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Review article

Andrographis paniculata (Burm.f.) Wall. ex Nees. - A potent herb with immense pharmacological potential

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ABSTRACT

Medicinal plants, as autochthonous sources of medications, have been utilized since primeval times. Andrographis paniculata (AP) is considered one of the most potent herbs around the world. Otherwise known as Kalmegh, the herb has traditionally been used as a cure for the common cold, diarrhoea, jaundice, and fever owing to numerous causes. Apart from this, the herbal parts can act as liver intoxicants as well as cardiovascular (CVD) vigor and are also considered to possess free radical scavenging activity. Habitation, season, and harvesting time of the crop have an effect on phytochemical composition that extensively differ from one part to another. In this review, we discussed the ethnobotany of this plant briefly. Apart from these various pharmacological activities with a specific focus on anticancer properties of the herb are also proposed.

***Keywords*:** *Andrographis paniculata* (AP), andrographolide, anticancer, ethnopharmacology, pharmacological potential.

INTRODUCTION

Andrographis paniculata (Burm.f.) Wall. ex Nees., belongs to the Acanthaceae Family- is an annual herbaceous plant and is generally cultivated in Southern Asia, China, and parts of Europe. The herb is usually renowned as kalmegh, bhumineeb, and chirayata. It is a valued traditional medicinal plant and has its usage in the Unani and Ayurvedic medicine systems (Chopra *et al*., 1956). The herb is commonly used to normalize body heat, dissipate toxic materials from the body; preclude common cold, infections of the upper respiratory tract as well as sinusitis and fever (Gabrielian *et al*., 2002). It also acts as an antidote against snakes and insects' venom (Samy *et al*., 2008). Phytochemical screening of various organic solvent extracts of AP plant parts, such as leaves and bark of the stem exhibited the availability of glycosides, phytosterol, saponins, tannins, flavonoids, and terpenoids. Terpenoids are considered to be the crucial constituents in the leaves as well as stem barks (Pandey *et al*., 2019). The occurrence of vital phytochemicals in AP makes the plant valuable for treating altered ailments and has the potential of providing useful drugs for human use (Pandey *et al*., 2019). This paper will provide a deep insight into the ethnobotany, a few agronomic techniques, a depiction of the potent chemical components, and the pharmacokinetics of Kalmegh. Furthermore, chemical possessions, biological roles, and their probable modes of action are also to be considered.

The plant has been reported to unveil several means of biological activities *in vivo* as well as *in vitro* such as, anticancer (Iruretagoyena *et al*., 2005; Li *et al*., 2007), anti-inflammatory (Wen *et al*., 2010), immunomodulating/ immunostimulatory (Calabrese *et al*., 2000), antihyperglycemic, antihyperlipidemic, hepatoprotective, cardiovascular, antiviral (Wiart *et al*., 2005), antimalarial and filaricidal, antidiarrheal, antibacterial (Singha *et al*., 2003; Mishra *et al*., 2009). The distinctive secondary metabolites encountered in this herb have substantially enriched its eminence in the arena of medicinal herbs. The quantitative determination of pharmacognostic parameters will facilitate in setting standards for crude drugs (Sharma *et al*., 2012).

Vernacular names

The herb is famous in diverse local languages in several parts of the world. In Hindi, the herb is famous as Kalmegh, Kiryat, and Mahatit. The herb is well known as Kiriyata and Olikiriyat in Gujrati. In Malyalam, it is called as Nilaveppu, Kiriyatta. Whereas NelaVemu and Nilavempui are the common names in Telugu and Tamil respectively. Bhuinimb, Kirata, and Mahateet are the local names in Sanskrit. English: Creat, Green Chirayta, King of bitters (Verma *et al*., 2019).

Origin

Kalmegh is an inhabitant of Taiwan, India, and Chinese province. The herb is usually located in Asia's tropical and warm temperate evergreen forests. The mainland of Southeast Asia (the Caribbean islands, Indonesia, Malaysia) and Sri Lanka, are the other continents, where the herb can be easily found. This plant is also found in altered geographical conditions of America, the West Indies, and Christmas Island (Verma *et al*., 2019).

Geographical distribution

The species is native to tropical South-East Asia and have its presence throughout warmer parts of India. The species is scattered in tropical Asian realms, frequently in remote areas. The herb can be sited in an array of habitations, for instance, plains, hillsides, coastlines, and disturbed and cultivated areas(roadsides and farms) (Niranjan *et al*., 2010; Mishra *et al*., 2007). Native populations of AP extend throughout south India and Sri Lanka which conceivably exemplify the epicenter of origin and diversity among species. The herb is an introduced species in northern parts of India, Java, Malaysia, Indonesia, the West Indies, and elsewhere in the Americas. The species also occurs in the Philippines, Hong Kong, Thailand, Brunei, Singapore, and other parts of Asia, where it may or may not be native (Perumal Samy *et al*., 2017). Contrasting other species of the genus, *A. paniculata* is of prevalent occurrence in most places in India, together with the plains and hilly areas up to an elevation of 500 m (1,600 ft). In India, the principal source of plants is collected from its wild habitat. According to the IUCN database, the plant is regarded as Low Risk or of Least Concern (Gowthami *et al*., 2021).

Climate and soil

The herb is a hardy species in tropical and subtropical regions and therefore thrives well in almost all types of soil. However, soils relatively superior in heavy metals such as Al, Cu, and Zn are preferred for the crop. Medium productive sandy loam to clay-loam soils with a pH range of 6.5 -8.5 is ideal for the cultivation of the crop. Though it is cultivated in open fields still can withstand partial shade of trees (Verma *et al*., 2019).

Propagation and nursery aspect

Both seed and vegetative methods can be utilized for raising the herb. However, propagation through seed is easy and economical when the cultivation is of commercial importance. Towards the beginning of September seeds of the herb have been sown in nursery beds, after soaking in water. About 650-750 g of seeds is the requisite for raising a nursery for one hectare of land (Verma *et al*., 2019). Seeds are usually sown in nursery beds prepared by taking a mixture of normal soil, sand, and organic matter in a ratio of 1:1:1 and at a spacing of 5 cm in rows. It takes around 8-10 days for germination to commence. The direct sown crop is scattered thinly with a seed proportion of 1.5 kg per hectare (Verma *et al*., 2019).

Agronomic techniques

Plants of Kalmegh traditionally grow through seed culture. The recommended time for sowing is usually from May to July. However, seed latency is a foremost limitation in the commercial farming of AP. Application of phytohormones and treatment with hot water has been recommended, to prevail over this problem (Kumar *et al*., 2011). The technique is not adequate as per the commercial measures concerned due to inconsistency amongst the scions derived from seeds and the deferred root system of seedlings (Martin *et al*., 2004). Numerous non-traditional proliferation approaches, for instance, micropropagation techniques are therefore the alternatives to generate huge amounts of saplings in a small duration, as well as to improve phytochemical contents in AP (Vijaylaxmi and Murthy, 2012). The yield of the herb is influenced by planting and time of harvesting (Nemade *et al*., 2003).

Land preparation and fertilizer application

The land should be organized well by frequent ploughing to make the soil pounded. Organic manure at a rate of 20 t ha^{-1} is applied as a basal application. An NPK fertilizer at a proportion of 75:75:50 kg ha^{-1} should be given in two split doses. The first dose is at the planting stage, followed by the second; that is 40 days after plantation. The employ of Azospirillium (5 kg) along with Phosphobacteria (5 kg) ha^{-1} will give satisfactory results.

Transplanting and optimum spacing

The nursery-raised seedlings are transplanted in the main field, upon reaching 10-25cm with an optimum distance of 30 x30 cm from plant to plant and row to row (Ram *et al.,* 2008; Kanjilal *et al.,* 2002).

Irrigation and weeding

Light irrigation as per prerequisite can be provided at regular intervals to aid the early establishment of seedlings, once the transplantation has been done. There is no need for additional irrigation as the crop is transplanted in the season of monsoon. Whenever there is a requirement for water, the field can be irrigated at intervals of 20-25 days (Verma *et al*., 2019).

Kalmegh is a short-duration crop and is cultivated in the *Kharif* season. Thus, weed invasion is an immense issue that deteriorates the quality and quantity of herbs. Since it is an herbaceous plant, the field should be free from weeds. Two to three weddings, usually one after 20 days and another after 60 days of plantation are essential during the crop season. The appliance of the herbicide pendimethalin at the rate of 1 kg ha^{-1} as pre-emergence followed by quizalofop ethyl at the rate of 50 g per hectare as post-emergence was found to be an efficient weed control agent. This should be followed by one mechanical weeding for accomplishing high biomass yield (Meena *et al.,* 2017; Semwal *et al,* 2016). There were no major insect pests or disease infestation has been recorded in Kalmegh. Brown Scale, *Parasaisettia nigra*, was found to causing considerable damage, with the affected plants showing stunted growth and drying. Semilooper, *Panilla albopenstata* dented the plants by feeding on young leaves, flower buds, and tender pods (Rani and Sridhar, 2005). Application of Azadirachtin (1- 2%) leaf extract upon the herb was found to be helpful (Suganthy and Sakthivel, 2013).

Crop maturity and harvesting

Generally, the crop matures after 120 days of sowing in October and November. Most pods mature fully in January and February (Verma *et al*., 2019). The planting and harvesting period influences the yield and quality of the crop. The active principle of the plant varies with time intervals and diverse environmental conditions (Kumar *et al*., 2002). It is harvested when most plants are in bloom. Upon maturity, the plants should be uprooted leaving few healthy plants in the field for seed production. The fruits once mature, should be picked up and dried in the sun for seed collection (Wankhade *et al*., 2005).

Postharvest management and storage

After uprooting the plant, it should be sun-dried for two days followed by shade drying. Once dried, the materials were packed in gunny bags and stored in dark, airy, and moisture-free places. The crops harvested after three months of cultivation showed the utmost sum of andrographolide, a foremost bioactive compound of AP followed by that just beforehand the flowering period (Tipakorn, 2002).

Yield and cost of cultivation

Most of the States of India have been reported for the commercial cultivation of Kalmegh, with an average yield (as a whole plant) of 2.5 t ha^{-1}. The approximate cost of the crop for one hectare comes to around INR 25,000/-.

Trade

Under the trade appellation Kalmegh, on normal 2,200-5,500 t of the herbage is traded in India.

Phytochemistry (chemical constituents)

Andrographolide (AD) is one of the foremost compound among the active principles that were extracted from the leaves and roots of Kalmegh. The compound is highest in the leaves; where as seeds have the lowest possible content (Sharma *et al*., 1992). Diterpenoids such as deoxy and rographolide-19-ß-D-glucoside and neo-and rographolide are two of the bitter components that have been isolated from the leaves (Weiming and

Xiaotion, 1982). The roots were found to be rich in apigenin-7, 4'-di-O-methyl ether, andrographolide, and a novel natural flavone, 5-hydroxy-7, 8, 2', 3'-tetramethoxy flavone ($C_{19}H_{18}O_7$). Andrographlide D (14-deoxy-11, 12-didehydroandrographolide), homoandrographolide, and rographan, andrographon, andrographosterin and stigmasterol are the other components reported in the herb (Siripong *et al*., 1992). Flavonoids, comprising 5, 7, 2', 3'- tetramethoxyflavanone, along with other flavonoids, andrographolide diterpenoids, and polyphenols, were acquired from the entire plant (Koteswara Rao *et al*., 2004). The above said phytoconstituents are considered to be accountable for various pharmacological activities of the herb.

Pharmacological potential (medicinal properties)

According to Ayurveda the herb "Kalmegh" is astringent, pungent, cooling, laxative, wound healing (vulnerary), antipyretic, antiperiodic, anti-inflammatory, expectorant (Mucoactive agents), depurative, sleep-inducing (soporific), anthelmintic, digestive, and stomachic. The herb is a valuable agent against hyperdispsia, burning sensation, ulcers, fevers, skin diseases, leprosy, colic, flatulence, diarrhoea, dysentery, and haemorrhoids, etc. (Mall and Tripathi, 2016). Kalmegh is also renowned as an apparent Homoeopathic drug. Fresh leaves of the herb are used to prepare "Alui", a well-known household medicine in Bengal (India), and are given to children suffering from stomach complaints. It has also been proven to be a hepatoprotective drug. Blocking of the voltage-operated calcium channels as well as inhibition of Ca (+2) influx by the dried herbal extract induces relaxation of the uterus (Hancke *et al*., 1995). Initial studies in animals ascertained that AP might help cure cardiac disease (Zhao, and Fang, 1991; Zhang, and Tan, 1997). It appears to stimulate gallbladder contraction, as well as prevent blood clots (Varies, 1993). The herb also showed certain other phytochemical properties as described below:

1. Anticancer effect

Andrographolide, the major bioactive compound of Kalmegh showed prominent effects on malignant cells due to hindrances in the proliferation of cells, arrest of the cell cycle, or cell discrepancy. Through induction of programmed cell death of malignant cells, these compounds also enhance the immune system. The anticancer ability of AD in colon cancer cells (HT-29) was evaluated by Khan *et al*. (2018) and a reduction in cell viability was observed. In a time and dose dependant manner, they observed reduced viability of the cells. The cell cycle in the G2 and M phases was surprisingly interrupted by AD at lower doses. While the cell cycle at the G0/G1 phase was arrested at a higher amount. Through ameliorating intracellular free radical levels and disruption of mitochondrial membrane potential, these phytoconstituents caused apoptosis of the cancer cells. AD along with another flavonoid isolated from Kalmegh leaves remarkably reticent U251 (CNS) and M14 (melanoma) human carcinoma cells and thereby proven the anticancer effect of the herb (Agarwal, 2015).

Diterpenoids along with andrographolide and isoandrographolide isolated from Kalmegh have also been explored by Chen and Colleagues (2009) taking human leukaemia (HL-60) cells. These two compounds were more effective compared to others. A different flavonoid, 7, 8- dimethoxy-2á - hydroxy-5-O-β-d-glucopyranosyloxyflavone, with a dose value (IC50) of 3.50 μM extracted from the aerial portions of the herb, revealed persuasive antiproliferative activity against leukaemia cells. An improvement in G0/G1 phase cells and a significant decline in the cell amount at the Synthesis Phase and G2 and M stages were observed against HL-60 cells with the use of andrographolide at a dose of 12 μg/mL.

The effective cytotoxic activity of the herbal extracts has been studied by Geethangili *et al*. (2008) against several human cancer cells such as lymphocytic (Jurkat), prostate (PC-3), hepatoma (HepG2), and colon cancer cells. Noticeably inhibition in the proliferation of HT-29 colon cancer cells was observed with the dichloromethane fraction of the extract. Inhibition of human colorectal carcinoma (CRC) Lovo cell growth by andrographolide was also reported by Shi *et al*. (2011). Induction in the expression of inhibitory proteins (p16, p21, and p53) of the cell cycle and arrest of the G1-S phases is considered to be responsible for this. The activity of cyclin D1 along with Cdk4 and cyclin A1/Cdk2 necessary for the

transition from the G1 to S phase has been overwhelmed by these proteins. Growth suppression, CRC cell invasion, and stimulation of apoptosis have been shown by andrographolide recently.

AD repressed the proliferation of lung cancer cells through a reduction in the level as well as a transformation of the growth factor (Luo *et al*., 2014). Inhibition of melanoma tumour enlargement and metastasis through apoptosis and arrest of the melanoma cell cycle at the G-1 phase has also been shown by AD. In other ways, inhibition of TLR4/ NF-κB signaling pathways, followed by inhibition of mRNA and Bcl-6 and CXCR4 protein expression, is thought to be the underlying mechanism for this activity (Zhang *et al*., 2014).

2. Antihyperglycemic effect

Effective antihyperglycemic action has been shown by AP extracts and andrographolide with a reduction of blood glucose levels through α-glucosidase and α-amylase inhibition. Besides monitoring blood glucose levels, the compounds in a dose-dependent manner can also effectually prevent the onset of insulitis. Consequently, in NOD mice, diabetes development was suppressed and delayed. By improving glucose exploitation and oxidation, AP has been found to lower blood glucose levels in type 2 diabetic rats. Liver restoration of insulin signaling molecules and lower serum lipid levels are also responsible for this activity (Augustine *et al*., 2014). Additionally, andrographolide controls the T Helper cells balance, which enables it to restrict T-cell insinuation into pancreatic cells and maybe avert β-cell death. Thereby, preventing the expansion of type 1 diabetes. For the antihyperglycemic action, stimulation of the sugar uptake and peripheral tissues oxidation with an increase in insulin level is also taken into consideration.

In both insulin-deficient diabetics and healthy rats, AP had a diminishing effect on blood sugar levels. In diabetic rats, andrographolide significantly enhanced β-cell functioning, GLUT4 translocation, and blood glucose levels. There was an ominous decrease in the fasting blood glucose in humans upon administration of Kalmegh extracts. Another bioactive constituent, 14-deoxy-11, 12-didehydroandrographolide, has also demonstrated antihyperglycemic action. Therefore, researchers' main focus will be on finding more antihyperglycemic chemicals in Kalmegh along with other medicinal plants to develop a better diabetic treatment alternative.

3. Anti-hyperlipidemic effect

The major cause of atherosclerosis, which results in heart attack and stroke, is hyperlipidemia. Several medicinal plants have been utilized, either alone or in combination, for the treatment of *Diabetes mellitus* (DM) since time immemorial. However, in order to confirm the safety and efficacy of these medicinal plants further scientific and clinical studies are needed (Fallah *et al*., 2023). Chen *et al*. (2020) found that and rographolide and neo and rographolide from AD have antihyperlipidemic properties. Reports by Yang and Song (2014), mentioned the fruitful efficacy of and rographolide and neo and rographolide on hyperlipidemic mouse models. In a dose-dependent way, these two substances diminished the cholesterol level, which can be confirmed with a decrease in the enzymes aspartate transaminase and alanine transaminase. Due to the various regulation of iNOS as well as eNOS expression in the aorta of hyperlipidemic rats, lipid and lipoprotein-reducing actions of the substances are liable to increase. It is necessary to concentrate future research on the concurrent signaling pathway and molecular mechanism.

4. Cardiovascular effect

According to Wong *et al*. (2020), *A. paniculata* and andrographolide have bioactivities that decrease the inflammatory response, oxidative stress, apoptosis, cardiac fibrosis, and endothelial dysfunction. By inhibiting the inductive phase of the inflammatory response, mediated by several signaling molecules such as NF-κB, PI3K/ Akt, MAPK, and STAT3, the phytoconstituents reduce the signs of myocardial damage. A possible mechanism for the specific action might be the effect on oxidative stress caused by activating the nuclear transcription factor, Nrf-2, and decreasing the enzymes that produce free radicals. Additionally, the phytoconstituents have blocked profibrotic growth factors, which has reduced heart fibrosis and enhanced endothelial and fibrinolytic function (Wong *et al*., 2020).

5. Antimalarial and filaricidal activity

Antimalarial action of Kalmegh root xanthones against *Plasmodium falciparum* and *Plasmodium berghei* on animal models (Swiss Albino mice) have been investigated by Dua *et al*. (2004). After treating the mice with a dose level of 30 mg/kg, a generous reduction (62%) in parasitaemia was observed. 1, 2-dihydroxy-6, 8-dimethoxyxanthone, with a dose level of IC50 > 32 µg/mL was noncytotoxic against mammalian cells in an *in-vitro* cytotoxicity assay. Filaria, a condition in which there is an obstruction of the lymph channels that will lead to Elephantiasis can be efficiently inhibited with the herbal extracts. Four of the potent xanthones such as 4, 8-dihydroxy-2, 7-dimethoxy xanthone, 1, 8-dihydroxy-3, 7-dimethoxy xanthone, 3, 7, 8- trimethoxy-1-hydroxyxanthone, and 1, 2- dihydroxy-6, 8-dimethoxyxanthone were reported in the roots of the herb. However, substantial antiplasmodial activity against adult worms of *Brugia malayi* was reported by 1, 2-dihydroxy-6, and 8-dimethoxy-xanthone. Though, clues for the pharmacological targets along with the mechanism of action of this compound on *P. falciparum* are still unclear. However, the regulation of a transcription factor is considered to be responsible for this phenomenon. Erythrocytes, upon infected with *P. falciparum*, there was an induction of NF-κB regulated inflammatory pathways in human cerebral endothelium has been observed. For effective regulation of malaria, there is a need for the re-evaluation of probable anti-malarial activity of andrographolide against the blood stage of the plasmodial life cycle.

6. Antidiarrheal effects

Diarrhea is the second leading cause of death, especially among children under five years old in developing countries. *S. flexneri, S. aureus, E. coli, S. typhi*, and *C. albicans* are the foremost causative agents of diarrhea in humans (Ashrafuzzaman *et al*., 2016). Several plant extract comprises pharmacologically active substances with antidiarrheal properties. AP extracts exhibited substantial effects against *E. coli* bacterial infections. A substantial effect against *E. coli* bacterial infections has been reported. In a case study, patients with acute bacterial diarrhoea were given a 500 mg dose of andrographolide for six days on a daily basis. It was found that the patients responded favourably to the treatment with an overall effectiveness of 91.3%. Due to its antibacterial activity, the herbal extract was effective in bacterial dysentery as well as diarrhea (Perumal Samy *et al*., 2007).

7. Antibacterial activity

Deaths due to microbial infections were investigated and reported around 9.2 million in 2013, accounting for 17% of overall mortality (Gupta *et al*., 2019; WHO, 2013). The microbes acquire resistance to numerous antimicrobial drugs and this is considered to be the sole cause for the ineffectiveness of the drugs. To fight the health problems related to bacterial infections, there should be an imperious need for research on other therapeutic agents as an alternative source to existing ones (Brown, 2015; Ncube *et al*., 2007). Due to their immense therapeutic activities, plant based antibacterial components are thus a suitable remedy for infectious diseases (Shakeri *et al*., 2018).

The isolated phytoconstituents andrographolide from AP showed significant antimicrobial activity. Various organic solvent extracts of AP flowers exhibited antimicrobial action toward *S. agalactiae, S. aureus,* and *E. coli* were reported by Sivananthan (2013). An inhibitory effect and thus antimicrobial activity of Kalmegh extract against the urinary tract pathogens for example *E. coli, P. aeruginosa, K. pneumonia, and S. aureus* has been reported *by* Murugesan *et al*. (2018). However, maximum inhibitory action was reported against *E. coli*.

Discussion

There is mention of *Andrographis paniculata* (AP) possessing potent phytoconstituents in altered traditional systems of medicines. Due to various pharmacological activities, the herb has been used as a remedy for hepatic issues, malaria, common colds, cardiovascular diseases, leprosy, stomach problems, etc. The plant has been used to treat hepatic issues, malaria, common colds, cardiovascular diseases, leprosy, stomach problems, etc. due to various pharmacological activities. The prime component accountable for most of the pharmacological actions of the drug is Andrographolide- a diterpene lactone.

Along with other diterpenoids isolated from AP, andrographolide, and isoandrographolide have been reported for their antiproliferative activities by many authors against mammalian cell lines. Potent antiproliferative activity by parts of AP against leukaemia cell lines has been reported. An improvement in cells (G0/G1 phase) and a considerable decline in cell amount against HL-60 cells were mentioned with the use of andrographolide. The dichloromethane fraction of the extract reported a considerable inhibition in the proliferation of HT-29 colon cancer cells. A direct inhibitory effect on cancer cells was employed by andrographolide and its analogues due to induction in the expression of cell cycle inhibitory proteins along with depressing cyclin-dependent kinase (Cdk). As a result, the progression of the cell cycle has been blocked at various stages. In multiple myeloma cells, an inhibition in cell proliferation, and apoptosis, as well as an enhancement in the caspase cascade activation was perceived. AD at a dose of 5μg/g showed *in vivo* anticancer activity in B16 melanoma mouse cells through inhibition of the TLR4/NF-κB signaling pathway. Reports regarding *in vitro* anticancer activity by AD have also been available in human myeloma cell (OPM1).

AP decreases the blood sugar level by escalating its utilization and oxidation. Apart from this insulin signaling molecules restoration in the liver and fall in the serum lipid levels indicate the positive effects of the compound (Augustine *et al*., 2014). The blood sugar level lowering ability due to inhibition of α-glycosidase and α-amylase is another way that proves the antihyperglycemic effects of the AP extracts and andrographolide. Whereas, Yu *et al*., 2003; reported that the increase of plasma glucose in rats has significantly been attenuated by Andrographolide. Andrographolide and neoandrographolide- two of the potent phytoconstituents have shown hyperlipidemic activity in animal models (mice) (Yang *et al*., 2014). There was a significant reduction of aspartate and alanine transaminase levels in the plasma compared with simvastatin. The lipid and lipoprotein-reducing effects of the compounds in hyperlipidemic rats are possible due to various regulation steps of iNOS and eNOS expression. The active constituents along with the aqueous extracts of AP exhibited platelet anti-aggregation activity against *in vitro* mouse models (Thisoda *et al*., 2006). AP has the potential to enhance NO, cyclic GMP, and SOD activity (Wang *et al*., 2020).

Andrographolide showed a potential beneficial effect against H9C2 cardiomyocytes. Reports were there on the effective doses for *Kalmegh* herbal extract with treatment duration of 7-31 days against myocardial injury. Thus, Kalmegh can be considered as an alternative source for the treatment of cardiovascular diseases. However, the effectiveness of these phytoconstituents from the herb against myocardial injury in humans along with clinical trials is yet to be investigated. The mice upon treatment with the herbal extract of andrographis showed a substantial reduction in parasitaemia. The ring stage of the parasite, during the erythrocytic life cycle, is considered to be the key point where the activity of andrographolide was found to be prominent. Protein and nucleic acid synthesis are the key points in the parasite life cycle, upon which the mechanism of action of this compound relies. The efficacy of AP solvent extracts as an antibacterial agent in urinary tract pathogens have been reported by Murugesan *et al*. (2018). The herb showed an overall effectiveness of 91.3% among patients with acute bacterial diarrhoea after treatment for six days.

Researchers, around the globe attain various leads by structurally modifying and brographolide due to the variety of biological activities shown by the herb. The pharmacological activities of numerous and rographolide derivatives have been evaluated that have emerged in recent times. A pilot-scale clinical studies has confirmed the safety of the herb. The herb's efficacy against other diseases needs to be evaluated further in all age group populations. However, studies that have comprehensively summarized or analyzed *A. paniculata* and its derivatives have been minimal. As a prerequisite, there is a need for deep research to assess the potential of the plant in clinical practices. In conclusion, this review highlights the chief eminence of Kalmegh as an established medicinal herb. The various pharmacological activities of the herb make it valuable in ameliorating altered diseases and lay out crucial directions for future research.

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Review article

Azadirachta indica–Neem-A natural gift for prevention and treatment of chronic diseases- A review

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ABSTRACT

Neem plant (Azadirachta indica L.) was known since ancient times, one of the most significant medicinal herbs in the world. Every part of the plant viz., leaves, bark, stem, wood, seed are utilized for treatment and prevention of a large number of human diseases. It is due to containing more than 300 phytochemicals of which azadirachtin is the most prevalent. For avoidance and treatment of different human chronic illnesses neem is used in Indian subcontinent and worldwide in the ayurveda, Siddha, and Unani medical systems and recently specially for treatment of Covid related system of diseases. This article summarized the importance, health benefits, biological role, major phytochemicals present in different plant parts and their molecular action in curing diseases and cultivation tips of neem tree.

Keywords: *Azadirachta indica,* chronic diseases, COVID-19, cultivation tips, ethnobotanical uses, phyto-chemicals

INTRODUCTION

Azadirachta indica (*Neem*) is seen as a treasured gift from nature. Since the prehistoric age, India and its adjacent countries have benefited from its enormous medicinal and ethnomedical values for humanity (Tiwari *et al*., 2014). It is also known in various names like holy tree, Indian lilac tree, Nim and Nimba. According to the Persian it is derived from three Latin words these are- Azad means "free"; dirakht means "tree"; i-Hind means (Gupta *et al*., 2017). Neem is one of the most significant multi-reason trees to the Indian subcontinent, but cultivated throughout the world mainly Southeast Asia, Australia, East and Sub-Saharan Africa, Fiji, Mauritius and many countries of central and South America (Kumar *et al*., 2019). Humans were using neem from immemorial time as a remedy for smallpox and other diseases. In addition, it has long been believed to have antibacterial properties and the ability to ward off evil spirits.

Neem, belongs to same family of the mahogany i.e. Meliaceae, is quick growing plant propagated naturally by seeds. The maximum productive life span of neem tree ranges from 150-200 years. Every part of the tree contain some phytochemicals with confirmed antiviral, antiseptic, anti- ulcer, anti-inflammatory, antifungal and antipyretic uses. Azadirachtin is one of the neem plant's most prevalent chemicals, which having insecticidal properties as well as used as a bio-pesticide (Chaudhary *et al*., 2017; Kilani-Morakchi *et al.,* 2021). For maintaining dental hygiene by chewing sticks, millions of rural as well as urban people use neem twigs (Gupta *et al*., 2017).

In view of modern medicine and chronic disease recently, the *neem* tree has acquired popularity to the researchers. Because it contains new antimicrobials and it is also applicable to in the disciplines of endocrinology, dermatology, dentistry, and oncology etc. (Wylie and Merrell, 2022). A significant challenge has been presented to humanity by the recent epidemic brought on by the new coronavirus SARS-CoV-2 (COVID-19). Different variants (Alpha, Beta, Gamma, Delta & Omicron) are available in case of COVID-19. Now the distinctly observed variant is Omicron corona virus group. Omicron has a significantly greater rate of symptomatic carriage compared to others

variant of corona virus had given high preceding rates of SARS-COV-2 infection, and this high prevalence of symptomatic infection is probably a major role in the global and rapid spreading.

It is well known to people, SARS-CoV-2 cannot be effectively prevented by any potentially active medication or vaccination that was previously available. Despite the fact that a number of medications, including chloroquine and hydroxy chloroquine, are advised against COVID-19, few of these having unfavourable outcomes which was reported by many researcher (Baildya *et al*., 2020). Now a days, different kind of vaccines (Covaxin, Covishield, Covovax, Comirnaty etc.) are available which can be effective against COVID-19 but these also have some side effects (Beniwal *et al.,* 2021). In the beginning of pandemic different potential drugs from natural sources, like the restorative *neem* plant was find out effectively against COVID-19 by some research workers (Eze *et al.,* 2022; Sarkar *et al*., 2022). In this article, cultivation aspects and important biological role of *Azadirachta indica* has been discussed with special emphasized to COVID-19 and others chronic disease.

Chemical composition of *neem*

More than 300 distinct chemicals have been taken from various *neem* components. The researcher claims that extracted chemicals can be generally divided into two kinds. Both isoprenoids and non-isoprenoids are present (Latif *et al.,* 2020). Diterpenoids and triterpenoids, which contain limonoids, azadirone, protomeliacins, and their derivatives, are the chemicals that make up isoprenoids. Carbohydrates (polysaccharides), proteins (amino acids), sulfate-containing substances, polyphenolics including glycosides and related flavonoids, dihydrochalcone, tannins, and coumarins are among the substances included in non-isoprenoids Diterpenoids and triterpenoids, which contain limonoids, azadirone, protomeliacins, and their derivatives, are the chemicals that make up isoprenoids. Carbohydrates (polysaccharides), proteins (amino acids), sulfate-containing substances, polyphenolics including glycosides and related flavonoids, dihydrochalcone, tannins, and coumarins etc. are among the substances included in non-isoprenoids *etc*. (Biswas *et al*., 2002). Beside this, some other compounds which were extracted from *neem viz.,* margalonone, margalone, and isomargalonone (Dixit, 2015).

The compounds have been extracted from bark of *neem* are catechin, gallic acid, epicatichin, polysaccharides and NB-2 peptidoglucan which having anti-tumor, anti-inflammatory, antioxidant and antibacterial activity (Pankaj and Lokeshwar, 2011; Ghimeray *et al*., 2009; Susmitha *et al*., 2013).

β-sitosterol and Quercetin (poly-phenolic flavonoids) were extracted from *neem* leaves and were famous for their anti-bacterial as well as anti-fungal properties. (Alzohairy, 2016). Beside this other essential compounds found in the *neem* leaves are nimbanene, nimbandiol, nimbin, nimbolide, amino acid and nimbiol (Gupta *et al*., 2017). Phytochemical analysis of methanolic leaf extracts of *Azadirachta indica* has shown the presence of biological compounds like, Alkaloids, Flavonoids, Saponins, etc which are then compared to aqueous leaf extracts of the plant (Dash and Dixit, 2017). Irodin A, isolated from leaves of neem, effective against contributing anxieties of HIV/AIDS and malaria (Anyaehie, 2009) and Azadivactrin, possess capability to perform against disease causing viruses, parasites and fungi (Raj, 2015).

Compounds present in *neem* seeds are liminoids or triterpenoids together with azadirachtin that would be one of the most significant (Tindo and Amusant, 2012). Azadirachtin, a tetranor triterpenoid, is an important biologically active constituent of *neem* seed kernel, generally used for pest control (Gupta and Tripathi, 1998). Specifically, the separation of three products nimbidin, nimbidic acid and nimbolide was from seed kernel oil (Bansal *et al*., 2010; Latif *et al*., 2020).

Ethnobotanical utilizations of *neem*

Neem leaves are used as a diuretic, for headaches, heartburn, and boosting appetite, as well as for oral health, gastrointestinal problems, insect repellent, malaria, and a number of skin conditions. *Neem* leaves, on the other hand, are used to cure diabetes (Sujarwo *et al*., 2016). Traditionally in India, due to its medicinal qualities, we can get relief by lying on the leaves from chicken pox (Maithani *et al*., 2011). *Neem* stick is used to clean the teeth for preventing the tooth decay, bleeding gums and

foul smell. Traditionally *neem* oil is used for lightening purpose as fuel in village area. Additionally, it works well as an antiseptic for the treatment of intestinal worm infections, eczema, and furuncles (Eid *et al*., 2017).

Nigerians use this as a traditional malaria cure. Seed aqueous extracts are used to treat head lice. *Neem* is also used to combat insects and pests; its main form of action is the interruption of metamorphosis, and its bitter taste deters insects from feeding on its host plants. (Senthil Kumar *et al*., 2018).

Uses of *Neem*

Neem tree remedies for a variety of human problems are frequently utilised on all sections of the tree. In the southern states of India, Sri Lanka, Malaysia, and Singapore, the Siddha system of medicine is widely practised. The Siddha medical system first makes reference to the medicinal herb *neem.*

Neem is effective against rheumatism, malaria, intestinal worms, jaundice, tuberculosis, skin and as well as arthritis. *Neem* oil is an effective antiseptic for the treatment of intestinal worm infections, eczema, and furuncles on the skin (Eid *et al*., 2017).Calcium mining, which is a unique property, capability to neutralize acidic soils is predicted in *neem* (Latif *et al*., 2020). Beside this it is having astringent effect, bitter property is useful for loss of appetite, cough, tiredness, helpful for healing wounds and excessive thirst infestation to combat vomiting (Dixit, 2015). The neem oil is also used for fighting pimples, acne as well as refining skin elasticity (Manisha and Sachin, 2014).

A significant anti-diabetic potential has been demonstrated by the extract of Neem leaves and possibly will be reduce the 30%-50% use of insulin (Latif *et al*., 2020). It is also considered as the possible substance for the treatment of cancer patients and AIDS. In south-Asia, a huge amount of *neem* oil is available and it is non-edible and it is used for cosmetics such as nail polish, purified *neem* oil is used (Latif *et al*., 2020; Balami and Aliyu, 2014). *Neem* seed cake, which contains the needed macronutrients for plant growth, is used as a bio-fertilizer after *neem* oil has been extracted (Ramachandran *et al*., 2007). Neem oil posses various phyto-chemicals, used for human health and insect-pest control (Campos *et al.,* 2016).

Jagannathan and Vasuki (2015) reviewed the properties of neem as insecticide, antifeedant, hormonal, antifungal, antiviral and nematicidal properties in agricultural applications through use of *neem* leaves, leaf extracts, oil, seed cakes, seed and fruit extracts. The *neem* and its products are used in seed treatment, soil application, foliar spraying, increasing nutrient efficiency by which the rice yield was found to enhance and its sustainability was seen in rice cropping system.

Biological properties of *neem* and molecular action of its phyto-chemicals

Diabetes prevention

Chronic hyperglycemia, which causes endothelial dysfunction and atherosclerosis, is a serious global health issue associated with diabetes (Zhang *et al*., 2015). Due to hyperglycemia and hyperlipidemia, there was an increase in the formation of free radicals or oxidative stress. (Barbosa *et al*., 2013). *Neem* is tested to see if it can lower intestinal glycosidase activity (Joshi *et al.,* 2011). *Neem* leaf extract may be helpful in the treatment of type-2 diabetes by increasing the production of the protein GLUT4 and insulin signalling molecules, according to an experimental investigation (Satyanarayan *et al.,* 2015).

Anti-Inflammatory activity

A defense mechanism of the body includes inflammation. Our immune system is triggered when our body is exposed to harmful substances such as viruses, bacteria, or poisonous chemicals or when it sustains an injury. Inflammatory cells and cytokines trigger our immune system's initial defenses. These cells may begin the healing process or work to trap germs and other harmful agents. The result can be shown in various ways like pain, swelling, bruising or redness. Mainly two types of inflammation: i) **Acute inflammation:** the reaction to an immediate physical injury, like cutting our finger. ii) **Chronic inflammation:** Even when there is no threat from the outside, our body keeps sending inflammatory cells. For instance, in rheumatoid arthritis, inflammatory chemicals and cells assault the tissues of the joints. **Acute inflammation may cause** pain, tenderness, swelling, heat, etc. Symptoms of **chronic inflammation** could be more difficult. It may also

include mouth sores, skin rashes, joint pain or stiffness, exhaustion, fever, and chest trouble.

Bhowmick *et al*. (2010) concluded that a compound found in *neem*, called nimbidin, might possess anti-inflammatory and anti-arthritic activity. Rheumatoid arthritis, which is distinguished by inflammation and pain in the muscles and joints as a result of auto-immune reactions, may also benefit from it. Schumacher *et al.* (2011) studied the ability of neem leaf extract to reduce inflammation through the nuclear factor-κB (NF- κB) signalling system, which is connected to apoptosis, inflammation, and cancer and concluded that *neem* extracts have strong effect on pro-inflammatory cell signaling and apoptotic cell death mechanisms. *Later* Alzohairy (2016) also reported that *Neem* act as anti-Inflammatory agent. It functions as an anti-inflammatory by regulating the activity of proinflammatory enzymes such the cyclooxygenase (COX) and lipoxygenase (LOX) enzymes.

Hepatoprotective effect

The body's many physiological processes depend on the liver, which is the most significant organ in this regard. A liver inflammation known as hepatitis is distinguished by the presence of inflammatory cells in the organ's tissue. Uorakkottil *et al*. (2016) stated that liver damage or dysfunction is a significant health issue that poses difficulties for the pharmaceutical business, medication regulatory organisations, and healthcare providers.

For a very long time, liver illness has been treated with herbal medications. Polysaccharides, proteins, flavanoids, lignans, and other phytoconstituents originating from plants maintain liver disorders and boost the immune system. There are several herbs that have been reported to have hepatoprotective and immunomodulatory properties (Uorakkottil *et al*., 2016). *Neem* has been found to have hepatoprotective effects, according to numerous researchers. Kale *et al*. (2003) stated that hepatoprotective activity of *neem* leaf extract on antitubercular drugs-induced hepatotoxicity and resulted in significantly preventing changes in the serum levels of protein, bilirubin, alanine aminotransferase, aspartate aminotransferase, and alkaline phosphatase as well as significantly preventing the histological changes as compared to the group receiving antitubercular drugs.

Baligar *et al.* (2014) established azadirachtin's involvement in rat liver damage caused by carbon tetrachloride (CCl4). According to hepatoprotective investigations, there was liver damage induced by CCl4 since the CCl4 treatment group showed a drop in total protein and albumin levels compared to the vehicle-treated control. According to the study's findings, pretreatment with azadirachtin at higher dose levels only modestly improves the condition of the rat liver. This research demonstrates that azadirachtin has a stronger hepatoprotective effect.

Antiviral activity

Badam *et al.* (1999) noted that *neem* leaf extract has demonstrated virucidal efficacy against the B-4 cox sackie virus. The herpes simplex virus type 1 (HSV-1) can lead to encephalitis, chronic skin and corneal ulcers, and other serious health issues. Tiwari *et al.* (2010) reported that *Neem* plant bark blocks HSV-1 entry into cells at concentrations ranging from 50 to 100 ng/ml and is a powerful entry inhibitor against HSV-1 infection into natural target cells. Additionally, polykaryocytes and HSV-1 glycoprotein-mediated cell-cell fusion were suppressed in cells treated with *neem* bark extract. Later, Yerima *et al.* (2012) also verified the prior findings that, at doses ranging from 50 to 100 g/mL, neem bark extract effectively inhibited HSV-1 entrance into cells.

Antiulcer activity

Bandyopadhyay *et al*. (2004) showed that *neem* bark extract has therapeutic potential for reducing gastric hypersecretion and ulcers in the gastroduodenum and gastroesophageal junction. The duodenal ulcers were nearly fully cured after 10 weeks of treatment with the bark extract at a dose of 30–60 mg twice day. After six weeks, one patient's esophageal and stomach ulcers were completely healed. They came to the conclusion that the neem bark extract decreased the hypersecretion of stomach acid in those with gastro-duodenal and gastro-esophageal ulcers.

Antioxidant activity

Chronic conditions like cancer, diabetes, and cardiovascular conditions (CVD) are major worldwide health issues that claim the lives of millions of people and leave them disabled.

Numerous plants, including *neem,* have been shown to be protective against the emergence of these chronic disorders. Scavenging these oxidants is considered to be a useful strategy to reduce organisms' oxidative stress levels (Zhang *et al*., 2015). Antioxidants are the substances, bioactive non-nutrient compounds that inhibit oxidation or inhibits reactions promoted by oxygen or peroxides. They lessen the production of free radicals and support the body's defense against cell damage. Toxic metals cause the production of free radicals. They also cause cardiovascular diseases (CVD), cancer, diabetes, and oxidative stress. They are the source of the oxidative degradation of DNA, protein, and other essential components (Hla *et al*., 2011; Zhang *et al*., 2015). In a study using extracts of the Siamese *neem* tree's leaves, fruits, blossoms, and stem bark Sithisarn *et al*. (2005) reported that a substantial antioxidant potential has been found in leaf, flower, and stem bark extracts. Later Ghimeray *et al*. (2009) also noted that *neem* growing in the foothills' high antioxidant capabilities were also discovered in its leaf and bark extracts.

Experimental results showed that nimbolide and azadirachtin have reductive potential and concentration-dependent antiradical scavenging action in the following order: Nimbolid, Azadirachtin, and Ascorbate (Priyadarsini *et al*., 2009).

Antibacterial activity

Neem extracts' antibacterial activity was tested against a small number of pathogens, and the results showed that it may be effective for preventing the growth of spoilage organisms and foodborne diseases (Mahfuzul Hoque *et al.,* 2007).

Yerima *et al*. (2012) investigated the antibacterial activity of leaf, fruit, seed and bark extracts of neem by using agar well diffusion method. With increase in concentrations of the extracts, the zone of inhibition also increased. The antibacterial activity only at higher concentrations is shown by the extracts of fruit and seed. Therefore, the results confirmed the usage of neem in sustaining oral hygiene traditionally. Later, Mariana *et al.* (2017) concluded that Neem's effective antibacterial action against the oral strains under investigation, which may be attributed to the substance's flavonoids and saponins content.

Antimalarial activity

Deshpande *et al.* (2014) demonstrated that *neem* leaf extract has some active components that may be the cause of its effectiveness against *Plasmodium falcifarum* and *P. vivax*. Another experiment using albino mice infected with Plasmodium berghei was conducted to assess the antimalarial activity of extracts. The results showed that the extracts of *neem* stem, bark, and leaf reduce parasitemia in infected mice by 51–80% and 56–87%, respectively. Other research revealed that Azadirachtin, which is present in *neem* extracts, and other liminoids are potent against malaria vectors (Alzohairy, 2016).

Antifungal activity

The growth of the seed-borne fungus Aspergillus and Rhizopus was clearly suppressed and controlled by the antifungal activity of leaf extracts of neem (Alzohairy, 2016).

Antimicrobial activity

Three fungi, including *C. lunata, H. pennisetti,* and *C. gloeosporioides* f. sp. mangiferae, were inhibited from germination of their spores by the aqueous extracts of *neem* cake (Anjali *et al.,* 2013). The study demonstrated that neem extract in methanol and ethanol inhibited the growth of *Aspergillus flavus, Alternaria solani* and *Cladosporium* (Shrivastava and Swarnkar, 2014).

Antisnake venom activity

Mukherjee *et al*. (2008) reported that a compound, phospholipase A (an enzyme), extracted from the leaf of *neem* was effective against snake bite and worked as anti-snake venom drug. This substance prevented the cobra and Russell's viper (RVV) venoms from acting. The amount will be depending upon the dependent.

Wound healing effect

Osunwoke Emeka *et al.* (2013) demonstrated that leaf extracts stimulate inflammatory response and neovascularization, which boost wound healing activity.

Antifertility activity

Neem oil has a novel application that is described: reversible inhibition of fertility after a single intrauterine administration (Upadhyay *et al.*,1990). *Neem* extract can be used to improve birth

control, according to a report by researchers at the Defense Institute of Physiology and Allied Sciences (DIPAS). *Neem* oil can prevent pregnancy without having any negative effects on the vagina, cervix, or uterus when administered intravaginally before to sexual activity, according to the study (Bansal *et al*., 2010), but not absorbed from the vagina (Singha *et al*., 1984). Its active ingredients were discovered to be absorbed into the bloodstream through the vaginal mucosa and to have antifertility effects in addition to their direct spermicidal actions.

Khillare and Shrivastav (2003) determined that the aqueous extract of old and tender *neem* leaves could directly immobilise and destroy 100% of human spermatozoa within 20 seconds. When compared to untreated sperm, no morphological abnormalities were observed in the sperm head, mid-piece, or tail. Therefore, complete sperm death may result from a metabolic process, such as energy utilization, being blocked. Longer storage times (up to 4 years) have no effect on the potentiality or efficacy of lyophilized aqueous extract of old and tender leaves.

Anti-dental caries

Considering the health-awareness criteria scientists are exploring and recognized *neem* as one of the beneficial plant for the development of anti-dental carries medicine from plant origin. Gupta *et al.* (2017) studied and emphasised the therapeutic benefits of *neem* from the perspective of oral health, including its treatment of gingivitis, anti-microbial, anti-plaque, anti-candidiasis, anti-periodontitis, tooth erosion, and oral cancer.

Anti-cancerous activity

Cancer is a complex illness that is a huge global health issue. Allopathic medicine is helpful on one hand, but it also had negative effects on healthy cells (Alzohairy, 2016). The multistage carcinogenic process is thought to involve free radicals. DNA mutations, which are essential for the start of the carcinogenic process, can be brought on independently by peroxylradicals and lipid peroxidation. By preventing DNA damage, antioxidant phytochemicals may control the start of the carcinogenesis process. *Neem* and its components were thought to be crucial in the control of numerous cell signaling pathways. It contains a variety of components that can turn on tumor suppressor genes and deactivate the activity of several genes implicated in the onset and spread of cancer. According to reports, *neem* is an effective tumor suppressor gene activator. Additionally, it induces apoptosis, blocks NF- κB signaling, and activates the cyclooxygenase pathway (Alzohairy, 2016).

Anti-HIV activity

Acquired Immune Deficiency Syndrome (AIDS) is brought on by Human Immunodeficiency Viruses (HIV). *Neem* has historically been used to treat HIV-related illnesses. (Wylie and Merrell, 2022), have been investigated for their potential to shield the HIV-vulnerable $CD4^+$ T cell population. According to certain research, *neem* leaf extract can increase $CD4^+$ T cell counts in HIV patients on a daily basis while being safe to consume. Overall, the findings confirmed that *neem* possesses immunomodulatory properties that may be useful for some treatments and to enhance the health of people with persistent infections (such as HIV).

Anti-Dengue activity

Kaempferol 3-O-rutinoside and epicatechin were shown to be able to suppress dengue virus by 77.7% and 66.2%, respectively, after 49 different *neem* tree bioflavonoids were virtually screened for binding to the virus (Dwivedi *et al*., 2021). Triterpenoids called nimbin, which were extracted from neem leaves, were found to be efficient against the dengue virus's envelope protein (Lavanya *et al*., 2015). Due to its unique phytochemicals that functioned to inhibit the functions of both the protease and envelope proteins of the dengue virus, as well as may be for other viruses, *neem* may be a novel source for antiviral medications (Shanmugam *et al*., 2020).

Anti-SARS-CoV-2 activity

A family of viruses known as corona viruses can seriously harm the acute respiratory system and cause symptoms like the common cold. It was found to be the root of a disease outbreak in 2019 that originated in China. The virus is referred to as SARS-CoV-2, or severe acute respiratory syndrome coronavirus. The condition it brings on is referred to as coronavirus disease 2019 (COVID-19). The

World Health Organisation (WHO) classified the COVID-19 outbreak as a pandemic in March 2020. People with COVID-19 have reported experiencing a wide range of symptoms, from minor discomfort to serious sickness. 2 to 14 days after virus contact, symptoms may start to show. New variations and vaccination status may affect the severity and number of symptoms. The risk is larger for older folks and those with underlying illnesses including diabetes, heart disease, or lung disease. Acute respiratory infection and multiple organ failure should be present in extreme situations.

Over the past two years, COVID-19 has claimed the lives of more than five million people and has continued to pose serious hazards to the public's health (WHO, 2021). SARS-CoV-2 antiviral treatment is still not available. A thorough examination of this outbreak's dynamics indicated that Papain-like protease (PLpro), a multifunctional polyprotein, promotes SARS-CoV-2 replication and shields it from the host immune response via antagonistic cytokines and interferons, and may be viewed as a possible therapeutic target (Balkrishna *et al*., 2021). Elfiky (2020) suggested that Ribavirin, Remdesivir, Sofosbuvir, Galidesivir, and Tenofovir are the effective medicines against SARS-CoV-2 due to their ability to bind to its RdRp.

Numerous studies have successfully searched plant-based chemical compounds for SARS-CoV-2 viral inhibitors (Thota *et al*., 2020; Adithya *et al*., 2021). When compared to other medicinal plants, Neem's phytochemicals that combat SARS-CoV-2 performed a significant contribution (Wylie and Merrell, 2022). The action of phytochemicals in *neem* against SARS-CoV-2 has been demonstrated by various workers. For example, Baildya *et al*. (2020) studied the effect of *neem* extracts on the new coronavirus SARS-CoV-2's papain-like protease (PLpro). All of the Neem compounds under investigation exhibited respectable levels of inhibitory efficacy against the SARS-CoV-2 PLpro. Desacetylgedunin (DCG), which can be found in Neem seeds, had the strongest affinity for PLpro of all of them. They came to the conclusion that DCG on PLpro may aid in the treatment of SARS-CoV-2.

Parida *et al.* (2020) explored phytochemicals as potential inhibitors for SARS-CoV-2 by performing all atom molecular dynamics simulations using high performance computing for 8 rationally screened phytochemicals from *Withania somnifera* and *Azadirachta indica* and two repurposed drugs docked with the spike glycoprotein and the main protease of SARS-CoV-2. The results revealed that Withanolide R (−141.96 KJ/mol) and 2,3-Dihydrowithaferin A (−87.60 KJ/mol) were with the lowest relative free energy of binding for main protease and the spike proteins respectively. It was also observed that the phytochemicals exhibit a remarkable multipotency with the ability to modulate various human biological pathways especially pathways in cancer.

Balkrishna *et al*. (2021) experimentally proved that Nimbocinol and sage exhibited maximum inhibitory effect on replication against PLpro SARS-CoV-2 which was found to be better than remdesivir, chloroquine and favipiravir.

Potentiality of *neem* against SARS-CoV-2 was also supported by Lim *et al.* (2021) by highlighting antiviral research with favourable results specific to the SARS-CoV-2 coronavirus. Affect and remedy of SARS-CoV-2 upon the human body were clearly explained by Eze *et al*. (2022). Similar to SARS-CoV and the Middle East respiratory disease (MERS) virus, SARS-CoV-2 is an enclosed virus with a positive sense, single-stranded RNA genome. Because COVID-19 has a potential to damage many organs in a severe instance, which can result in multiple organ distress syndrome (MODS), it is unusual in that it can have a high morbidity and fatality rate. Severe COVID-19 risk factors include cardiovascular disease, diabetes and its consequences, and obesity. As a result of SARS-CoV-2 infection, the body produces excessive amounts of cytokines, chemokines, reactive oxygen species, nitric oxide, oxidative stress, acute phase proteins (such as C-reactive protein), and other pro-inflammatory compounds. A cytokine storm is produced in the worst case scenario.

Eze *et al*. (2022) hypothesised that an effective technique for preventing COVID-19 would be the displacement of the virus attached to the vascular endothelium (VE) and suppression of virus binding. As a result, *neem* leaf extract will be an efficient therapeutic formulation against COVID-19 by preventing the binding of SARS-CoV-2 to the vascular endothelium (VE).

The development of a vaccine and antiviral therapy are being challenged as a result of concurrent alterations in the SARS-CoV-2 genome. A useful investigation against SARS-CoV-2 was made by Sarkar *et al.* (2022) using *neem* bark extract (NBE). They looked into the SARS-CoV-2 and the m-CoV-RSA59 infections. On viral load, inflammatory response, and histological alterations in m-CoV-RSA59 infection, the effects of in vivo intranasal or oral NBE treatment were evaluated. Administration of neem bark extract (NBE) prevents SARS-CoV-2 and m-CoV-RSA59 infection and replication *in vitro* m-CoV-RSA59 infection is effectively inhibited *in vitro* by isolated fractions of *neem* bark extract (NBE) that are rich in nimbin isomers. *Neem* bark extract (NBE) contains triterpenoids, which may enable them to selectively target a variety of viral proteins to prevent mouse and various human coronavirus infections. It will be speculating on possible application as a pan-Coronavirus antiviral.

Cultivation tips

Agro-climatic requirements

It can be grown almost everywhere in the lowland tropics, with sub-arid to sub-humid conditions. It prefers tropical and sub-tropical climates at altitudes between 0-1500 m with annual temperature ranges between 21-32^{o}C but can tolerate up to maximum 40^{o}C and mean annual rainfall varies between 400-1200 mm. It does not tolerate cold temperature or saturated soils (Kumar *el al.,* 2019). It cannot tolerate the temperature below 5°C (Nisa *et al.,* 2022). It can also resistant against drought. Neem tree unable to withstand against waterlogged conditions and quickly dies in these conditions. It can grow on a range of soil types, including black cotton soils with a pH range of 5 to 10, from sandy to clayey soils, and other types of soil as well.

Propagation and plating

It can be propagated using seed as well as root shoot cutting. The viability of seeds lasts only for around two weeks. The endocarp is removed from seeds or the seed coat at the round end is cut off with a sharp knife in order to increase germination. Seeds are then immersed in cold water for 24 hours. In sandy nursery beds, de-pulped seeds should be distributed in lines (15 to 20 cm apart), 2.5 to 5 cm apart, at a depth of 1 to 2 cm, and lightly covered by soil. To avoid cracking, nursery beds should be irrigated frequently. It usually takes one to two weeks for germination. 70% to 90% of seeds sprout. Seedlings are transplanted in the main field during rainy season after 1 to 2 years at a spacing of 4.5 to 5.5 m (*Anon*., 2012).

Planting root-shoot cuttings

The stumps are made from seedlings that are 12 to 13 months old, with 2.5 cm of shoot section and 23 cm of root, and they are planted in crow bar holes at the end of the rainy season. In Tamil Nadu, stumps from plants that are two years old have a higher survival rate and better height growth than root stock that is one year old. 53% success with root-shoot cuts has been reported from Maharashtra. Rains are necessary for root-shoot cuttings to succeed; a protracted dry spell may significantly reduce survival rates (*Anon*., 2012).

Interculture operation

Young plantations can benefit from strip weeding for their survival and general health. The first year only needs two weedings, while the second year just needs one. Weeding can be carried out mechanically or manually. When seedlings are transplanted, the first mechanical thinning is carried out at the age of 5 years.

Plant protection

In nursery stage the plants are affected by different pests and diseases. After attaining 2-3 years age, they are capable to stand well against various pest and diseases attack. Tip borer (*Laspeyresia koenigiana*) and Tea mosquito bug (*Heliopeltis antonii*) are the two most frequent pests that attack plants in their seedling and younger stages. The main pest of *neem* is the scale insect *Pulvinaris maxima*, and *Heliothrips haemorrhoidalis* is a potential pest. Damping off disease, which is brought on by *Rhizoctonia* species, can have a serious impact on *neem* seedlings. There are other diseases as well, such as leaf web blight, leaf spot, and blights brought on by *Colletotrichum, Alternaria, and Pseudocercospora*. To manage the aforementioned insect pests and illnesses, systematic insecticides and fungicides are utilized.

Harvest and yield

Scented white flowers appear during March-April in abundance on auxiliary spikes. Fruits are ripening from June to August. Fruits are picked when they transform from green to a bright yellow tint. To collect the ripe fruits, the branches must be shaken firmly. About 8000- 10000 kg of fresh berries can be harvested from 1ha/ year (*Anon*., 1973). Fresh fruits give about 60% dry fruits, which yield 10% kernel and contain on an average 45% fixed oil.

Cost of Production

Approximate cost of production comes around Rs.37, 500/- per hectare (*Anon*., 1973).

Post-harvest management or processing

Neem oil

The seeds must first be cracked open and the kernels separated in order to produce *neem* oil. The kernels are subsequently squeezed in ghani or industrial expellers. Sometimes, the oil output might reach 50% of the weight of the kernel (Latif *et al.,* 2020).

The neem seeds were repeatedly washed to eliminate dirt and other clinging contaminants prior to oil extraction, and they were then dried in an oven at 50^0C until they reached consistent moisture content (Anon., 2022).

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Review article

Biodiversity of fruit plants in Azerbaijan: Prospects of conservation and utilization

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ABSTRACT

Azerbaijan considered as the primary or secondary center of origin, domestication and diversity of many fruits, grapevines and nuts, taking their basis from the wild relatives of the region. The land races and wild relatives in Azerbaijan are conserved in protected areas and botanical gardens, as well as ex-situ and on-farm conservation in field collections of the National Gene Bank. At present, more than 4000 accessions of fruit plants are maintained in field collections of GRI (Genetic Resources Institute (GRI), Ministry of Science and Education Republic of Azerbaijan). GRI maintains valuable collections of apple, pear, quince, medlar, pomegranate, grapevine, fig, cherry, apricot, almond, pistachio, almond, pomegranate, sea-buckthorn and other crops. Despite the broad utilization of native varieties in conventional agriculture, the traditions of usage of wild relatives for food, food additives or environmental purposes are still kept among populations like fresh fruit consumption as a raw material for processing and making traditional food, including in a food chain as a feed for domestic animals, initial materials for breeding. Except for fruit utilization, wild relatives are also used as rootstocks, live fences; honey, decorative and medical plants, and wind-brake in plantations.

***Keywords*:** Conservation, fruit crops, landraces, utilization, wild relatives

INTRODUCTION

Azerbaijan is one of the richest countries regarding agricultural biodiversity of plants, animals and microorganisms in the world. Agricultural biodiversity can only be protected and sustainable by rural communities using traditional and ecological agricultural techniques and implementations. Many of these globally important environmental and human friendly traditional agricultural production and information systems are abandoned by producers due to the development of modern agricultural techniques and marketing problems. Thus they are under danger of vanishing.

Traditional agricultural knowledge which is related to indigenous technologies and traditional farming and crop beliefs associated with different cycles of crop cultivation or utilization such kind of species, is deteriorating faster than their biological/genetic diversity which are used by local farmers for different purposes of their consumption. Local people are often excellent resource managers when they are allowed to manage their own resources for their own benefit. Land races are the products of their selection and even multiplication over long periods of time. Therefore, their participation to the conservation of bio/agro-biodiversity is essential to identify how more complex traditional systems can be adapted to modern needs, while still retaining the bio/agro-biodiversity of both agro-ecosystem and its surroundings.

Flora of Azerbaijan Republic contains more than 5000 species of vascular plants, including 800 ether-oil yielding, 600 medicinal, 500 spices-aromatic, 500 of vitamin-important, 850 dyeing and 1500 species with tannin, have been revealed, 237 of which are endemic. They grow along Major and Minor Caucasus slopes and in subtropics of Talish Mountains.

The **South Caucasus**, also known as **Transcaucasia** or the **Transcaucasus**, is a geographical region on the border of Eastern

Europe and West Asia, straddling the southern Caucasus Mountains. The South Caucasus and the North Caucasus together comprise the larger Caucasus geographical region that divides Eurasia. The South Caucasus in particular, Azerbaijan, considered is considered to be the primary or secondary center of the origin, domestication and diversity of many fruits, grapevine and nuts, taking their basis from the crop wild relatives (CWR) of the region, while the wild relatives (WR) of major fruits and nuts like grapevine, apple, pear, quince, medlar, pomegranate, fig, cherry, apricot, almond, hazelnut, walnut, chestnut, pistachio and others are presented with many genera in the flora of these countries. According to Zhukovskii (1964), the South Caucasus should be considered as a hearth of evolution for cultivated plants of great independent importance.

Vavilov (1926), in his works about the world centers of origin of cultivated plants includes South Caucasus in the Southwest Asia Center and pointed out (Vavilov,1931) existance of about 80 species and genus of fruit trees in the Caucasus. He specifies the South Caucasus as the major region for fruits in the Caucasus, the centre of origin and center of domestication of grapevine, pears, cherry plum, pomegranate, sweet cherry, walnut, quince, almond, fig, medlar, Cornelian cherry, *Punica granatum* L., *Cidonia oblonga* Mill., *Diospyros lotus* L., *Pyrus elaeagnifolia* Pall., *Pyrus syriaca* Boiss., *Cerasus incana* Spach., *Cerasus micocarpa* Boiss., *Amigdalus georgica* Desf., *Ficus carica* L., *Corylus colurna* L., *Corylus colchica* Alb., *Prunus divaricata* Lebed., *Cerasu savium* (L.) Moench, *Mespilus germanica* L., *Vitis vinifera* L., *Cornus mass* L.). He also indicates growing and wide form origin processes for *Laurocerasus officinalis* L., *Castanea sativa* Mill., *Amygdalus fenzliana* (Fritsch) Lipsky., *Amygdalus orientalis* Mill, *Pyrus communis* L., *Corulus avellana* L., *Juglans regia* L. here. The area of distribution for *Pyrus communis* L.*, Malus domestica* Borkh.*, Corylus avellana* and *Jeglans regia* are wide and goes out of the Caucasus, but in the South Caucasus they have rich local germplasms.

MATERIALS AND METHODS

Materials for research included the wild relatives and local varieties of fruit crops grown in field collections of the Genetic Resources Institute Ministry of Science and Education Republic of Azerbaijan and other institutions and by farmers in different regions of the country.

Phenological phases, growth, biomorphological description and productivity, fruit quality traits, resistance to disease and pests were studied by using the common description methods of fruit plants as described by Marozova (1987); Smirnov *et al*. (1987); Michurinsk (1980); Sedova (1999). The different varieties and forms of fruits from different regions were collected from following geographical locations as follows:

N 41^{0}17.5470/ E47^{0}2.4890/ 504,4 m;
N 40^{o}52.127 E 48^o 05.769 475 m;
N 40 ^{o}58/33.09// E47^{0}51/48.46// 839 m;
N 41^{o}04.883 E 47^o 17.933 526 m;
N 41 ^{o}21/ 59,5// E 048^0 24/ 03,0// 905m;
N 41 ^{o}29/ 53,4// E 048^0 43/ 17,3// 122 m;
N 38 ^{o}53,357/ E048^{0}43,556/;
N 38 ^{o}32,555/ E048^{0}49,582/;
N 41^{0}8.1130/ E47^{0}10.9500/ 351,9 m
N 40^o 36.811 E48^o 26.688 1044 m.
N 40^{o}47.781 E 48^o 19.141 835 m.
N 41^{o}00.695 E47^o 27.435 481 m
N 41^{0}17.9240/ E47^{0}6.6430/ 764,4 m
N 41^{o}38.611 E 48^{o}45.156 18 m.
N 41^{o}33.504 E 48^o 42.920 67 m.
N 38 ^{o}32,571/ E048^{0}49,539/
N 39^o 26' 46// E 45^o 35' 31" 1304 m.

OBSERVATIONS

Each region of Azerbaijan has its own specific and qualitative fruit varieties. Below we described the names of some varieties of national selection of fruit crops discovered during expedition to the regions of Azerbaijan:

1. **Shirvan region:** Its location is 39°552 553 N 48°552 133 E. It is famous for its pomegranate and quince. A number of local varieties of pomegranate (*Punica granatum* L.) namely Guloysha, Malas, Shahnar, Balmursal, Girmizigabig, Nazikgabig and etc.; of quince

(*Cydonia oblonga* Mill.) namely Jardam, Garaheyva, Sari heyva, Armuduheyva, Qaraman, as well as of plum, sloe, grapevine and etc. are cultivated in Shirvan region.

2. **Shaki-Zagatala region:** Zagatala location is Latitude: 41° 37' 53.76" N Longitude: 46° 38' 41.24" E. It borders Russia to the north and Georgia to the west, as well as the economic regions of Quba-Khachmaz, Mountainous Shirvan, Central Aran, and Ganja-Dashkasan. This region for its walnut and hazelnut; In this region ancient landraces of hazelnut (*Corylus avellana* L.) namely Ata-Baba, Yaghlifindig, Sachaglifindig, Ganja findigi; of walnut (*Juglansregia* L.) namely Jar, Dundi, Gum, Tala, Zagatala; of chestnut (*Castanea sativa* Mill.) namely Khanlig, Ashig, Farash, Barguvara and etc. are grown.
3. **Guba-Khachmaz region:** It location is N 41° 24' 7.758", E 48° 40' 30.3924". This region possesses more than hundred varieties of apple namely, Qizil Ahmedi, Sari tursh, Jirhaji, Sikhijani, Ayyubi, Shirvangozeli, Jibir, Gand alma; pear varieties like Peyghambari armud, Abasbeyi, Agh armud, Alpanqalý, Qarpýz armud, Nar armud, Kurduku, Daqur, Qefeyi, Jýr Nadiri armud, Dash armud, Sapýburma armud, Qara tuluq, Sheker armud, Zencirbend, Ýspigi, Pas armudu, etc. were spread widely in fruit gardens, especially in amateur gardeners' courtyards. But some of these varieties are on the verge of extinction, they are very rare and in very small quantities, for example - apple varieties Sari tursh, Qara Tursh, Ayyubi, Jibir, Gand alma; pear varieties Peyghambari armud, Alpanqalý, Qarpýz armud, Daqur, Qefeyi, Bardaq armud, Buz armud, Sapýburma armud, Qara tuluq, Sheker armud, Zencirbend, Ýspigi; grape varieties Hinbi and Shikhveli (Hajiyev and Musayev, 2022).

   In Nabran forests of **Guba-Khachmaz region** dark and dark purple coloured grape forms were found. While expedition in Guba-Khachmaz region it was known that, Guba region is enriched with wild grape. In forests of this region (Uzunmeshe, Alpan, Khujbala, Digah, Aghbil, SusayGishlag, Dallakand villages) along Guruchay, Gusarchay, Gudyalchay rivers lots of wild grape forms were found. In forests of Khachmaz (Pir forest), Shaky (Oraban), Lankaran (Seligavul) and Gabala (Shongar) regions small seedy dark wild grape varieties were also determined.

   Typical forest formation of wild grape was found in Agharehimoba, Godekli, Gimilgishlag, Gadashoba, Nerecan and etc. villages and forests (forest number 1, Pir forest) of Khachmaz region, Seligavul forest of Lankaran region and Shongar spring of Gabala region and coastal formation was found in Nabran forests of Khachmaz region (Musayev and Akparov, 2013)

   On the banks of Kungutriver (Oraban village) of Sheki, Guruchay, Gusarchay, Gudyalchay rivers (Uzunmeshe, Alpan, Khujbala, Digah, Akbil, SusayGishlag, Dallakand villages) of Guba region wild grapevines spread mainly in tugay forests densely and widely.

   Wild sea-buckthorn brushes are widely spread in most regions of the Republic – Guba Khachmaz, Shaky-Zagatala, Shirvan, Talysh, Garabagh, Nakhchivan AR and etc. While natural seabuckthorn populations are mainly spread in Ismayilli, Shamakhi, Aghsu, Gabala, Shaky, Gakh, Zagatala, Guba, Gusar and etc. regions. Sea-buckthorn is growing very well almost in all places, being part of riparian forest 'tugay' (Musayev and Akparov, 2012; Musayev, 2013; Letchamo *et al*., 2018).
4. **Absheron region:** Its geographical position is **N** 40° 10' 17.9292", E 49° 42' 23.5908". The region contains varieties of grape, fig, pistachio, almond, oleaster (*Elaeagnu sangustifolia* L., *E. caspica* Grossh.), mulberry, quince and pomegranate where these crops grow naturally or cultivated by farmers in their holdings and orchards. Aboriginal varieties of olive (*Olea europaea* L.) namely Shirinzeytun, Azerbaijan zeytunu, Armuduzeytun, Bakizeytunu; of fig (*Ficus carica* L.) namely Absheron sari injiri, Buzovburnu, Goy injir, Garainjir, Bozinjir, Sumakhinjiri, Payizinjiri; of almond (*Amygdalus communis* L.) namely Nazikgabig, Sarayi, Mardakan; of pistachio (*Pistacia vera* L.) namely Amirjan, Bulbula, Narinji, Zumrud and etc. are cultivated in Absheron region.

5. **Garabagh region:** It location is N 39°45' 36" E 46°45' 01" This territory possess a number of fruit crops like landraces of apple, pear, quince sweet cherry, cherry, pomegranate, fig, grape and other fruits which were used by local people.
6. **Nakhchivan Autonomous Republic:** It location is N 39°12' 32" E 45°24' 44" This region have more valuable varieties of grapevine and stone-fruits are cultivated. There are some famous varieties of apricot (*Armeniaca vulgaris* Lam.); varieties of peach (*Persica vulgaris* Mill.) namely Salami, Zafarani, Juyur, Aghkustu, Aghnazli; varieties of plum namely Garaalbukhara, Sari albukhara, Khatini; varieties of alycha (*Prunus cerasifera* Ehrh. var. *divaricata* (Ledeb.) L.H.Bailey) namely Goychasultani, Shabrani, Payizmalasi, Aghalycha; varieties of walnut namely Sugra, Seyfi, Araz, Disar and etc.
7. **Ganja-Gazakh region:** It location is N 40°40' 58" E 46°21' 38" This region, there are many aboriginal varieties of stoned-fruits and berries and subtropical fruits as well as grapevine, for example, cornelian cherry (*Cornus mas* L.) namely Armuduzogal, Challakzogal, Girdazogal, Dilimlizogal, Garazogal, Sari Kahrabazogal, Irimeyvalizogal are available.
8. **Lankaran-Astara region:** Latitude: 38.7537311; Longitude: 48.8539286. It borders Iran to the south and west, Caspian Sea to the east and Shirvan-Salyan Economic Region to the north. In this region wild medlar (*M. germanica* L.) widely spread together with wild pomegranate (*P. granatum* L.) and quince (*C. oblonga* Mill.). Some of cultivates medlar varieties have names like 'Khan ezgili', 'Nelbeki', 'Kitil', 'Aghezgil', 'Arkivanezgili' (Akparov and Musayev, 2012.,). It was reported that the medlar was domesticated by Caucasian inhabitants, especially in the Lankaran-Astara region of Azerbaijan.

In the territory of Azerbaijan: 149 species of fruit crops belonging to 39 genera and 15 families are distributed.

In forests and rural regions of Azerbaijan: Big number of genera and species of wild fruit and fruit-berry plants are available which provides the greatest diversity of fruit crops, viz., *Amygdalus communis* L., *Armeniaca vulgaris* Lam., *Berberis vulgaris* L., *Castanea sativa* Mill., *Cerasus avium* (L.) Moench, *C.vulgaris* Mill., *Cornus mas* L., *Corylus avellana* L., *Crataegus orientalis* Pall. ex M. Bieb., *Cydonia oblonga* Mill., *Ficus carica* L., *Fragaria vesca* L., *Hippophae rhamnoides* L., *Juglans regia* L., *Malus domestica* Borkh., *Mespilus germanica* L., *Morus* L., *Persica vulgaris* Mill., *Pistacia mutica* Fisch. & C. A. Mey., *Pistacia vera* L., *Prunus cerasifera* Ehrh., *P. domestica* L., *P. spinosa* L., *Punica granatum* L., *Elaeagnus angustifolia* L., *Pyrus communis* L., *Rubus* L., *Vitis vinifera* L. subsp. *sativa* D.C., *V. vinifera* L. subsp. *sylvestris* (C. C. Gmel.) Hegi. and etc.

In river valleys and other places: many wild forms of apple (*Malus* L.) are available.

In Coastal forest area of Caspian Sea: Wild forms of quince are found. These forests represent service tree (*Sorbus* L.) with 11 species (5 of them are endemic to the Caucasus); hawthorn (*Garataegus* L.) has 9 species; plum (*Prunus* Mill.) with 3 species, almond (*Amygdalus* L.) – 2 species, cherry (*Cerasus* Juss.) – 5 species, blackberry and raspberry (*Rubus* L.) –14 species and currant (*Ribes* L.) with 2 species. Furthermore, wild medlar (*Mespilus germanica* L.), sloe (*Prunus spinosa* L.), alycha (*Prunus divaricata* lebed.), pomegranate (*Punica granatum* L.), sweet cherry (*Cerasus avium* (L.) Moench.), dog-rose (*Rosa sp*.), sea-buckthorn (*Hippophae rhamnoides*), cornel (*Cornus mas* L.), grape (*Vitis sylvestris* Gmel.), nuts and other fruit and fruit-berry crops in the forest are availble. (Akparov and Musayev, 2012; Musayev and Akparov, 2013; Asadov and Asadov, 2001; Maghradze *et al*., 2012 and Mammadov *et al.,* 2000)

Among 27 Caucasian pear species, 19 are grown in Azerbaijan. These are *Pyrus boisseriana* Buhse., *P.hyrcana* Fed., *P. grossheimii* Fed*., P. communis* L., *P. caucasica* Fed., *P. eldarca* A.Grossh., *P.voronovii* Rubtz., *P. syriaca* Bioss., *P. salicifolia* Pall., *P. zangezura* Maleev, *P.elata* Rubtz, *P. raddeana G.*Woron, *P. serotina* Rehd., *P. nutans* Rubtz, *P.vsevolodi* Heidemann., *P. oxyprion* G.Woron., *P. complexa* Rubtz., *P. medvedevii Rubtz*. vY *P. georgica* Kuth.) with a number of spontaneous hybrids.

In Azerbaijan there were more than 400 landraces of pear and half of them were endangered (Rajabli, 1966). Some varieties of pear (*Pyrus*

communis L.) are: Abbasbeyi, Agh gulabi, Aghagormez, Bildirchin budu, Chaxma, Nurun-burun, Qorxmazý, Talýbý, Kup armudu, Uzun Mustafa, Xemzeyi armud, Jirnadiri, Ispigi, Kurduku, Nargila, Qýrmýzý yanaq, Zerqava armud, Hazar armud, Letenzi, Nar armud, Nelbeki armud, Peyghambari armud, Shekeri, Sulu armud, Tir armudu, Tursh sini armud, Turshmalasi armud, Usun armud, Usun sap armud, Yag armud, etc. These varieties differ for ripening time (summer, autumn and winter), size, taste quality, productivity and different factors. For instance, Aghagormez, Bildirchin budu are early ripening, whereas Goy armud is productive (1 ton per tree) (Akparov and Musayev, 2012).

Cornelian cherry (*C. mas* L.) is widely spread in the countries and used in local cuisine. It grows in forest with other fruit species like cherry plum, sloe, hawthorn, dog-rose, apple, pear, quince, medlar, hazelnut, currant, raspberry and others. There are variations of cornelian cherry in Azerbaijan having different color, size and shape of fruits. More than 40 forms of Cornelian cherry was reported by Mammadov and Musayev (2011).

The Common (syn. Persian, English) walnut *J. regia* L. wildly grows in Azerbaijan in lower and middle slopes of the Major and Minor Caucasus and in subtropics of Talish Mountains.

According to Safarov (1981), the total surface of walnut forests is more than 25.000 ha in Azerbaijan. Based on fossils observation, it was approved that the walnut was spread in Azerbaijan during the Tertiary Period and it is a relict plant of Cretaceous Period. The local wild forms of *J. regia* being in basis of native walnut germplasm. The Azerbaijan selective forms are 'Kaghizi', 'Katankoynak', 'Araz', 'Disar', 'Darvishpapag', 'Nazikgabig'. However, 'Evrica' and 'Blecmer' cultivars had been selected from 'Kaghizi' cultivar (Akparov and Musayev, 2011, Ibrahimov, 2007).

Wild grapevine - *V.vinifera* L. subsp. *sylvestris* (C. C. Gmel.) Hegi., the wild ancestor of the cultivated grapevine *V. vinifera* ssp. *sativa* D.C., is a typical plant of flora in Azerbaijan, spread widely in large areas and in the banks and shores of river, lake and sea and mountain slopes. This wild grapevine together with native varieties is interesting, while the Azerbaijan - as one of the main centre of origin and domestication of cultivated grapevine. Confirmations of this opinion are high number of autochthonous varieties with ample diversity of berry colour and technological aptitudes; historical information; linguistic and folk data; and certainly, rich palaeobotanical and archaeological findings discovered since "*Shomutapa culture*", dated back to VI-IV millennium BC (Pipia *et al.,* 2012; Amanov *et al.,* 2012; Dong *et al*.,2023).Wild grape spread on the whole territory of Azerbaijan is very ancient formation. In general, more than 3000 samples of wild grapes were found in expeditionary regions and phytocenotic features of their spreading areas were described. (Musayev and Akparov, 2013.) It is spread on the territory of Azerbaijan from 18 m below sea-level (Kyur riverside, Salyan region) to 2000 m above sea-level (Gusar region). There are two kinds of wild grape in Azerbaijan: *typical* Negr. (with hairs) and *aberrans* Negr. (hairless). On the banks of Kondalanchayriver in Fuzuli region dark, dark red, dark purple coloured grape seed forms were observed. At the result of investigations it was determined that, different populations of wild grape in republic of Azerbaijan, spread mainly in three locations *viz*., tugay (streamside forest), typical broad-leaved forests and coastal area of the Caspian Sea.

Utilization of native varieties

1. **Direct fresh fruit consumption**: The fruit crops like almond, pear, medlar, wild strawberry, raspberry, blackberry, barberry, hawthorn, sea-buckthorn, hazelnut, walnut, cherry plum, cornelian cherry, chestnut, mulberry, Caucasian persimmon, pomegranate and others are directly consumed as fresh.

2. Utilization as raw materials for processing and traditional cuisine: wild fig, mulberry, pomegranate, walnut, sloe, apple, pear, apricot, cornelian chery, persimon, oleaster, ash berry, bilberry, current, cherry plum, cherry laurel, gooseberry, hazelnut, hawthorn, Caucasian persimon, sea-buckthorn, snowball, quince, wild rose and others. These fruits are used for preparation juice, syrup ('Behmez', 'Doshab'), puree, preserve ('Muraba'), dried fruits ('Axta', 'Movuc', Alana', 'Mianpur'), dried layers ('Lavashana'), jam, morse, alcoholic (Wine, 'Araki, Liqueur) and non-alcoholic ('Limonade') beverages, candy, species

and souces per dishes ('Lavangi', 'Abgora, 'Sujuq', 'Narsharab'), surrogates (tea, coffee), marinade, 'Kiesel', confectionery ('Badambura', 'Halva', 'Shakarbura', 'Pakhlava', 'Fasali'), others;

3. In a food chain as a feed for domestic animals like sea-buckthorn, mulberry, nuts and others;

4. **Utilization in breeding**, when the old autochthonous varieties took origin from the native WR of apple, pear, apricot, pomegranate, cherry plum, quince, grapevine, sweet cherry, fig, hazelnut; and when the advanced forms for cultivation were selected within cornelian cherry, cherry plum, pear, walnut, almond in the XXth century;

5. **Utilization in rootstock selection:** the Wild Relatives of pear, apple, (peach), quince, cornelian cherry, cherry plum, sloe, Caucasian persimmon, wild rose, hawthorn are used.

6. Utilization in for live fences - blackberry, barberry, sea-buckthorn, hawthorn, oleaster, cherry plum, sloe, Pyrus salicifolia and others are used.

7. As honey, decorative and medical plants- suitable for making anti-erosion and wind-brake line plantations; forest garden construction.

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Review article

Invasion, population build-up and possible management strategy of rugose spiraling whitefly (*Aleurodicus rugioperculatus* Martin) on minor fruit crops as well as on medicinal, aromatic, and ornamental plants in India – Current scenario

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ABSTRACT

The occurrence of rugose spiraling whitefly, an invasive pest, was established in 22 counties worldwide including India. Its invasion in the country was first reported from coconut plantation in Tamil Nadu during 2016. Presently its presence has been noticed in almost all the southern and peninsular states of the country along with Gujarat of western part and West Bengal and Assam of eastern part. Till date 67 plant species have been identified as the host of this pest including minor fruits, medicinal, aromatic, ornamental, avenue plants. This review article aimed to create awareness to all the stakeholders including scientists, researchers, farmers, students and common man regarding its host diversity among minor fruits, medicinal, aromatic and ornamental plants in the country at present situation so that attention may be paid on its sustainable management in time.

***Keywords*:** *Aleurodicus rugioperculatus*, aromatic plants, invasion, medicinal and ornamental plants, minor fruits, population build-up

INTRODUCTION

Rugose spiraling whitefly (*Aleurodicus rugioperculatus* Martin; Aleyrodidae; Homoptera) is an invasive and destructive pest of different crops. This whitefly is endemic in America and was previously termed as gumbo limbo spiraling whitefly. Since a relatively newer pest which was discovered just few decades ago, not so much information is available for this pest. This pest was first described from the infested coconut leaves collected from a coconut plantation located at Belize, Florida. The natural habitat of this pest was located at Belize, Guatemala and Mexico and the pest belongs to the *niveus* species-group under the genus *Aleurodicus*. From the originating region the whitefly has been amplified its habitat towards 22 Central and South American countries including Florida and USA. They are characterized by their unique polyphagous nature with huge host range involved with 118 plant species representing 43 plant families covering the plants having economic importance in America (Francis *et al.,* 2016). The occurrence of this whitefly species was confirmed in 22 counties. Its introduction in India is believed to be occurred by trading of ornamental plants (Shanas *et al.,* 2016). Invasion of this pest in India was first reported in the month of September, 2016 from a coconut plantation located at Coimbatore, Tamil Nadu (Sundararaj and Selvaraj, 2017). In Kerala and Tamil Nadu coconut plants were severely infested by this pest and 17 plant species were found to be susceptible to this pest in Kerala (Sundararaj and Selvaraj, 2017; Selvaraj *et al.*, 2017). Subsequent report of occurrence of this pest have been came from different states including Andhra Pradesh, Assam, Karnataka and Kerala (Rao *et al.,* 2018), West Bengal (Selvaraj *et al.,* 2019a) and Gujarat (Jethva *et al.*, 2020b). According to Selvaraj *et al.* (2019a) this pest was probably introduced to Purba Midnapur and Howrah district through transport of plant materials from infested areas either from Orissa or South India. Potential suitable areas for establishment of this pest were mostly found in all coastal and southern states of India. It prefers a warm and humid climate, indicating that the tropics,

subtropics and even temperate regions like Mexico (Evans, 2008) are ideal for its spread and invasion. The results of Maruthadurai *et al*. (2023) highlighted that the suitable habitat area for *A*. *rugioperculatus* was predicted to increase and highest probability of invasion and spread in 2050 and 2070 under future climate change scenarios. On the other hand, India represents diverse agro-climatic conditions having unique soil and plant types across the country. Diverse agro-climatic conditions enable the country for bearing different fruit plant species.

Apart from the major fruits and minor fruits like Cape gooseberry (*Physalis peruviana* L.) (Dahiya *et al*., 2022), wood apple (*Feronia limonia*) (Mahato *et al*., 2021), walnut (*Juglans regia* L.) (Kumar *et al*., 2020) *etc.,* India is also a hotspot for different aromatic, medicinal and ornamental plants. Majority of these plants are somewhat ravaged by different insect pests. Ornamental plants are more preferred by this invasive pest (Stocks and Hodges, 2012). It is also observed that the ecological conditions of Andhra Pradesh, Odisha, Madhya Pradesh, Bihar, Uttar Pradesh, Uttarakhand, Chhattisgarh and West Bengal are very congenial for multiplication of the pest (Selvaraj *et al.*, 2017). Therefore, it will not be an exaggeration to say that this rugose spiraling whitefly is also a potential threat to the production of minor fruit crops as well as medicinal and aromatic plants. Again, information regarding the susceptibility of different minor fruit crops along with other ornamental, medicinal and aromatic plants including weed hosts against this pest is necessary for implementing effective, environmentally safe and economically viable management strategies for the respective crops. Limited studies have been conducted focusing on range of minor fruit, medicinal, aromatic and ornamental crops acted as hosts of this pest. Aim of this study is to aware the scientists, researchers, planters and common people regarding the identification of host crops of rugose spiraling whitefly among the aforementioned groups of crops in Indian context so that the concerned people may pay the attention on its sustainable management.

Infestation of rugose spiraling whitefly on different host plants

The presence of rugose spiraling whitefly can be identified by the presence of egg spirals on the underside of the infested leaves. Eggs are laid on the leaves in a spiraling pattern with a tiny stalk. Yellowish nymphs create white waxy filament that covered all the life stages of the whitefly. Adults are quite larger in size and possess two pale brown wavy makings on their forewings. Infested plants can be easily diagnosed from a distance by noticing the heavy coating of white filamentous waxy materials on the underside of the leaves. Presence of honeydew along with black sooty mold and premature leaf drop is also an important diagnostic character for rugose spiraling whitefly infestation (Mayer *et al.*, 2010). Occurrence of rugose spiralling whitefly in different host plants in India has been evaluated. Srinivasan *et al*. (2016) recorded 70% whitefly infestation on banana. Observations of Selvaraj *et al.* (2016) suggested that coconut palms were severely damaged by the pest where infestation ranged from 40-60% and 25-40% leaf infestation was observed in banana. Severe drying and scorching of leaves due to whitefly infestation in banana orchard was noticed in certain places of Tamil Nadu and Kerala. In Mangaluru and Udupi, 20 to 35% infestation of coconut plantation and 24 to 38 % banana plantation were infested by this whitefly during 2017 (Selvaraj *et al.,* 2017). In West Godavari district of Andhra Pradesh it was observed that per cent infestation caused by rugose spiraling whitefly ranged between 75.85-95.0%, 82.93% and 63.50-79.0% on coconut, banana and guava respectively, whereas the incidence was ranged from 81.42-100.0%, 83.13-100.0% and 83.13-100.0% on coconut, banana and guava, respectively (Sushmitha *et al.,* 2020b). A Tamil Nadu based survey by Elango and Nelson (2020a) revealed that a total of 20 host plant species from 15 families were recorded as hosts of *A*. *rugioperculatus*. Among them, 8 hosts viz. *Cocos nucifera*, *Musa paradisiaca*, *Annona squamosa*, *Citrus limon*, *Myristica fragrans*, *Psidium guajava*, *Theobroma cacao* and *Manihot esculanta* had all the life stages of whitefly, whereas in other 12 host plants viz. *Areca catechu*, *Azadirachta indica*, *Jatropha curcas*, *Mangifera indica*, *Abelmoschus esculentus*, *Piper nigrum*, *Achras zapota*, *Solanum melongena, Gossypium hirsutum, Zea mays, Pennisetum glaucum* and *Hibiscus rosasinensis* the eggs stages only were observed. Another survey based

experiment in Bhubaneswar (Sri *et al.*, 2022) highlighted that 85.0-100.0%, 100.0% and 35.0% whitefly incidence was occurred on coconut, banana and guava, respectively. In a study, almost 100% incidence was observed on the plant belonging to the family Amaryllidaceae, Moraceae, Malvaceae, Magnoliaceae, Combretaceae, Myrtaceae and Sterlitzaceae. The plant families Annonaceae, Arecaceae and Sapotaceae experienced more than 90% whitefly incidence. A survey work in Tamil Nadu (Nandhini and Srinivasan, 2022) revealed that amongst the 67 host plants recorded, 17 fell under the ornamental plants category, followed by 14 under fruit crops, 9 under medicinal plants, 5 under vegetables, 4 under fibre crops, 2 each under biofuels, flower crops, green leaf manures, millets, oilseeds, plantation crops and timber crops, besides 1 host each under pulses, avenue trees, fodder crop and tuber crops. Das *et al*. (2023) from Bangladesh reported that 49.18% fruit plants, 21.31% ornamental and floral plants, 15-20% arecanut plants, 8.19% field crops, 8.19% forest plants and 13.13% miscellaneous plants were infested by this pest.

Bio-ecological features of rugose spiraling whitefly

The concerned whitefly *A. rugioperculatus* pertained to the subfamily Aleurodicinae under the family Aleyrodidae belongs to the suborder Sternorrhyncha under the order Hemiptera. Characteristic depositing of eggs in a spiral manner is the reason for describing them as spiraling whitefly and since they have irregularly corrugated or fluted (Rugose) operculum in their pseudopuparial stage they are termed as rugose spiraling whitefly. They are comparatively larger (almost three times) than other whitefly species with sluggish movement. They can be differentiated from other whiteflies with distinct simple and compound pores. The 4th instar i.e. the puparium is distinctly thick and larger than the commonly occurring species *Bemisia tabaci* (Mondal *et al.*, 2020). Their puparium is mainly used for taxonomic identification. A pair of small compound pore is present in the seventh and eighth abdominal segment of the puparium (Selvaraj *et al.*, 2017). They lay their dark yellow to creamy whitish elliptical eggs on the underside of the leaves and covered the eggs with whitish filamentous wax like coatings. Egg was observed to be translucent yellowish green or light green to yellow, though according to Pradhan *et al.* (2020), the eggs changed its colour from translucent to pale yellow and before hatching to brown. They have four developmental instars before reaching the adulthood. First three instars are termed as nymph and they are continuous feeder. First instar nymphs are generally considered as crawlers as they are only mobile stage after hatching. They move randomly on the host surface and after finding suitable feeding site they fix themselves and start feeding. After molting crawler turns into immobile oval and flat instars and with the progression of life they become oval and more convex in shape. The golden yellow nymphs also produce whitish waxy filamentous cottony growth on themselves and surrounding area as well. Adult flies have light brown paired band present on their wings. Males possess long sword shaped pincer like structure at the abdominal end. Limited information regarding the biology of rugose spiraling whitefly is available, except for few studies in Florida and in India (Taravati and Mannion, 2014; Taravati *et al.*, 2018; Elango *et al.*, 2019). The research work regarding its life history on our target crops is negligible. The observations by Pradhan *et al*. (2020) in NBAIR, Bengaluru, India revealed that the fecundity, duration of nymphal instars and duration of total life cycle were 56.60± 5.04, 25.7± 1.66 and 42.9± 2.95 days, respectively on *Canna indica*, an ornamental crop.

Invasion in minor fruits

Several authors reported the invasion of rugose spiraling whitefly from different minor fruit crops at low to moderate level in different regions of the country. Observations from the survey in Karnataka by Selvaraj *et al*. (2017) suggested that different fruit crops including sapota, India almond, water apple, laurel ball tree or punnai were invaded by rugose spiraling fly and had severe (11-20%), very severe (>20%), moderate (6-10%) and very severe (>20%) infestation, respectively. In Kerala the pest invaded the crops like wild jack fruit (*Artocarpus hirsutus*), jack fruit, Malabar tamarind (*Garcinia gummi-gutta*) and Indian almond (Karthick *et al.,* 2018). Survey report in West Bengal stated that different minor fruit host community comprising

water apple, jamun, custard apple [Plate 1] and jack fruit [Plate 2] were infested by rugose spiraling whitefly (Selvaraj *et al.*, 2019a). In 2020, several minor fruit crops including sapota, custard apple, White wax jambu (*Syzygium samarangense*) and Indian almond was invaded by *A. rugioperculatus* in Junagadh district, Shaurastra region of Gujarat (Jethva *et al*., 2020a). Elango and Nelson (2020a) reported that custard apple recorded relatively higher number of eggs laid per female (26.9 eggs per spiral), highest male emergence (46.13%) and medium in female emergence (53.57%). The sex ratio (female: male) was 1: 0.86 on custard apple according to them. Sri *et al.* (2022) from Bhubaneswar reported that the minor fruit crop species comprising Bael, custard apple, jackfruit, jamun, phalsa, Ramphal (*Annona reticulata*), rose apple (*Syzygium malaccense*), sapota were infested by rugose spiraling whitefly with a per cent incidence of 32.50%, 65.30%, 51.30%, 100.00%, 5.60%, 93.20%, 85.50% and 91.30%, respectively, though in case of bael and phalsa only egg stage was found. They also recorded lowest number of nymphs of this pest (15.0 per leaf) on jack fruit and lowest number of pupae (12.0 per leaf) on sapota. Rajmohana and Sushama (2022) observed the concerned pest infestation in wild date palm (*Phoenix sylvestris*) from West Bengal. Nandhini and Srinivasan (2022) from their survey based study in Tamil Nadu observed that rugose spiraling whitefly survived up to nymphal stage in *Annona muricata, A. squamosa, A. reticulate* and *Theobroma cacao*. The other minor fruits they found as the hosts which supported egg stage only were bael, karanda (*Carissa carandas*), star apple, wood apple (*Feronia elephantum*), sapota, avocado (*Persea americana*), Indian gooseberry (*Phyllanthus emblica*), jamun, Sour sop (*Annona muricata*) and Spanish cherry (*Mimusops elengi*). Several fruit crops were infested by this pest in Bangladesh having almost similar kind of climatic condition like West Bengal and they are date palm, palmyra palm, jackfruit, deua or monkey jack (*Artocarpus lacucha*), hog plum or amra, cashew nut, water apple or jamrul, Indian black berry, custard apple, sharifa (*Annona squamosa*), pomelo or jambura (*Citrus maxima*), amlaki (*Phyllanthus emblica*), olive (*Olea europaea*), rambutan (*Nephelium lappaceum*), jujube (*Ziziphus jujube*), elephant apple or chalta and tamarind (*Tamarindus indica*) with relatively less infestation where less than 10% plants were infested with 10 or less nymph or adult per leaflet (Das *et al*., 2023).

Invasion in ornamental plants

Apart from the different major and minor fruit crops, some of the ornamental plants are also invaded by the rugose spiraling whitefly. Just after introducing in India the pest also infested butterfly palm (*Dypsis lutescens*) and ruffled fan palm (*Licuala grandis*) severely in Karnataka (Selvaraj *et al*., 2017). Sri *et al.* (2022) from Bhubaneshwar enlisted several ornamental plants invaded by the pest includes Acalypha (*Acalypha macrophylla, A. wilkesiana*), Asoka (*Saraca asoca*), beach spider lilly (*Hymenocallis littoralis*), bird of paradise (*Sterlitzia reginae*), butterfly palm, Chinese fan palm (*Livistonia chinensis*), garden croton (*Codiaeum variegatum*), Dracaena (*Dracaena marginata*), golden thryallis (*Galphimia glauca*), Ixora (*Ixora chinensis*), Jatropa (*Jatropa curcas*), Philodendron (*Thaumatophyllum bipinnatifidium*) and song of India (*Dracena reflexa*) with 68.2, 66.2, 53.5, 100.0, 100.0, 95.0, 70.0, 41.0, 10.5, 53.0, 19.0, 41.0, 65.0 and 11.5 per cent incidence, respectively. Among these, bird of paradise was recorded with highest number of nymphs, pupae and adults (38.5, 28.9 and 78.5 per leaf, respectively) and lowest number was noticed on golden thryallis (12.0, 5.0 and 16.3 per leaf, respectively). Das *et al*. (2023) recorded low infestation of this pest from Bangladesh in Florida royal palm (*Roystonea regia*). According to Mondal *et al.* (2020), potted palm and bird of paradise were severely infested by the pest in West Bengal.

Invasion in medicinal, aromatic and avenue plants

Important medicinal and aromatic plants were also invaded by this notorious pest. Among them two crops betel vine (*Piper betle*) and rubber fig (*Ficus elastic*) faced minor infestation in Karnataka (Selvaraj *et al*., 2017). Different avenue trees like brown salwood (*Acacia mangium*), portia tree (*Thespesia populnea*), wire weed (*Sida acuta*), Brahma's banyan (*Ficus exasperate*), Rangoon creeper (*Combretum indicum*), golden trumpet (*Allamanda cathartaca*) and oleander (*Nerium*

Plate 1: Incidence of rugose spiralling whitefly on custard apple

Plate 2: Incidence of rugose spiralling whitefly on jackfruit

oleander) were also reported to be attacked by this insect in Kerala (Karthik *et al*., 2018). Infestation of this whitefly was also recorded from West Bengal on areca nut (*Areca catechu*), betel vine, Spanish cherry, akashmoni (*Acacia auriculiformis*) and areca palm (*Dypsis lutescens*) (Selvaraj *et al.*, 2019b). From Gujarat there was a report of whitefly infestation in curry tree (*Murraya koenigii*) (Jethva *et al*., 2020). Bhavani *et al*. (2020) observed severe infestation on areca palm and Brahma's banyan, moderate infestation on crotons (*Helicona* sp.) and traces of infestation on teak (*Tectona grandis*) in Andhra Pradesh. Medicinal plants like Aloe (*Aloe vera*), neem (*Azadirachta indica*) and turmeric (*Curcuma longa*) with an infestation of 66.60%, 56.50% and 40.20%, respectively were also recorded at Bhubaneswar, Odisha (Sri *et al.*, 2022; Nandhini and Srinivasan, 2022). Sri *et al.* (2022) recorded the highest number of eggs (187.0 per leaf) on neem and lowest number of eggs (35.0 per leaf) in Aloe among medicinal plants and among avenue trees maximum number of nymphs (36.5 per leaf) was recorded on Indian almond while minimum number of nymphs (21.0 per leaf) was registered on Cassia. Indian almond also recorded highest number of pupae (26.0 per leaf) and adults (83.5 per leaf) whereas Cassia recorded least number of pupae (14.6 per leaf) and adults (32.0 per leaf). In Tamil Nadu, several other plants were invaded by this notorious pest like Alexandrian laurel (*Calophyllum inophyllum*), banyan (*Ficus benghalensis*), Fern leaf tree (*Filicium decipiens*), Gliricidia (*Gliricidia sepium*), Indian elm (*Holoptelea integrifolia*), Jatropha, mahua, Indian beech (*Milletia pinnata* L.), Philodendron (*Philodendron hederaceum var aureum)*, false Asoka tree (*Polyalthia longifolia*), Indian sandal wood (*Santalum album* L.), Asoka tree (*Saraca asoca*), mahogany (*Swietenia macrophylla*), peepal (*Ficus religiosa*) and teak (*Tectona grandis*). The entire host supported only egg stage except teak, which supported all the three stages viz. egg, nymph and adult (Nandhini and Srinivasan, 2022). A survey conducted by Mondal *et al.* (2020) in West Bengal revealed the pest's severe to medium infestation on rubber fig, deodar and Acacia leaves. Das *et al*. (2023) reported that croton and Ixora experienced higher whitefly infestation while bakul

(*Mimusops elengi*), banyan, debdaru (*Polyalthia longifolia*), akashmoni, mahogany and haritaki (*Terminalia chebula*) experienced less infestation in Bangladesh.

Population dynamics of rugose spiraling whitefly

Numbers of research works (Srinivasan *et al.,* 2016 on coconut; Sundararaj and Selvaraj, 2017 on coconut; Mohan *et al*., 2017 on coconut; Josephrajkumar *et al.,* 2018 on coconut; Mane, 2019 on coconut; Elango and Nelson, 2020b on coconut; Sushmitha *et al*., 2020a on coconut and guava; NBAIR, 2021; Chavan *et al*., 2022 on coconut) are available on the population dynamics of this pest and most of the studies were concentrated on coconut. Weather parameters have significant effect on the infestation of the whitefly. Reduced rainfall, prolonged dry spell, high temperature, low humidity, bright sunshine hours, availability of large area of host plants and absence of natural enemies are very much congenial for up-surging of its population according to them. Information regarding its population dynamics in other crops particularly minor fruits, medicinal and aromatic plants is scanty. Avitha *et al.* (2022) in their study on wild date palm in West Bengal supported the report of NBAIR (2021) who opined that severe infestation of this pest in West Bengal might be due to hot and humid climatic condition, the availability of number of host plants and the lack of natural enemies.

Possible management strategies

Taravati *et al.* (2013) and Francis *et al.* (2016) reported number of natural enemies associated with rugose spiralling whitefly such as parasitoids like *Encarsia guadeloupae, E. noyesi, Aleuroctonus* spp. and predators like *Nephaspis oculata, Azya orbigera orbigera, Chilocorus cacti, Cryptolaemus montrouzieri, Delphastus pallidus, Harmonia axyridis, Hyperaspis bigeminata, Cy-bocephalus* sp. and *Ceraeochrysa* spp. in Florida. Sel-varaj *et al.* (2016) documented three species of natural enemies from Tamil Nadu, Andhra Pradesh and Kerala e.g. *E. guadeloupae*, *Mallada* spp. and *Cybocephalus* spp. and among them *E. guade-loupae* was found as a dominant one with highest parasitism (20.0-60.0%). Survey by Selvaraj *et al.* (2017) in Karnataka revealed natural parasitism of rugose spiraling whitefly by the parasitoids *E. guadeloupae* and *E. dispersa* with percent parasitism of 5-15% in coconut, 7-18% in banana and 22-30% in sapota. Gupta *et al.* (2017) recorded *Xenasteia* members (Diptera: Brachycera: Cyclorrhapha) on rugose spiraling whitefly colony in the coastal areas of Karnataka. Sushmitha et al. (2020a) reported spiders and coccinellid beetles (*Menochilus sexmaculata*) as predators in coconut and guava. Natural enemies *viz*., lady bird beetles, *Cryptolaemus montrouzieri*, *Chilocorus nigrita*, *Scymnus nubilus* and the parasitoid wasp, *Encarsia guadelopae* were recorded in sugarcane ecosystem from Andhra Pradesh (Bhavani *et al*., 2020). Elango and Nelson (2020a) recorded the predators viz. *Cybocephalus* spp., *Cryptolaemus montrouzieri*, *Chilocorus nigrita*, *C. sexmaculata*, *Curinus coeruleus*, *Mallada astur*, *Mallada boninensis* and *C. zastrowi sillemi*, praying mantis, spiders and one parasitoid *E. guadeloupae* as natural enemies of *A. rugioperculatus* in Tamil Nadu. Several natural enemies including predators like *Cybocephalus* spp*., Chrysoperla* sp*., Nephaspis oculata, Oxyopes salticus* and *Uloborus* sp. and a parasitoid, *Encarsia* sp. were reported on this whitefly from Bhubaneswar (Sri *et al.,* 2022). Rajmohana and Sushama (2022) recorded *E. guadeloupae* as natural parasitoid and *Dichochrysa astour* as predator of rugose spiraling whitefly in *Phoenix sylvestris* from West Bengal though earlier survey conducted by Selvaraj *et al.* (2019a) revealed the absence of *E. guadelouape* from West Bengal and they noticed chrysopid, *Dichochrysa astour* as the sole natural enemy. Selvaraj *et al.* (2019b) observed that among the natural enemies, *E. guadeloupae* was dominant and potential to provide about 41% natural parasitism in Andhra Pradesh and it was less than 20% in Kerala and Karnataka. Drastic reduction in rugose spiralling whitefly was noticed by Visalakshi *et al.* (2021) in coconut orchards sprayed with *Isaria fumosorosea* fungus (NBAIR-Pfu-5) @ 5 g/ l with sticker @ 10 g/ l, two times with introductory release of parasitoid, *Encarsia guadeloupae*. The attempt made by Elango *et al.* (2022) revealed that under laboratory conditions, *Isaria fumosorosea*, a microbial pathogen with a spore load of 1×10^8 cfu caused 34.54% egg mortality, 37.39% nymphal mortality and 48.30% adult mortality of rugose spiraling whitefly,

followed by *Lecanicillium lecanii* (24.54, 30.76 and 28.01%) and *Metarhizium anisopliae* (20.56, 32.51 and 42.92%) for egg, nymphal and adult mortality, respectively in coconut under laboratory condition, whereas, under field conditions, *I. fumosorosea* caused nymphal mortality of 29.60%, followed by *M. anisopliae* (24.30%) and *B. bassiana* (21.00%) at 15 days after spray. This study also proved the safety of *I. fumosorosea* to the chrysopid predator, *Mallada boninensis.* So, there is a huge scope for utilizing natural enemies specially *Encarsia guadeloupae*, *Isaria fumosorosea* and *Mallada boninensis* in suppressing this notorious pest. The incidence and intensity of the whitefly was significantly reduced from 75.5 to 37.7% and 85.7 to 42.9%, respectively on coconut treated with ecofriendly IPM practices in Tamil Nadu (Alagar *et al.*, 2021). The IPM treatments included installation of light traps @ 5/ ha, fixing yellow sticky trap sheets @ 25/ ha, spraying three rounds of 0.5% neem oil at 15 days interval on the under surface of leaves, three rounds of jet water spray at 10 days interval about 15 days after spraying of neem oil and stapling of leaflets containing*, E. guadeloupae* parasitised puparia on palm leaflets. According to Saranya *et al.* (2022), antibiotic treatment, carbenicillin 100 ìg/ mL + ciprofloxazin 5 ìg/ mL significantly reduced the oviposition and % egg hatchability of rugose spiraling whiteflies reared in coco-nut (13 eggs/ spiral and 61.54%), banana (15 eggs/ spiral and 60.00%), sapota (15 eggs/ spiral and 66.67%) and guava (16 eggs/ spiral and 56.25%). The experiment conducted by Navin Kumar *et al.* (2022) suggests that Neemazal- T/S @ 10000 ppm concentration could effectively kill the early and late instars nymphs of *A. rugioperculatus* in coconut, mango, banana and guava. Trunk application including basal bark sprays and trunk injection with systemic insecticides *viz.,* Acetamiprid, Clothianidin, Dinotefuran and Imidacloprid can be recommended in coconut (Mannion, 2010). A substantial control of this pest on coconut plantation in West Bengal was achieved by spraying with Flonicamid 50% WDG and Lancer Gold (Acephate 50% +Imidacloprid 1.8% SP) @ 5 g and 2 g/ l respectively and root-feeding with Monocrotophos 36 SL @ 4 ml/ plant as immediate remedy measure (Mondal *et al.*, 2020).

CONCLUSION

From the earlier research works and surveys it is concluded that rugose spiralling whitefly has become a serious threat invading several hosts such as plantation crops, fruit crops, medicinal plants, spices, ornamentals, avenue trees, vegetables and field crops belonging to different plant families, though coconut, banana and guava were proved to be preferred host for this pest. Indiscriminate use of pesticides to control this pest in its preferred host plants like coconut, banana or guava may lead to population shifting on other host plants like minor fruits, medicinal, aromatic or plantation crops of our concern. Though, climate and host availability play significant roles in determining its population fluctuation, accidental dispersal may also play a crucial role in its new outbreaks. Inoculative releases of natural enemies identified in the early stages of whitefly establishment may be beneficial to manage the pest. Therefore, suitable devices of integrated pest management should be advocated against the pest with special emphasis on utilization of biocontrol agents, microbial pathogens and botanicals.

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Review article

Nutritional and therapeutic attributes of neglected and underutilized fruit crops in Sri Lanka and their potential applications in value addition

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ABSTRACT

Sri Lanka endowed a rich biodiversity, making it a significant source of plant species used as food sources. Fruits serving as both desserts and refreshments. Major and minor fruits contribute vitamins to the Sri Lankan diet, but underutilized fruit crops, lacking developed cultivation protocols and commercial production, remained as unexplored resources. Soursop (Annona muricata), Lavalu (Pouteria campechiana), Madam (Syzigium cumini), Nam-nan (Cynometra cauliflora), Mangosteen (Garcinia mangosteen), Beli (Aegle marmelos), Kamarankha (Averrhoa carambola), Jambu (Syzygium jambos), Sapodilla (Achras sapata), Veralu (Elaeocarpus serratus) are some underutilized fruit crops available in Sri Lanka. This review article highlights the nutritional, medicinal, and value-added potential of selected minor fruits in Sri Lanka.

***Keywords*:** Medicinal properties, nutritional value, underutilized fruits, value addition

INTRODUCTION

Sri Lanka is one of the biologically diverse hotspot in Asia, serving a home to more than 100 species of fruits. There are numerous underutilized wild fruit species that are used by the locals to suit their dietary needs (Tripathi, 2021). However, only a limited number of species are considered major fruits, including Banana, Mango, Pineapple, Passion Fruits and Avocado, which are cultivated on a commercial scale (Premathilake and Wathugala, 2013). Neglected or underutilized fruit crops are domesticated plant species that have been historically valued for their food or medicinal properties (Dahanayake, 2015). Neglected and underutilized agricultural species hold an enormous promise for combating malnutrition, poverty, and global hidden hunger (Ali and Bhattacharjee, 2023). Indeed, underutilized fruits are not only a source of food, vitamins, and minerals, but also a source of income due to their nutritional and medicinal capabilities. Fruits are high in vitamins and minerals. Aside from their nutritional worth, some fruits offer therapeutic properties due to presence of flavonoids, quinolizidine, alkaloids, stilbenes, tannins, steroids, coumarin, and saponins, quinolizidine, triterpenoids, glycosides, and fatty acids. They are having a variety of pharmacological effects, including anti-inflammatory, analgesic, antidiabetic, antipyretic, antioxidant, hypoglycemic, hepatoprotective, and anticancer, dysentery, cholera, wounds, and sores (Tripathi, 2021). This review will present the nutritional and medicinal properties of selected minor crops in Sri Lanka, along with their potential for value-added products.

*Beli (*Aegle marmelos*), Woodapple* (Ferronia Limonia), *Mangosteen* (Garcinia mangosteen), *Nelli* (Embelica officinalis), *Sapodilla* (Achras sapata), *Star fruit* (Averrhoa carambola), *Lovi* (Flacourtial inermis), *Amberalla* (Spondias dulcis), *Ambul Dodam* (Citrus aurantium), *Heen Naran* (Citrus crenatifolia), *Pani Dodam* (Citrus sinensis (L.), *Veralu* (Elaeocarpus serratus), *Ugurassa* (Flacourtia indica), *Katu anodha* (Annona muricata), *Rose apple* (Syzygium jambos), *Bilin* (Averrhoa bilimbi), *Tamarind* (Tamarindus indica), *Damina* (Limonia acidissima), *Madan* (Syzygium caryophyllatum), *Damba* (Syzygium rotundifolium), *Kon* (Schleichera oleosa), *Karanda* (Carissa carandas), *Nam-nan (*Cynometra cauliflora), *Jambola* (Syzygium cumini) are some important underutilized fruit species available in Sri Lanka (Fig. 1).

These species lack developed scientific agronomic packages, organized supply and marketing channels, processing techniques as well as export share (Premathilake and Wathugala, 2013). Over time, significance of these fruit species has diminished due to factors such as limited supply, constraints like poor shelf life, insufficient consumer awareness, and their nutritional and medicinal value going unrecognized (Dahanayake, 2015). One of a major reasons for their limited popularity include the lack of knowledge regarding their nutritional and medicinal value. Inadequate scientific research, particularly on their potential for medical applications, has also contributed to their lack of attraction. Furthermore, insufficient value addition as well as rising demand for imported fruits contributed to this decline in popularity (Premathilake and Wathugala, 2013).

"Value addition" would be the greatest approach not only to ensure year-round availability of underutilized fruit species but also enhanced their popularity and will help to eliminate the surplus waste. Exporting processed and semi-processed products ensures profits and economic benefits. Most important thing is the prioritizing in-depth researches on nutritional quality and anti-nutritional elements, processing, value addition, product development, and successful marketing methods. Additionally, close linkages between growers, merchants, processors, and consumers are needed to be developed to have an efficient value chain and stimulate the use of these potentially neglected crops (Ali & Bhattacharjee, 2023).

In order to overcome the major constraints regarding underutilized crops in Sri Lanka, a well-organized surveys must be launched to build a database on the origin, distribution, habitat, agroclimatic requirements, and scientific application of potential underutilized crops. Then, suitable plant types with desirable characteristics should be generated. Further introducing underutilized crops into existing agricultural systems can be also done.

Important underutilized fruit species in Sri Lanka

Soursop (*Annona muricata*)

Soursop belongs to the Annonceae family and is commonly known to Soursop in English, locally referred to as *"Katu anoda"* in Sri Lanka. It is a climactric fruit with a short shelf life (Badrie and Schauss, 2010). The tree is an evergreen slender, reaches a height of around 8-10 meters. The tree produces dark green oval to conical/heart shaped fruits in the unripen stage, which gradually turn light green as they ripen (Sanusi and Abu Bakar, 2018). Typically, many fruitlets fuse together to form a single soursop fruit. The fruit has whitish flesh that is juicy, with dark brown seeds and the number depends on the size of the fruit. Outer skin has a leathery appearance with short spines. The average weight of a fruit is around 4 kg. The primary propagule is the seed which can be stored for several months before planting. However, vegetative propagation have not been tried in Sri Lanka or with low percentage of success.

The well ripe (WR) stage fruit contains a significant amount of moisture (80.52% w/w), crude fat content (3.28% w/w), crude protein content (2.98% w/w), Brix value (11.5 degrees Brix), titratable acidity (1.02% w/w), pH (3.7), and a low amount of ash content (78% w/w). (Wijerama *et al.,* 2023). Additionally, soursop is rich in Vitamins B_1, B_2, and Ascorbic acid (Badrie and Schauss, 2010). Antioxidant compounds such as ascorbic acid, β-carotene, α-carotene, and different xanthophylls have been detected in soursop and may have contributed to the antioxidant activity of the fruit extract (Wijerama *et al.,* 2023). *A. muritica* is often referred to as a miracle fruit and is widely used in traditional medicine around the world to treat various conditions such as abscesses, bronchitis, diabetics, intestinal colic, palpitations, gallbladder problems, diarrhoea, dysentery, inflammation, liver disorders, fungal infections, asthma, blood cleansing, cancer, arthritis, malaria, intestinal parasites etc (Sejal and Jayvadan, 2016).

Lavalu (*Pouteria campechiana*)

Lavalu belongs to the Sapotaceae family and is one of the underutilized fruits of Sri Lanka, while it is commonly referred to as Canistel in most parts of the world (Lim, 2013). It is an evergreen tree that can be found in tropical and sub-tropical areas around the world and it is well adapted to a wide range of soil conditions. *P. campechiana* is an erect medium-sized tree that reaches a height of 8-20 m. Every part of the tree having whitish gummy latex. The fruit is a berry with obovoid to variable shapes,

measuring 7-12.5 cm in length and 4-7.5 cm in width and it has a golden yellow coloured skin (Lim, 2013).

The edible part of the plant is the fruit pulp, which resembles the texture of a hard-boiled egg yolk, giving rise to the name "Egg Fruit" (Lim, 2013 and Sethuraman *et al.,* 2020). Sethuraman *et al.*(2020) indicated that the fruit pulp of *P. campechiana* contains 52.96% moisture, 40.19% carbohydrate, 1.16% protein, 4.97% fat, 2.12% fiber, and 0.71% ash content. Ripe fruits also contain 2.7% fructose, 3.3% glucose, 15.6% sucrose and 6mg/100g of vitamin C. Due to its lower sugar content, it is potentially suitable for the people with obesity and type II diabetics (Sethuraman *et al.*, 2020). Moreover, *Pouteria campechiana* has many medicinal uses, such as preventing heart failures, cardiac arrhythmia, intestinal disorders, allergies and cancers (Mehraj *et al.*, 2015).

Madam (*Syzigium cumini*)

Syzigium cumini, locally known as "Madam" is an evergreen drought tolerant tropical tree belongs to the family Myrtaceae. It is commonly referred to as "Jamun" around the world (Madani *et al.*, 2021). Other synonyms for this crop include Portuguese plum, black plum, Malabar plum (Rehaman, 2021). The tree can grow up to a height of 30 meters with a girth of 3-4 m (Jadhav *et al*., 2009). In Sri Lanka, these trees are mostly found in rural forest areas as wild trees bearing purple coloured berries that mainly serve as a food source for birds and mammals. The fruit of *S. cumini* is a berry with deep purple coloured fleshy pulp. The pulp is astringent and sour in taste, and it contains a hard seed embedded within it (Benherlal and Arumughan, 2007). The fruit is perishable and typically have a short postharvest shelf life at room temperature. Fresh fruit pulp is rich in carbohydrates, proteins and minerals. It contains 80% moisture, 0.81% protein, 12.7% Sugar (Swami *et al.*, 2012). The fruit is an excellent source of antioxidants, iron, and vitamin C (Rizvi *et al.*, 2022).

S. cumini plays a significant role in primary healthcare and traditional systems such as Ayurveda, Unani and Sidha. It is used as a treatment for diabetes mellitus, ulcers, inflammations, stomach pains, chronic diarrhea (Jadhav *et al.,* 2009 and Swami *et al.*, 2012). Additionally, it possesses antimicrobial, anti-inflammatory, anticancer, gastroprotective, anti-viral activities and can be used to boost up immune system (Rehaman, 2021).

Nam-nan (*Cynometra cauliflora*)

Nam-nan is a member of family Fabaceae and one potential underutilized fruit crop in Sri Lanka. It is a widely spread evergreen small tree that can grow up to a height of 15m. This tree typically bears greenish yellow or brown kidney-shaped pods (fruits) on its trunk, which have a rough surface with wrinkles (Tajudin *et al.*, 2012). Nam-nan can be found in village areas as a homegrown tree and it produces fruits with a savory taste that can be consumed either as a raw fruit or as a salad (Khoo *et al.*, 2016). It is rich in vitamin A and vitamin C (Duraisamy *et al*., 2020). According to Lim (2012), the edible fruits are used in traditional medicine to treat skin diseases and loss of appetite.

Mangosteen (*Garcinia mangosteen*)

Mangosteen belongs to Clusiaceae family (Ketsa and Paull, 2011). It is locally known as "*Mangus*" in Sri Lanka. Mangosteen plants can be found in Sri Lanka as garden plants or small cultivations in both the mid and wet low-country districts. This plant has a slow growth habit and a long juvenile stage of 8-12 years to develop fruits, thus seedlings should be kept at the nursery for at least two years. It is considered to be one of the most popular tropical fruits with a strong economic potential (Wickramasingha *et al.*, 2021).

Pedraza-Chaverri *et al*. (2008) described mangosteen as an evergreen tropical tree that exhibits very slow growth in an erect habit, reaching heights of 6-25m. The fruit is externally smooth and round shaped, with a dark purple to reddish purple in colour. Inside the fruit, there are 4 to 8 triangular, white juicy arils, that form the edible portion of the fruit. The flesh has slightly to distinctly acid flavour. Rizaldy *et al.* (2022) stated that edible portion of 100g contains 80.94 g of moisture, 0.41 g of protein, 0.58 g of fat, 17.94 g of carbohydrates, minerals 73 mg (calcium, phosphorous, potassium) and ascorbic acid (2.9 mg). Aril has pleasant taste but contain low nutrient content (Ketsa and Paull, 2011). They also reported minor amount of vitamin B_1, B_2, niacin and vitamin C in the edible portion.

Rizaldy *et al.* (2022) stated in their review that the main abundant bioactive compound in mangosteen is xanthone. Kurniawati *et al*. (2014) suggested that the xanthone found in the rind of mangosteen has antidiabetic effect. Furthermore, the pericarp is utilized for various medicinal purposes such as overcoming abdominal pain, dysentery, eczema, cystitis, chronic ulcers and diarrhea (Al-Massarani *et al.*, 2013). Mangosteen has been found to exhibit a range of pharmacological effects including anti-diabetic, anti-obesity, antimalarial, anti-parasitic, antioxidant, anti-acne, anti -aging, anti-bacterial and anti-inflammatory properties (Rizaldy *et al.*, 2022).

Beli (*Aegle marmelos*)

'Beli' or 'Bael' belongs to the Family Rutaceae. The fruit is native to Southeast Asia (Jayawardene, 2019) and it can be found in most parts of home gardens in Sri Lanka. Bael is an arid fruit crop with high nutritional content, hardiness, high processing quality, and extensive adaptability across the tropics and subtropics (Kundu & Ghosh, 2017). Bael trees are commonly observed in Hindu temple gardens in Sri Lanka. The tree can grow up to 18 meters in height and bears fruits with a smooth woody skin that measures 5-15 cm in diameter. It is regarded as a hardy tree that can flourish in both marshy alkaline soil and strong acid soil with pH ranging from 5 to 10 (Kundu & Ghosh, 2017).

Inside the fruit, a thick, glue-like aromatic pulp, which is yellow and fibrous, contains embedded seeds. The taste resembles marmalade. The Bael fruit is rich in Vitamins B_1, B_2, and Proteins (Jayawardena, 2013). Maity *et al.*(2009) stated that bael may play a vital role in future medicine systems. Bael is used as a treatment for cancer, and cardiovascular diseases and has many medicinal properties, including being astringent, antidiarrheal, antipyretic and anti-inflammatory property (Maity *et al.,* 2009). Bael fruit also shows numerous potential health benefits, such as radio-protective effects, antibacterial, gastroprotective, antiviral, antidiabetic, anti-ulcerative colitis, as well as cardioprotective effects (Venthodika *et al.*, 2020). Fruit drink of bael is an excellent "restorer of vigor" for those engaged in outdoor activities (Jayawardene, 2019). The fruit shell can be boiled in water and consumed to provide a cooling effect for the body.

Kamarankha or Star fruit (*Averrhoa carambola*)

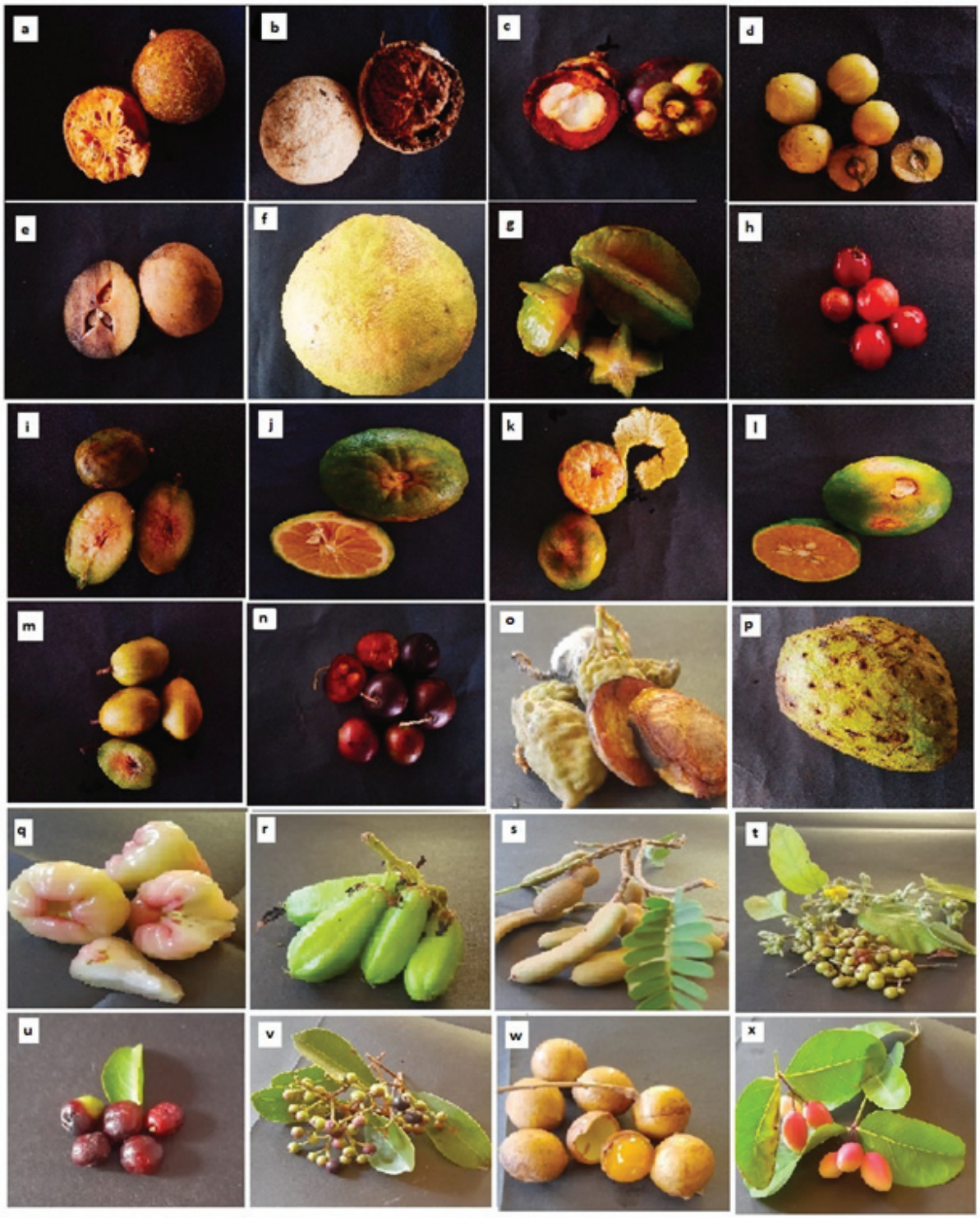
Kamarankha, are called as "Star Fruit" in English and is one important underutilized fruit in Sri Lanka belongs to the family Oxalidaceae (Dasgupta *et al*., 2013). Star fruit originated from Ceylon and the Moluccas. It is a small tree that grows up to 6 m in height, mainly in tropical and sub-tropical areas (Manda *et al*., 2012). It bears green to greenish-yellow, fleshy fruits that are about 6 cm long. The fruits are distinct with five longitudinal angular lobes The fruits are green at the unripe stage and turn yellow or orange when ripe (Dasgupta *et al* ., 2013). The fruit has a slightly tart, sweet and acidic taste (Muthu *et al.*, 2016).

In their review, Manda *et al.* (2012) cited out that star fruits contain a package of nutrients. In 100g of fruit, there is 38g of protein, 9.38g of carbohydrates, 0.9g of fiber, 0.08g of fat and Ascorbic acid (15.4/100ml). Patil *et al*. (2010) conducted an analysis of star fruit at various stages of ripening and found that fully ripe fruit contains 0.85% protein, 1.32% reducing sugar, 2.25% total sugar, 18% ascorbic acid, 5.11% pectin and 0.17% amino acids. They also noted that the chemical composition of edible star fruit differs with the maturity stage.

Star fruits can be eaten as raw fruits and can also be used in the preparation of juices, salads, jams, wine and pickles (Patil *et al.*, 2010 and Muthu *et al*., 2016). Lakmal *et al*. (2021) listed several beneficial medicinal effects of star fruit including anti-inflammatory, anti-infective, antitumor, antioxidant, hypotensive, hypocholesterolemic and immune boosting effects. They also mentioned that star fruits are commonly used in Ayurvedic medicine to treat skin disorders, fever, cough, diarrhea, chronic headache etc.

Jambu or Rose apple (*Syzygium jambos)*

Jambu is known as Rose Apple belongs to the family Myrtaceae (Subbulakshmi *et al*., 2021). It has a dense crown with wide spreading branches. The fruit is a berry and has a fleshy pericarp that is 10-15mm thick. The epicarp of the fruit is reddish and thin. The fruit has succulent whitish flesh (Ochieng *et al.*, 2022). Subbulakshmi *et al.,* (2021) stated that the fruits are sometimes yellowish in colour, except for the Malaysian variety, which is reddish slightly pear shaped and crowned by a 4



a. Beli - *Aegle marmelos,* **b.** Woodapple – *Ferronia limonia,* **c**. Mangosteen - *Garcinia mangosteen,* **d.** Nelli – *Embelica officinalis*, **e.** Sapodilla - *Achras sapata,* **f.** Jambola - *Syzygium cumini,* **g.** Star fruit - *Averrhoa carambola,* **h.** Lovi – *Flacourtial inermis,* **i.** Amberalla- *Spondias dulcis,* **j.** Ambul Dodam- *Citrus aurantium,***k.** Heen Naran- *Citrus crenatifolia* , **l**. Pani Dodam -*Citrus sinensis (L.)* , **m**. Veralu – *Elaeocarpus serratus,* **n.**Ugurassa – *Flacourtia indica***,o.** Nam-nan - *Cynometracauliflora,* **p**. Katu anodha –*Annona muricata,* **q**. Rose apple – *Zyzygium jambos,* **r.** Bilin – *Averrhoa bilimbi,***s.** Tamarind – *Tamarindus indica,* **t.** Damina- *Limonia acidissima* , **u.** Madan-*Syzygium caryophyllatum* , **v.** Damba – *Syzygium rotundifolium,* **w.** Kon- *Schleichera oleosa,* **x**. Karanda - *Carissa carandas*,

Table 1: Processed products that can be prepared from the selected minor fruits

Minor Fruit	Value Added Products	Sources
Soursop	Juices, juice blends, fresh pulp bottled, frozen pulp, concentrates, nectar, ice cream, sherbets, syrups, jams, jellies, yoghurt, powder, dehydrated products, candies, canning, flakes, fruit bars, champola	Badrie and Schauss, 2010; Sanusi and Abu Bakar, 2018
Lavalu	Custards, ice cream, milkshakes/eggfruit nog, pancakes, cupcakes, jam, marmalade, pie butter	Lim, (2013)Sethuraman *et al.* (2020)
Madam	Juices, seed powders, jellies, jam, squash, hips, wine, vinegar, pickles, syrups, sauces, brandy	Rehaman, 2021; Babak Madani *et al.* (2021)**;** Swami *et al.* **(**2012)
Nam-nan	Jam, pickles, salads, fried products	Duraisamy *et al*. (2020)Lim, (2012)
Mangosteen	Paste, leather, candy, juice, dried arils, pericarp powder, freeze dried mangosteen, wine, vinegar, ice cream, yoghurt, cosmetics	Ketsa and Paull, (2011)
Beli	Juices-RTS, jam, toffee, preserve, panjiri, squash, dehydrated products, Powder, Slab	Singh and Chaurasiya, (2014) Ullikashi *et al*. (2017)
Starfruits	Puddings, tarts, stews, pickles, jam, preserves, canned syrups, carambola juice, sherbets, jelly, salads, squash	Manda *et al*. 2012 ; Patil *et al*. (2010); Anuar and Salleh, (2019); Monalisa *et al.* (2014)
Rose Apple	Wine, jam, jelly, syrups, sauce, drinks, candy, rosewater	Subbulakshmi *et al*. (2021)
Sapodilla	Fruit bars, nectar, puree, juice, jams, butter, powder, candy, dried slices, halwa, milkshakes, Shrikhand	Madani *et al*. (2018); Jadhav *et al*. (2018)
Veralu	Juice, smoothies, ice cream, jelly pickles	Jayawardane, (2019)

lobed calyx. It is a non-climacteric tropical fruit with a rose fragrance, which is due to the presence of linalool (Mondal *et al.*, 2023). The fruit possess vitamins, fat, minerals and antioxidants as nutrients. Subbulakshmi *et al.* (2021) found that 100g of the fruit contains 84-93g of water, 0.6g of protein, 0.3g of total lipids, minute amounts of minerals (Ca, Mg, Fe, Zn, K etc.), 22.3mg of **v**itamin C, and minutes amount of Vitamin B_1, B_2 and A. Scientific studies have found that rose apple has uses in traditional medicine, such as antimicrobial, anti-inflammatory, analgesic, antiviral, anticancer, and hepatoprotective activities, while the seeds are used in treatments for diabetes, diarrhoea and dysentery (Subbulakshmi *et al*., 2021).

Sapodilla or Sapota (*Achras sapata*)

Sapota belongs to the Sapotaceae family (Bano and Ahmed, 2017 and Kulkarni *et al*., 2007) and is locally known as Sapodilla in Sri Lanka. It is a medium to large sized tree that reach up to 20-30 meters bears brown coloured fruit that measures 5-10 cm in width, course texture**d** when unripe, which gradually turns soft as it ripens (Bano and Ahmed, 2017). Sapota has an average moisture content of 75.2% , an average fruit weight of 52.19g, a brix value of 19.45, an average acidity of 0.16 , 0.5% protein, 19.5% carbohydrates, 0.49% fat,2.5% fiber and average total sugar content of 48.5% with reducing sugar accounting for 16.3% (Jadhav *et al*., 2018).

A decoction of young fruits help relieve diarrhea, while an infusion of young fruits and flowers together is used to treat pulmonary complaints. Additionally, seeds have a diuretic action that helps expel stones from the kidney and bladder (Kulkarni *et al*., 2007). A paste of seeds is known to be used as an application for the stings and bites of venomous animals. Sapodilla has anti-inflammatory, anti-arthritis, anti-fungal, anti-oxidant, anti-tumor anti-diabetic activities. Ripe fruits are used to treat diarrhoea, dysentery, haemorrhage, pulmonary problems, muscle spasms and breast cancers (Bano and Ahmed, 2017)

Veralu or Ceylon olive (*Elaeocarpus serratus*)

Ceylon olive belongs to the family Elaeocarpaceae which is known as Veralu in Sri Lanka (Ananda *et al.*, 2023). It is a medium sized tree with drupe shaped fruits which have an astringent taste (de Lima *et al.*, 2019). The results of physical, chemical and pharmacological study carried out by them showed that average total weight of 19.45 g where pulp is about 82.16% of it and seed represent 17.89%. 100g of edible portion have 84.62g of moisture content, 6.1 g of ash, 3.15 g of total sugar, 1.1g lipid 4.92g of protein, 17.5 g of crude protein and 5.93mg of vitamin C (de Lima *et al.*, 2019). *Elaeocarpus serratus* can be utilized in traditional systems of medicine to treat arthritis, diabetic, analgesic activities, diarrhea. Further olive plants possess significant effect on melanogenesis inhibition and antioxidant properties, antibacterial activity and antifungal activity (Maheshwari *et al*., 2022).

Value addition of underutilized fruits

Most of the time, the surplus of these underutilized and neglected fruits are subjected to loss, as they are not harvested and instead left under trees or become a food source for wild animals, without being optimally utilized. On the other hand, many of these minor fruits available only for a certain period of the year, due to their seasonal bearing habit. Furthermore, these fruits are not very popular among the native population due to the lack of awareness about their nutritional and medicinal traits, as well as lack of continuous supply. To address these issues related to underutilized fruits, "value addition of minor fruits" emerges as an excellent option. The approach can potentially fulfill the off-season nutritional requirements of the local population by producing value added products that are available throughout the year, even during the off-season. This strategy can help to reduce post-harvest losses and increase the shelf life of these specific fruits. Despite some limitations, such as concerns about product quality, potential health issues, and nutrient loss during processing, the concept of "Value addition" can facilitate consistent supply against the demand. Moreover, it has the potential to enhance foreign exchange revenues and uplift livelihoods. Various value-added products that can be processed from selected minor crops in Sri Lanka has been presented in Table 1.

CONCLUSION

This review provides insights into selected underutilized fruit crops in Sri Lanka, focusing on their nutritional and therapeutic attributes and potential applications in value addition. In conclusion, these fruit crops have immense potential to gain popularity among the local community, primarily due to their nutritional and therapeutic qualities. To ensure year-round supply and demand, there is a need for value addition and commercialization of these fruits. Industrial workshops, promotional campaigns, and awareness programs involving both the government and private sector are essential to promote the local consumption of these minor fruits. Furthermore, additional studies are required to explore the nutritional and therapeutic properties of these fruits and to develop products through value addition.

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Review article

Essential oils uses in post-harvest management of fruits and vegetables : A Review

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ABSTRACT

Fresh fruit and vegetable consumption is steadily rising because of their nutritional value. It is estimated that about 30 to 40 percent of fruits and vegetables spoiled after harvesting due to poor post harvest management. Presently, this sector is facing lots of challenges in storage and supply chain of fresh fruits and vegetables to the consumers. The majority of post-harvest losses of horticultural produce are managed by using synthetic chemicals that may have detrimental effect to human health and nutritional value of horticultural produce. Therefore, using natural resources to control its damage and post-harvest management has been emphasized in order to prevent the decay of fresh fruits and vegetables. One of the natural sources and alternatives to the synthetic chemicals for reducing post-harvest losses and to improve overall quality and extending the shelf-life of perishable goods through natural essential oils and its constituents. Essential oils are used as preservatives, fragrant & flavour, vapour and coating wax as post-harvest management of fresh fruits and vegetables. The post-harvest losses of fruits and vegetables can be reduced by using essential oils of various aromatic crops such as cinnamon, geranium, citronella, mint, eucalyptus, lavender, thyme, rosemary, citrus, lemongrass, and basil, among the others. The objective of this review is to gather and discuss up-to-date applications and effects of the essential oils in post harvest management.

***Keywords*:** Essential oil, horticultural produce, natural resources, post-harvest management, shelf-life,

INTRODUCTION

India is the world's largest producer and consumer of fruits and vegetables, producing approximately 12.40 and 13.30 percent of global production, respectively (Gajanana *et al.,* 2011). Fruits and vegetables are the richest sources of vitamins and minerals, both of which are essential in human diets. The recommended dose of fruits and vegetables are 120g and 300g, respectively per capita per day, but availability of fruits and vegetables are still around 104g fruits and 207g, respectably per day per person (Gajanana *et al.,* 2011). To prevent the various diseases and to provide nutrients for many people who are low in micronutrients, an average of 400 g of fruits and vegetables per person per day are required (World Health Organization 2003). It is estimated that 30 to 50% losses of fruits and vegetables occurred world-wide, which is the main reason for low availability to the people (Gajanana *et al.,* 2011). During post-harvest handling, about 20 to 30 percent of fresh fruits and vegetables are also damaged. Post-harvest losses of various fruits and vegetables are brought through fungi, such as brown rot and rhizopus rot that *Rhizopus stolonifer* and *Monilinia fructicola* induce in peach (Sholberg and Gaunce, 1996; El-Sheikh Ali and Baraka, 2000). Nectarine fruits exposed to *Botrytis cinerea* induced grey mould, Rhizopus rot caused by *Rhizopus stolonifer*, and *Penicillium expansum* generated blue mould (Zhang *et al.,* 2008). Fungicides are a first line of defense against post-harvest losses of fruits and vegetables. The majority of synthetic chemicals used to minimize post-harvest losses in fruits and vegetables which are bad for consumer safety, hence, the uses of natural compounds and natural goods should be prioritized (Alzoreky and Nakahara 2003). One of the healthiest and safest ways to prevent post-harvest infections is to utilize essential oils extracted from plant sources. Essential

oils include a variety of secondary metabolites, most of which have antibacterial, fungicidal, antioxidant, and bio-regulating activities (Asghari *et al.,* 2009). Utilizing essential oils offers an alternative strategy for preventing fruit and vegetable storage losses and post-harvest pathogen growth.

Reason of spoilage of horticultural produce

In horticulture production, there is a major problem to save the produce from various damages such as physiological disorders, insects, pests and diseases attack because maximum loss of these produce occurred by these factors at various stages from field to final consumption. Most of the spoilage of the produce occurs by fungal, bacteria, viruses and insects. Post-harvest diseases affect the production, mainly because of the lack of proper post-harvest storage facilities.

After harvesting of crops, storage should be proper because most of the fruits and vegetables have very short shelf-life and some of those started spoilage in between one to two days. Important pathogens responsible for damage of the horticultural produce are given in Table 1.

Essential oils and its properties

Aromatic plants are the richest sources of secondary metabolites which have a wide range of properties like antioxidant, analgesic, anti-inflammatory, antitumor activity, larvicidal, anticancer, antimicrobial, anti-parasitic, insecticidal, anti-fungal and antibacterial properties (Saranraj *et al*., 2017; Srivastava *et al.,* 2014). The volatile compounds of the essential oils have a wide range of properties such as eugenol- local aneasthetic, neuro- protective, anti-cancer (Pramod *et al.,* 2010), Linalool- Anti-hyperalgesic, anti-nociceptive (Kim *et al.,*2015), Limonene- anti-inflammatory, motor-relaxant (Vale *et al.,* 2002), 1,8-cineole- monoterpenoid wound healing, antiulcer (Rocha Caldas *et al.,* 2015), Menthol- Anaesthetic (Balakrishnan *et al.*, 2018), Camphor- Anti-pruritic, decongesting (Zuccarini, 2009), Chicoric Acid- Immuno-stimulatory, antioxidant (Kuban-Jankowska *et al.,* 2016), Farnesene- Anti-insecticidal, antioxidant (Sun *et al.,* 2011), β-cadinene- Anti-proliferative (Cascaes *et al.*, 2015), Menthyl acetate- Antioxidant, Antibacterial (Singh *et al.,* 2015), Germacrene- Anti-inflammatory, analgesic (Del-Vechio-Vieira *et al.,* 2009), Silv Erio *et al.,* 2013), α-bisabolol- Analgesic, Antibiotic, Anticancer (Queiroz and Cajaiba 2015; Kamatou and Viljoen 2010).

Skrinjar and Nemet (2009) reported antibacterial properties of various essential oils like rosemary, turmeric, clove, pepper, fennel, round cardamom, angelica root, etc. against *Escherichia coli*, *Listeria monocytogenes* and *Pseudomonas fluorescens* which are prevalent as spoilage pathogenic bacteria. The anti-pathogen properties of essential oil are shown in Table 2.

Uses of essential oils in post harvest management

Essential oils have been used in management of diseases and pests, minimizing post-harvest losses, the extension of shelf-life, and the addition of flavour & fragrance and nutritional value to foods. The essential oil from Mints, Lemongrass, Thyme, Rosemary, Lavender, Bergamot, Dill, and Coriander, etc. have the efficacy for enhancing the shelf-life and also maintaining the quality of fruits and vegetables (Serrano *et al.,* 2005).

Essential oils as enhancement of self-life

According to studies of Abd-AllA *et al.* (2011), lemongrass oil has antifungal, antibacterial, antioxidant, antiviral, and antimicrobial properties. It also reduces the incidence of diseases, weight loss and firmness loss of fruits. It also enhanced acceptable texture and has a higher acceptance rate overall when avocado fruits are stored in cold storage under market shelf conditions (Mpho *et al.,* 2013). Rosemary (*Rosmarinus officinalis* L.) oil is utilized as an antioxidant and has antibacterial effects (Almela *et al.,* 2006). Against several bacterial and fungal strains, the oils of lavender (*Lavandula hybrida*), dill (*Anethum graveolens* L.), and coriander (*Coriandrum sativum* L.) demonstrated strong antibacterial and antifungal action. During the six weeks in cold storage of Crimson seedless grape, bergamot (*Citrus bergamia*) oil used for delayed the changes in total soluble solids, anthocyanin content, firmness, vitamin C, respiration rate, titratable acidity, and also reduced decay and weight losses (Abd El wahab *et al.,* 2014). It also had positive effects on titrable acidity, total soluble solids, weight loss

Table 1: Food borne Pathogenic microorganism

S.N.	Pathogen	Associated foods	References
1.	*Escherichia coli*	Salads, Green beans, Apple pieces, Spinach leaves, Fresh Lettuce,	(Severino *et al.,* 2015)
2.	*Salmonella typhimurium*	Green beans, Fresh Lettuce,	(Bhargava *et al.,* 2015)
3.	*Salmonella enteric*	Spinach leaves	(Ruengvisesh *et al.,* 2015)
4.	*Penicillium digitatum*	Green mould (post-harvest)	(Abdolahi *et al.,* 2011)
5.	*Penicillium expansum*	Post-harvest mould (Apple)	(Znini *et al.,* 2013)

Table 2: Anti-microbial/Anti-pathogen properties of essential oils

S.N.	Name of the medicinal/ aromatic plants	Anti-microbial properties of essential oils	Inhibited Micro-organisms	References
1.	Basil	Antibacterial	*Escherichia coli, Pseudomonas aeruginosa (Positive gram), Bacillus cereus, Staphylococcus aureus (Nigative gram)*	(Moghaddam *et al.,* 2011)
		Antibacterial	*Saphylococcus aureus, Streptococcus mutans (Positive gram), Citrobacter freundii, Escherichia coli, Proteus mirabilis, Salmonella choleraesuis, Yersinia enterocolitica (Nigative gram), Enterococcus faecalis, Sarcina sp, Enterococcus faecalis, Acinetobacter sp., Proteus vulgaris, Shigella flexneri, Aeromonas sp., Serratia marcescens, Staphylococcus epidermidis, Klebsiella pneumonia*	(Gaio *et al.,* 2015)
		Anti-fungual	*Fusarium spp.*	(Antunes and Cavacob 2010)
		Anti-yeast	*Candida albicans*	(Adigozel *et al.,* 2005)
		Anti-Fungi	*Alternaria alternate, Aspergillus flavus, Fusarium oxysporum, Penicillium spp.*	(Adigozel *et al.,* 2005)
2.	Lemongrass	Anti-fungal	*Enteriobacteriacae, S. aureus*	(Grohs and Kunz, 2000)
			Rhizopus	(Antunes and Cavacob 2010)
			Alternaria spp.	(Antunes and Cavacob 2010)
			Aspergillus niger	
3.	Mentha		*S. typhimirium, Vibrio parahaemolyticus*, *S. aureus*	(Sokovic *et al.,* 2009)
4.	Eucalyptus		*Escherichia coli*	(Selim, 2011)
5.	Orange oil	Anti-fungal	*Stolonifer*	(Antunes and Cavacob 2010)
6.	Thyme		*Clostridium perfringens*	(Radaelli *et al*., 2016)
7.	Cinnamon		*Escherichia coli, Staphylococcus aureus*	(Zhang *et al.,* 2016)
8.	Salvia		*Enterococcus faecalis, P. aeruginosa, P. vulgaris, Klebsiella pneumoniae*	(Hassanshahian *et al.,*2014)

Table 3: Essential oils used as fumes to control postharvest diseases of fruits and vegetables

S.N.	Name of the plants used for essential oils	Diseases/pathogen causing post harvest loss	Infected crops	References
1.	Thyme (*Thymus* sp.)	*Botrytis cinerea* (Grey mould rot)	Sweet cherries	(Chu *et al.,* 1999)
		Monilinia fructicola (Brown rot)	Sweet cherries	(Chu *et al.,* 2001)
		Anthracnose (*Collectotrichum gloeosporioides*)	Avocado	(Sellamuthu *et al.,* 2013)
			Mango cv. Banganapalli & Totapuri	(Perumal *et al.,* 2017)
2.	Eucalyptus (*Eucalyptus globulous* L.*)*	Antimicrobial properties	Tomato, Strawberry	(Tzortzakis, 2007)
3.	Cinnamon *(Cinnamomum zeylanicum* Blume*)*	Antimicrobial properties	Tomato, Strawberry	(Tzortzakis, 2007)
4.	Lemongrass and cinnamon oils and combination of both	*Colletotrichum musae, Collectotrichum gloeosporioides*	Banana, Papaya	(Maqbool *et al*., 2011)
5.	Oregano and lemongrass oils, vanillin	Psychrophilic microorganisms, yeasts, moulds, L. innocua	Fresh-cut 'Fuji' apples	(Rojas-Grau *et al.,* 2007)

Table 4: Properties of essential oil as preservative agents in fruits and vegetables

S.N.	Essential oils	Effect on produces	Remark	References
1.	Lemongrass Oil	Prepared formulation effective on minimized weight loss, total plate count, respiration rate, yeast, firmness and mold counts during low temperature storage of pineapple fruit.	Coated formulation prepared by using 0.5% (w/v) essential oil of lemongrass	(Azarakhsh *et al.,* 2014)
2.	Lemongrass Oil	After coating the formulation with wax on cut pieces of Fuji apple the activity of *E. coli* minimized	Activity of *E. coli* minimized after coating with formulation	(Via-Trujillo *et al.,* 2015)
3.	Chitosan–lemon essential oil	During the storage of Strawberry fruit mixture of chitosan and lemon oil maximize the antifungal activities and its respiration rate.	The combination of lemon and chitosan oil enhanced the shelf life of strawberry	(Perdones *et al.,* 2012)
4.	Gelatin-based edible coating incorporated with *Mentha pulegium* Essential Oil	The formulations reduced the effect of yeasts, mold and total flora on strawberry fruit	The rate of microbial activities on fruits was dependent on concentration of essential oil and method of application.	(Aitboulahsen *et al.,* 2018)
5.	Basil oil mixed with Beeswax	Application of prepared coat of basil oil with beeswax reduced the post harvest diseased, delayed ripening and enhance shelf life by reduction of weight loss of fruit	To control anthracnose was very significantly of 600 600µL L-1 with beeswax mixture coat	(Karunanayake *et al.,* 2020)

percentage, and the length of time in plum fruits for storage (Aminifard and Mohammad 2013).

3.2 Uses as fumigant

Few essential oils have been used as fumigants to reduce post-harvest diseases in fruits and vegetables because they have volatile compounds which evaporate easily in ambient temperature (Ding and Lee 2019). Table 3 shows essential oils used as fumes to control the diseases of fruit and vegetable.

Use as preservative

Some essential oils have been used as preservative because they have anti micro-organisms properties which inhibit the growth of

Table 5: Major volatile aroma compound available in fruits

S.No.	Fruits	Major volatile compounds	Reference
1.	Mango	ä-3-carene, ethyl butanoate, limonene, terpinolene, R-phellandrene	(Pino *et al.,* 2003)
2.	Banana	(E)-2-hexenal, acetoin, 2, 3-butanediol, solerol, hexanal, isoamyl acetate, 3-methylbutyl acetate, 3-methylbutyl butanoate	(Araguez *et al.,*2013; Nogueira *et al.,* 2003)
3.	Papaya	Methyl butanoate, ethyl butanoate, 3-methyl-1-butanol and 1-butanol	(Pino *et al.,* 2003)
4.	Guava	Acetic acid, 3-hydroxy-2-butanone, 3-methyl-1-butanol, 2,3-butanediol, 3-methylbutanoic acid, (Z)-3-hexen-1-ol, 6-methyl-5-hepten-2- one, limonene, octanol, ethyl octanoate, 3-phenylpropanol, cinnamyl alcohol, R-copaene	(Jordan *et al.,* 2003)
5.	Citrus	Limonene, â-myrcene, linalool, hexanal, ethyl butanoate	(Moufida *et al.,* 2003)
6.	Grape	Linalool, geraniol, (E)-2-hexenal, hexanal, phenylethyl alcohol, octanoic acid	(Rosillo *et al.,* 1999)
7.	Strawberry	Ethyl acetate, ethyl butanoate, butyl acetate, methyl hexanoate, ethyl hexanoate, hexenyl acetate, hexyl acetate, 2-hexenyl butanoate, benzyl acetate, phenylethyl acetate, 1-butanol, 1-hexanol and 2-heptanone	(Prat *et al.,* 2013)
8.	Apple	Acetaldehyde, ethyl butanoate, ethyl methyl propanoate, 2-methyl butanol, ethyl 2-methyl butanoate, 2-methyl butyl acetate, hexyl acetate, butyl acetate, hexyl butanoate, hexyl hexanoate, (E)-2-hexenal, (Z)-2-hexenal	(Araguez *et al.,* 2013)
9.	Pear	Hexanal, hexyl acetate, ethyl hexanoate, ethyl 2-methylbutanoate, ethyl butanoate	(Li *et al.,* 2012)
10.	Plum	(E)-2-hexenal, hexanol, hexanal, (E)-2-hexen-1-ol, (Z)-3-hexen-1-ol, hexyl acetate, (Z)-3-hexenyl acetate, ethyl acetate	(Bononi *et al.,* 2012)

Table 6: Essential oil components use in beverage and soft drink

S.N.	Name of the crops for essential oils used	Components of EOs in Soda (in % w/w)	Name of product
1.	Lemon	Lemonene 54.6, B-pinene 14.5, geranial 2.3, y-Terpinene 19.1, Myrcene 1.5 and a-pinene 3.9	Citrus Soda
2.	Orange	Lemonene 94.2, Myrcene 2.1-4.3, a-Pinene 0.7-1.4 and B-pinene 0.2-1.0	Citrus Soda
3.	Lime	Lemonene 74.8, Myrcene 7.1, B-pinene 1.4, a-pinene 0.3 and y-Terpinene 0.1	Citrus Soda
4.	Neroli Flower Oil (0.8-1 w/w)	Lemonene 27.5, a-Terpineol 14.0, Nerolidol 17.5, a-Terpinyl acetate 11.7 and B-Terpinyl acetate 1.7	Citrus Soda
5.	Cola Leaf Oil	Methyle salicylate 0.6-0.13,	Cola Soda
6.	Nutmeg seed oil (5-15w/w),	Sabinene 50, a-pinene 20, lemonin 8, linalool 6, borniol 6, geraniol 6,	Cola Soda
7.	Cinnamon bark oil,	Cinnameldehyde (65-80%), linalool, ethyl eugenol	Cola Soda
8.	Orange oil	Lemonene 94.2, b-Pinene 02-1.0, a-pinene 0.7-1.4, myrcene 2.1- 4.3	Orance Soda/Fanta

Resource: Ameh *et al.,* 2016

micro-organisms of the preserved materials and also protect from spoilage with enhance shelf-life. The essential oil of *Thymus capitates* L. has been used to prevent several diseases of fruit plants (Abd-AllA *et al.,* 2011). Table 4 shows the use of essential oils as a preservative.

Use as flavouring and fragrant agent

Essential oils are mainly used as flavouring agent compounds that are supplemental, enhanced, or modified in modest doses to the food of natural flavour or aroma without adding any of its own distinctive flavours or aromas. There are three

Table 7: Patents on use of essential oils in post-harvest technology

S.N.	Patent No.	Inventor	Title	Field date/Application no./ Publication date
1.	WO 93/06735	(Vassilis D and Panagiotis T 2017)	Post-Harvest fruit protection using components of natural essential oil in combination with coating waxes	Filed date: 09.10.92 Application No. PCT/GR92/ 0018 Publication date: 15.04.1993
2.	EP1106070A2	(Freire GM and Maria J 2001)	Natural source composition for the control of post-harvest pathologies and method of application	Filed date : 10.11.2000 Application No. 00500230.8 Publication date-13.06.2001
3.	US9,957,215 B2	Louis Perez, Camille Mol, Ronald C. Bakus, James Rogers, Gabriel Rodriguez (2018)	Plant extract compositions for forming protective coatings	Filed date: 18.08.2017 Application No.15 / 680, 541 Publication date-15.02.2018
4.	US10,517,310B2	Louis Perez, James Rogers, Ronald C. Bakus, Chance Holland, Jenny Du (2017)	Plant extract compositions and methods of preparation thereof	Filed date: 26.07.2017 Application No. 15 / 660,260 Publication date: 9.11.2017

categories of flavouring agents i.e. natural, nature-identical, and artificial, according to European Union law. There are so many volatile compounds show in Table 5, presented in fruits that are responsible for fragrance and flavour. The same volatile compounds are available in different essential oil (Table 6) extracted from different aromatic plants which are used in food industries for the preparation of natural flavour and fragrance.

Use of essential oils in beverage and soft drink

Methyl eugenol, which is found in many plants and vegetables, is another widely used flavoring agent to enhance the taste and aroma. The European Union establishes maximum levels for the use of this substance in food, and the permitted amount in nonalcoholic beverages is up to 1 mg/kg.

Available patent on minimizing post-harvest losses

Various studies are available on essential oil used for reducing post-harvest losses. Some patents (Table 7) are also assigned on the method which is related to coating wax used to reduce post-harvest losses.

CONCLUSION

Since last decade, a large number of studies have been conducted to demonstrate the benefits of essential oils in post-harvest handling of fruits and vegetables. In various studies, it has been found that the essential oils are used as coating wax, vapour, preservative, fragrant & flavour, beverages and soft drink to enhancement the overall quality and shelf-life of fruits and vegetables. The essential oils of some plants like lemongrass, mint, basil, rosemary, thyme, eucalyptus, salvia, cinnamon, etc. have properties against various diseases and plant pathogens, especially which are responsible for losses in horticultural produces. Recently, the use of natural essential oils increased because of it is safe and non-toxic for human health and environment. Hence, the demand and interest towards use of essential oils in post harvest handling has also enhanced in recent years. Although, various research works have been done on advantages of essential oils, but further research on effect and uses of essential oils in post harvest management needs to scientifically prove for reduction on the losses and improve the overall quality of horticulture crops.

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Review article

A Review: Phytochemical constituents and medicinal importance of Dashapushpam

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ABSTRACT

The Dashapushpam group consists of ten holy flowers, which are used in the Ayurvedic medical system and have amazing medicinal and prospective effects. It contains many flowers with distinctive personalities. These flowers, which have divine power, belong to various families, including Amaranthaceae, Oxalidaceae, Sapindoidae, Asteraceae, Convolvulaceae, Poaceae, and Hypoxidaceae. It covers things like anti-inflammatory, antimicrobial, cancer-fighting, anti-fungal, antipsychotic, anti-rheumatic, antiarrhythmic, anti-diarrheal, anthelmintic, and anti-diabetic activities. It offers a well-established conventional medical system. They also provide therapeutic advantages without any negative side effects. The main goal of this article is to inform readers about these herbs generally and their value as medicines.

***Keywords*:** Dashapushpam, phytochemicals, phytoconstituents, sacred plants, therapeutic potential

INTRODUCTION

Plants have satisfied all of man's needs for food, shelter, taste, aroma, not to mention medicine and other essentials, since the dawn of civilization. These plants additionally play a role in people's religious practices. Kerala is one of the main states of South India, where the use of plants is evident in both traditional and Ayurvedic medicines. The populace of Kerala has ten herbs that are revered as sacred, and they are known as "Dashapushpam" or "Ten Sacred Plants" (Fig.1). Dashapushpam literally translates to "ten flowers" (where "Dasha" stands for "ten" and "Pushpam" for "flowers") in Sanskrit. Dashapushpam is a collection of ten species of angiosperms that come from several plant groups and are valued by South Indians, especially Keralites, for their religious use and medicinal or therapeutic properties. A brief description of Dashapushpam has been given in the Table 1. The Western Ghats, situated in the state of Kerala, is one of the 34 biodiversity hotspots worldwide. Dashapushpam, the sacred plant of Kerala history and culture, is one of the more than 200 taxa of plants employed in Ayurveda. Over 4,500 different blooming plant species, including 1,500 endemic taxa, can be found throughout the state. Kerala is renowned for its abundance of medicinal plants and its history of using an indigenous medicine called Ayurveda. Herbs are important in the pharmaceutical industry because they are natural sources of medicine that can save lives. In Karkkidamasam (mid-July–mid-August), dashapushpas are ingested in the form of Karkkidaka kanji to avoid aggravating vatadosha during the forthcoming varsharitu (monsoon season). Folk medicine has long utilized these herbs to treat a wide range of ailments, including vomiting, lithiasis, gonorrhoea, indigestion, intermittent fever, skin disorders, and urogenital diseases. Majority of them have been medicinally verified for several bio-activities.

Different Dasapushpam along with its phytochemical's constituents

Aerva lanata

According to the World Health Organisation's most recent data, 80% of people worldwide rely on herbal medicines for a portion of their basic healthcare (Adepu *et al*., 2013). One such gift among the many medicinal herbs that nature has provided is *Aerva lanata*. It is a prostate, upright undershrub (Goyal *et al*., 2011). In Hindi, it is known as Kapurijadi or Gorakhabooti, while in English, it is known as Mountain Knotgrass. It is

also known as Gorakshaganja, Satkabhedi, and Aadaanpaak in Sanskrit (Nagaratna *et al*., 2014). It could be considered a form of Pashanbheda, which is a remedy used in Ayurveda to remove urinary tract stones. It is native to Saudi Arabia, tropical Africa, Sri Lanka, Philippines and Java, where it grows to a height of 30 to 80 cm (Rajesh *et al*., 2011). It has a wide range of therapeutic uses in conventional and folk medicines across numerous geographical contexts (Bitasta *et al*., 2016). A paste made from the entire plant is utilised, for the treatment of spermatorrhoea, while a paste made from the leaves is used to treat wounds (Malik *et al*., 2012). The plant extract is effective as a diuretic, emollient, vermifuge, astringent, and in the treatment of diabetes and ureteral stones (Kumar *et al*., 2011; Sharma *et al*., 2011). It cures bleeding piles and malaria and serves as a snake venom antidote (Sukumaran *et al*., 2014). The plant's leaves are utilized in the treatment of diabetes, hypertension, fever, and cough (Chander *et al*., 2014). The entire plant as well as root portion is used for lithotriptic, astringent, and cough purposes (Sivasankari *et al*., 2014).

Phytochemicals constituents in *Aerva lanata*

As per Ragavendran *et al*. (2012) Indian water lily is rich in diverse array of plant-based constituents. Through phytochemical screening, it has been found to contain various types of phytochemicals. Furthermore, FTIR analysis of different parts of the plant, including roots, stems, leaves, and flowers (Yamuna *et al*., 2012), unveiled the existence of various functional groups, such as alcohols, aldehydes, nitro compounds, amides, carboxylic acids, ethers, amines, alkyl halides, and phenols. This indicates the diverse array of chemical constituents present in *Aerva lanata*. Additionally, the entire plant includes necessary trace elements like calcium, silica, magnesium, potassium, chloride, carbon, and oxygen (Ragavendran *et al*., 2012). According to an ethanolic extract HPTLC study, the roots contain a significant amount of gallic acid. Quinones, phenols, triterpenoids, phytosterols, and phlobatannins were all detected during a phytochemical screening of the root extract (Vijaylakshmi and Ravindhran, 2012).

When analysed by HPLC, the soluble extract that comes from the stem included 3,5,7,3,4,5,-OH (myricetin), apigenin-7-O-glucoside (apigetrin), quercetin-3-O-rutinoside (rutin), and 3,4,5-OH (gallic acid) (Kumar and Kumar, 2011). Total chlorophyll content, methoxykaempferol chlorophyll a, and chlorophyll b were reported to be present in the white and yellow-coloured varieties of *Aerva lanata* (Denniand Mammen, 2013). When the leaves, stems, roots, flowers, and seeds were analysed using GC-MC, a variety of different substances were found, including pyridine, hydroquinone, monobenzyl ether, docosane, dotriacontane, R,Z-12-hydroxy-9-octadecenoic acid, 2-isopropyl-2,5-dihydrofuran, and a wide variety of other substances (Mariswamy*et al*., 2013).

Biophytum sensitivum

The *Biophytum sensitivum* is primarily found in tropical Asia, America, and the Philippines. According to Ayurvedic tradition, it is bitter, expectorant, stimulant, and tonic, and it is mainly used to treat fever, malaria, tuberculosis, burns, phthisis, arthritis, back pain, bone spurs, bursitis, carpel tunnel syndrome, cervical spondylitis, degenerative joint disease, degenerative neck disease, fibromyalgia, and leg cramps (Bharati *et al*., 2012). It has been used historically through medicine to treat indigestion, asthma, somnolence, convulsions, chest pain, and tumours. Whole plant decoctions are taken orally. In the foothills of the Himalayas in India, Ayurveda prescribes the root for gonorrhoea and lithiasis and uses it typically for inflammation and chronic skin conditions.

According to Nagbeli, an ancient form of medicine practiced in Nepal and the foothills of the Himalayas, leaves are taken orally to treat diabetes. In Mali, Africa, powdered leaves and seeds are applied topically to treat wounds. In India, both Ayurveda and Siddha claim that snake envenomation is cured by an entire plant decoction swallowed internally (Sakthivel *et al*., 2012).

Phytoconstituents constituents of *Biophytums ensitivum*

Numerous chemical components, primarily polyphenolic and phenolic compounds, saponin, polysaccharides, essential oils, and pectin, have

been identified by studying the phytochemistry of B. sensitivum. Two bioflavons, amentoflavone and cupressuflavone, three flavoids, isoorientin, 3-methoxy-uteolin 7-O-glycoside, and luteolin-7-methyl ether, as well as two acids, 4-caffeoilinic acids, and aerial parts of B. sensitivum were wet-isolated to make up the major bioactive components. Additionally, it contains proanthocyanidins, which are condensed forms of tannins, 3'–8 pipigenin, and a few phenolic compounds. The airborne portion of the system was separated from these compounds (Sakthivel and Guruvayoorppan, 2012; Pawar andVyawahare, 2014).

Eclipta alba

Eclipta alba (L.) an annual, multi-branched herbaceous plant. It typically reaches a height of 30–50 cm and can have an prostrate growth habit. The plant have white-colored hairs in it, and its leaves have hairs on both surfaces. The fruiting period of *E. alba* occurs between September and October (Fenz *et al*., 2019 and Sunitaand Mishra, 2016). The stem of this plant is red in color. It is considered a weed and can grow up to 2000 meters above sea level in subtropical and tropical regions around the World, including Africa, Asia, and South America. It is particularly widespread in countries such as Taiwan, Brazil, Japan, Indonesia, the Philippines, India, China, Bangladesh, Thailand, and the United States. In India, it is commonly found in states like Bihar, U.P, Assam, and Manipur (Mithun *et al*., 2011; Singh *et al*., 2017; Soni and Soni, 2017; Sinha *et al*., 2016; Thenmozhi and Jayanthi, 2019). The medicinal properties of this herb are more known than its other pharmacological effects, they have beneficial rejuvenating qualities as well as analgesic, anti-inflammatory, anti-hepatotoxic, anti-hyperglycemic, antioxidant (Thenmozhi and Jayanthi, 2019) etc.

Phytochemicals constituents of *Eclipta alba*

Bhringraj, also referred to as *E. alba*, is a plant that includes a variety of phytochemical molecules, among which are flavonoids, coumestans, polyacetylenes, alkaloids, glycosides, triterpenoids and, as well as phenolic acids, sterols, saponins, sesquiterpeneulactones, amino acids, proteins, and carbohydrates (Saxena *et al*., 2016; Latha *et al*., 2017). Detail Phytochemical constituents are presented in Table 2.

Cyanthillium cinerea

Cyanthillium cinerea is a widespread common plant that is utilized in various traditional Indian medicines. It is a perennial herb that features flat-topped clusters of many flower heads with rosy ray petals. Various traditional uses of the plant have therapeutic potential. Infections of the eyes and fever are treated using the entire plant. To obtain anthelmintic and alexipharmic medication, seeds are employed as a source. The leaves of *Cyanthillium cinerea* possess analgesic, antipyretic, and anti-inflammatory effects. Whole plant is used to treat renal problems, dermatitis, menstrual cramps, stomach ache, diarrhea, and as a diuretic decoction. Children who wet the bed are given this plant's juice to help them (Abdullahi *et al*., 2015).

Phytochemical constituents of *Cyanthillium cinerea*

The triterpenes are the main components. Luteolinmonobeta-D glucopyranoside was produced via aerial components. The entire plant produced potassium chloride, phenolic resin and triterpene chemicals like beta amyrin acetate, lupeol acetate, lupeol and beta amyrin. It also produced sterols like beta-sitosterol, stigma sterol and alpha-spinasterol. The flower (used to treat conjunctivitis), seeds (used as an anthelmintic), roots (used to treat dropsy) and juice (used to treat piles) are among the helpful parts (Raj *et al*., 2013).

Evolvulus alsinoides

A flat perennial herb with a weakly branched wood rootstock is called *Evolvulus alsinoides* belongs to Convolvulaceae family. They are rounded in shape and have several annual branches that are about 30cm long on the outside. The branches typically crouch and encircle lengthy hairs. The elliptic-shaped small leaves are found in *Evolvulus alsinoides*, they are acute, sensitive and densely branched and every part of this plant is utilized to make Ayurvedic medication to cure fever, a cough and a cold. Inflammation related neurodegenerative diseases are also treated with them (Prochazkova *et al*., 2011).

Phytochemicals constituents of *Evolvulus alsinoides*

Three alkaloids, namely betaine, shankhapushpine, and evolvine, are found in the *Ecliptaalba* plant. Additionally, fresh plants of this species contain volatile oil, a yellow neutral fat, organic acids, and saline materials. Several substances were identified from E. alsinoides through chemical analysis, including 1,3-di-O-caffeoyl quinic acid methyl ester, caffeic acid, and 3-Hydroxy-4-(2,3,4-trihydroxy-2-methylbutoxy)-phenyl-2,3,4-trihydroxy-3-2-propenoate (1). kaempferol-3-O-glycopyranoside, quercetin-3-O-glycopyranoside, kaempferol-7-glucopyranoside, and 6-methoxy-7-glucopyranoside coumarin (Raj *et al*., 2013).

Emilia sonchifolia

In Malayalam, it is Muyalcheviyan. The herb is glabrous, slender, and 30–40 cm tall. The flowers are purple, and the leaves are obviated. It is sometimes referred to as Cupid's razor. Typically, waste grounds and damp regions are where we can find the plant. The entire plant is utilized for a variety of therapies. Dysentery is treated using a tea brewed from the leaves. The leaf juice is applied topically to treat wounds, sore ears, eye inflammation, and night blindness. Roots are used to treat diarrhea. There are claims that the methanoic extract and fresh juice of *E. sonchifolia* leaves have anti-inflammatory and antioxidant properties. The water extract of the plant shows antibacterial activity (Raj *et al*., 2013).

Phytochemical constituents of *Emilia sonchifolia*

Alkaloids, flavonoids, and terpenes have all reportedly been found in the plant's aerial parts. Senkirkine, doronine, and pyrrolizidine alkaloids are present in the aerial portions. Similar sitosterol, palmitic, and triacontannic acids are also said to be present in the plan (Raj *et al*., 2013).

Cardiospermum halicacabum

This is the common term for balloon vines. Tamil for "Mudakkathan". Annual climbers have tenders that have a light pubescence. Biternate leaves are actually trifoliate leaves with three leaflets on each section and coarse serration on each leaflet. The axillary heads of the flowers are normally white with a yellow centre, and there are three flowers by abortion. Fruit is a green, inflated, membranous capsule that is over 2cm long and turns brown as it dries. Round dark seeds with noticeable heart- or kidney-shaped markings (Jayanthi *et al*., 2012). This plant thrives on the plains of Bangladesh, India, Pakistan, and America and is common throughout the world's tropical and subtropical climates (Rajesekaran *et al.,* 2014). The entire plant has long been used to treat rheumatism, stiffness in the limbs, snake bites, and anxious diseases like diaphoretic, diuretic, stomachic, and sudorific; its leaves and stalks are also used as a poultice for swellings and diarrhoea and are used to treat dysentery and headaches. (Vinoth *et al*., 2013). For the treatment of bone fractures, it is used in Sri Lanka. On the market, there are a number of *C. halicacabum* products available, including gel, lotion, shampoo, and spray. Dry, itchy skin and scalp may benefit from these products (Patil *et al*., 2011).

Phytochemical constituents of *Cardiospermum halicacabum*

Proteins, carbohydrates, tannis, alkaloids, glycosides, steroids, and saponins were found *in C. halicacabum*, according to a phytochemical investigation (Raza *et al*., 2013; Patil *et al*., 2011). In addition to other active compounds, gas chromatography identified 1,2,4-trioxolane-2-octanic acid, 5-octymethyl ester, ricinolenic acid, [1,1-bicyclopopyl], 2-[9-octadecenyloxyl], and 1,2,3-trioxolane-2-octanic acid in *C. halicacabum* (Kumarand Kumari, 2012).Oleic acid, 2-hexyl-methyl ester, 11-octadecenoic acid, methyl ester, 7-methyl-7-tetradecan-1-ol acetate, and 1,2,3-propanetriyl ester are among the substances that mass spectrometry has identified (Rajesh *et al.,* 2016). Apigenin-7-o-glucuronide, arachidic acid, chryoerior-7-o-glucuronide, linoleic acid, luteolin-7-o-glucuronide, and stearic acid are the components that make up (+) pintol (Aishwarya *et al*., 2014).

Curculigo orchioides

The plant called "*Kali musali"* in Sanskrit, is a little tuberous herbaceous plant. Flowers across the entire region are yellow and have narrow, linear

Table 1 : Brief description of Dasapushpam

Names in Latin	In Sanskrit	In English	Family	Part utilized	References
Aerva lanta (L.) AL Juss. Ex Schultes	Bhadra	Indian water lily	Amaranthaceae	Whole plant	Bitasta *et al*. (2016)
Biophytum sensitivum (L.) DC	Viparithalajjalu	Sensitive wood sorrel	Oxalidaceae	Whole plant	Sakthivel and Guruvayoorappan (2012)
Cardiospermum halicacabum Linn.	Indravalli	Balloon vine	Sapindaceae	Shoot/ leaves	Shree and Azamthulla(2019)
Curculigo orchioides Gaertn.	Musali	Black musali	Amaryllidaceae	Tubers	Nagesh *et al*. (2016)
Cynodon dactylon (Linn.) Pers	Murva	Bermuda grass	Poaceae	leaves	Parihar *et al*. (2021)
Eclipta alba (L.) Hassk	Bhringaraja	False Daisy	Asteraceae	Shoot/ leaves	Kumari *et al*. (2021)
Emilia sonchifolia (L.) DC	Akhukarni	Canada Flea-bane	Convolvulaceae	Shoot/ leaves	Xu *et al*. (2020)
Evolvulus alsinoides Linn.	Harikrantha	Slender Dwarf Morning Glory	Convolvulaceae	Whole plant	Arun *et al*. (2013)
Ipomoea sepiariakoen. Ex Roxb	Lakshamana	Ipomoea	Convolvulaceae	Whole plant	Arun *et al*. (2013)
Cyanthillium cinereum (L.) H.Rob.	Sahadevi	Ash coloured Flea-bane	Asteraceae	Whole plant	Arun *et al*. (2013)

Table 2: Phytochemical constituents of *Eclipta alba* Hassk

Nature of phytoconstituents	Phytoconstituents	References
Coumestans	Wedelolactone, demethylwedelolactone, demethylwedelolactone-7-glucoside	Kumari *et al*. (2021)
Tereterpenoids and their glycosides	Eclalbasaponins VII-X (taraxastanetriterpeneglycosides), Eclabasaponins I-VI (oleananetriterpeneglycosides), Eclabasaponins I-VI (triterpene glycosides), ecliptasaponins C and D (triterpenoid glycosides), á-amyrin, Oleanolic acid, ursolic acid (triterpenoids).	Rafif *et al*. (2022)
Sterol	Stigmasterol, daucosterol, Stigmasterol-3-O-glucosideâ-sitosterol.	Kaur *et al*. (2011)
Alkaloids	[(20S) (25S)-22,26 imino-cholesta-5,22(N)-dien-3â, 23-ol] (verazine), [20-epi-3-dehydroxy-3-oxo-5,6-dihydro-4, 5-dehydroverazine], [(20R)-20-pyridyl-cholesta-5-ene-3 â,23-diol] (ecliptalbine), [25â-hydroxyverazine].	Rafif *et al*. (2022)
Flavonoids	Luteolin-7-glucoside, leteolin, apigenin, orobol (isoluteolin)	Kumari *et al*. (2021)
Volatile oils	Heptadecane, 6,10,14-trimethyl-2-pentadecanone, n-hexadecanoic acid, pentadecane, eudesma-4(14),1-diene, phytol, octadic-9-enoic acid, 1,2—benzenediacarboxylic acid di-isooctyl ester, (Z,Z) -9,octadecadienolic acid, (Z)-7,11-dimethyl-3-methylene -1,6,10-dodecateriene, (Z,Z,Z)-1,5,9,9-tetramethyl1,4,7-cycloundecatriene	Kumari *et al*. (2021)
Saponins	Eclalbatin (triterpenesaponin), dasyscyphin C	Kumari *et al*. (2021)

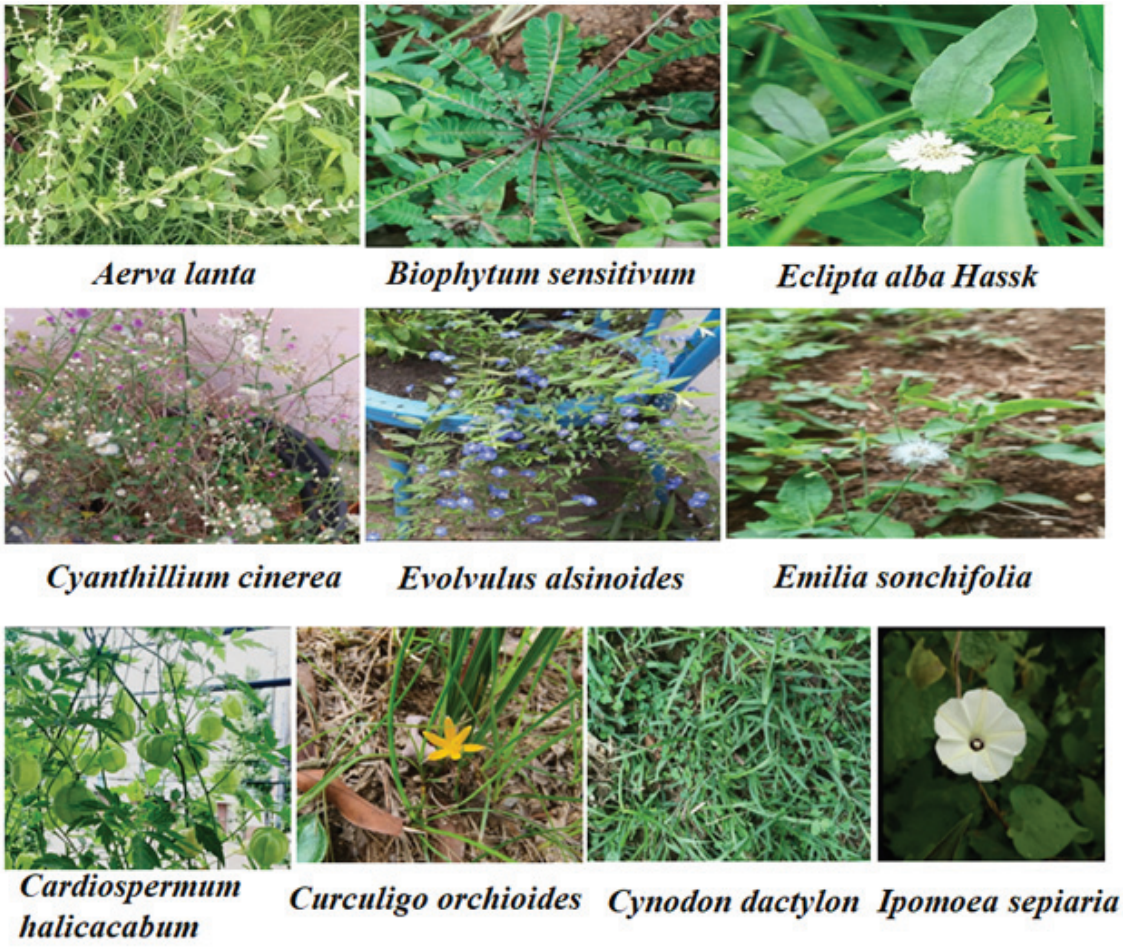

Fig. 1: Different types of Dashapushpam

leaves. This medicinal plant is used to cure asthma, osteoporosis, diabetes,jaundice, make tonics, prevent the disease and fight cancer (Nagesh *et al*., 2016; Xia *et al*., 2016; Sharma *et al*., 2017). Numerous phenol and phenolic glycosides, lignans, lignans glycosides and polysaccharides were discovered as a result of chemical analysis of *C. orchioides* (Nie*et al*., 2013; Wang *et al*., 2019; Xia *et al*., 2016).

Phytochemical constituents of *Curculigo orchioides*

The fruit of the plant has been used to isolate Curculin C, which is a peptide containing amino acids (Raj *et al*., 2013). The rhizome also contains additional saponins, sapogenins, phenolic glycosides, a triterpene alcohol, and curculigosaponin C and F.

Cynodon dactylon

Cynodon dactylon (Linn.) Pers called "Dhruva" in Sanskrit, may flourish in unfavorable conditions. Bermuda grass, also known as Dhrub grass, was highly regarded by the Hindus and is still used in temple devotion today. A huge twining herb with soft-to-the touch extremities and deeply furrowed corky bark on older stems. It flourishes all over India's southern region. It thrives in open spaces that frequently experience disturbances like grazing, flooding, and fire.

Phytochemical constituents of *Cynodon dactylon*

Triticin**,** hydrocyanic acid, and cynodin are all present in Bermuda grass. The plant has been linked to the p-coumaric, syringic, phenolic phytotoxinsferulic, vanillic, p-hydroxybenzoic, and o-hydroxyphenyl acetic acids. The leaves contain flavone C glycosides and flavonoid sulphate (Raj *et al*., 2013).

Ipomoea sepiariakoen

This plant is known as "Lakshmana" in Sanskrit (Majumder *et al*., 2013). The perennial climber *Ipomoea sepiaria* is a significant ethano medicinal plant having phytochemical constituents like carbohydrates, alkaloids, glycosides, saponins, flavonoids, phenolic compounds and tannin. Human prostate cancer cell lines known as PC3 (PC-3) are widely utilised to study the biochemical alterations that occur in prostate cancer cells (Meesala *et al*., 2017).

Phytochemical constituentsof *Ipomoea sepiaria*

Ipobscurine A, B and C, non-ergoline type indole alkaloids, as well as Ipobscurine C, a serotonin alkaloid, are found in the seeds of Ipomoea resin (Majumder *et al*., 2013).

CONCLUSION

These ten sacred plants have limitless therapeutic potential and are underutilized for treating a variety of diseases. The creation of novel therapeutic compounds from these plants has to be fostered through research and development. Its criteria, pharmacological activity, toxicity, and clinical trials should all be carefully examined in order to produce novel drugs for a range of illnesses.Research on phytomedicinehas significantly increased on a global scale. Therefore, the potential for developing drugs from these plants is enormous. Over the past few decades, certain research projects have been conducted on these plants, which has sufficiently inspired the scientific community to learn more about these revered plants. The potential of Dashapushpam should be the subject of research and development.

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Review article

Roscoea purpurea (Kakoli): Exploring the imperative for conservation of an endangered Ashtawarga plant through comprehensive review

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ABSTRACT

In COVID Era, achieving holistic health through Ayurveda is gaining popularity day by day. However, market fails to satisfy this ever-increasing demand due to unavailability and extinction of some of the major medicinal flora. Being an Ashtawarga plant, Roscoea purpurea, known for its multiple Ayurvedic formulations, medicinal and rejuvenating properties, is also available in very limited quantity and considered endangered. Traditionally, various parts of the plant like leaves, roots and flowers are used for treating diseases like diarrhoea, diabetes, inflammatory disorders, rheumatic pain and fever. On account of its rich chemical base consisting polyphenols and flavonoids, it is reported to have anti-cancerous and anti-oxidant activities. Howbeit, due to its endangered status, Department of AYUSH, Govt. of India has permitted to swap these kind of rare herbs with accessible substitutes. Adulteration with other plants opens the ground for exploitation in drug industry and degrades the quality and credibility of Ayurvedic medicine. So, the study aims to provide the importance of this endangered plant in lieu of its traditional uses, Ayurvedic properties and bioactivities. It also tries to emphasize that, owing to its rich medicinal record, not only the plant needs to be explored further but conservation is important as well. Hence, a strategic conservation plan has also been presented.

***Keywords*:** *Ashtawarga*, Ayurveda, conservation, herbal drugs, *Roscoea pupurea*,

INTRODUCTION

Roscoea is an important genera belonging to the family Zingiberaceae which constitutes nearly 24 herbaceous tuberous geophytes, according to Royal Botanic Gardens, Kew having various medicinal uses (Dhyani *et al*., 2020; Zhao *et al*., 2017). *Roscoea purpurea,* known by the common name of *Kakoli*, is a large lush green perennial rhizomatous herb between 15-30 cm in height with light brown, thick, fleshy, and bundled roots (rhizomes). The leaves are fleshy and flowers are hooded expressing a purple tone shade (Sahu *et al*., 2010; Gopal *et al*., 2014). It is an important species of the "*Ashtawarga*" group with *Jivaka-Rishibhak, Mahameda-Meda, Riddhi-Vriddhi* and *Kakoli-Kshirakakoli* as clan members. These plants are claimed to be useful in healing and treating fractured bones, weakness, pyrexia, body overweight, and regulating diabetes; also renowned for their effect in balancing *vata*, *pitta*, and *raktadoshas* (Dhyani *et al*., 2010; Chinmay *et al*., 2011; Virk *et al*., 2015; Rajashekhar *et al*., 2015; Misra *et al*., 2015). Due to lack of clarity, several different authors used the substituted species of *Roscoea procera* Wall., *Roscoea alpina* Royle and others but it is evident that the genuine species of *Kakoli* is *Roscoea purpurea* Smith, which can be characterized by purple colour stout stem and flowers as described in ancient literatures of *Shaligram* and *Bhav Prakash Nighantus* and also indicated by Balkrishna *et al*. (2012). Although, due to its rare nature, the Ministry of Ayush has now permitted to replace it with other available sources like *Aswagandhâ* (*Withania somnifera* (L.) Dunal) and *Krsna-mûsalî* (*Curculigo orchioides* Gaertn) but the real nature of the plant should be conserved, for which its conservation and cultivation is mandatary (Virk *et al*., 2017).

Further, in order to correctly characterize the true identity of species, some compounds have been isolated and purified from different plant parts to be used as chemical markers. Kaur *et al*., (2020a)

has reported the presence of a compound, named Sitostanol caffeate in the roots of plant, which can be used as chemo-marker. Several other studies reported the presence of flavonoids, steroids, terpenoids, alkaloids and phenolic acids. Rhizomes of *R. purpurea* are reported to possess significant antidiabetic and hypolipidemic activity. In other studies, immunomodulatory, spermopiotic, and anti-tubercular potency have been revealed (Bairwa *et al*., 2012; Subramoniam *et al*., 2013; Kumar, 2014). Traditionally, roots (rhizomes), leaves, and flowers are used in the treatment of diabetes, diarrhoea, high blood pressure, hyperthermia, haematemesis, polydipsia, rheumatic pain and inflammation etc. Immuno-stimulating properties of rhizomes, extracted in ethanolic extract have also been demonstrated (Handa, 1980; Singh and Rawat, 2011; Gopal *et al*., 2014). The rhizomes and tubers are essential ingredients of number of herbal formulations prepared like tonic and Chyawanprash (Handa, 1980; Singh and Rawat, 2011; Kaur*et al*., 2020a). In Nepal, boiled tubers of the plant are eaten and traditionally used in veterinary medicines (Handa, 1980; Singh and Rawat, 2011). So, with an objective to present a comprehensive and upgraded review of this highly valuable Ayurvedic species, this study has been drafted to include traditional uses, major phytochemicals and biological studies conducted till now. The present review summarizes the updated information regarding the chemical constituents, pharmacological study, medicinal uses and ayurvedic significance along with its various formulations as well as also mentioned the different ways to conserve and cultivate it which is a pressing need.

Botanical description

Kakoli is a stout perennial herb, 25-38 cm tall. Leaves are 4-8, elliptic, lance-shaped to oblong-ovate, 14-20 cm long, sometimes sickle-shaped, tip tapering and slightly eared at a base on lower leaves, side veins parallel; leaf eared fringed with hairs. Flowers are borne in a cluster at the top, the cluster stalks are enclosed by upper leaf sheaths, only the upper part of bracts and flowers visible. Typical structured flowers are slightly purple, mauve, lilac, pink or white with purple markings where only one or two flowers open at a time. Outer calyx is tubular-shaped, three petals arising from the calyx form a slightly longer tube than the calyx and terminates with trilobular structure raised vertically having one hooded central lobe surrounded by two slightly smaller side lobes. Staminodes, constituting four sterile stamens, are formed inside the petals where the two lateral staminodes appeared as small straight petals and the other two central ones got partially fused at the base and forms a lip. Lip is non-deflexed, 4.5-6.5 × 2-5 cm (Flowers of India).

Geographical distribution

As a native species of Nepal, *R. purpurea* is distributed and scattered in India and Bhutan also, where it prefers to grow along green hillsides, alpine grassland, stony ridges and slopes of Central to Eastern Himalaya extending from Uttarakhand to Assam and Sikkim, upto an elevation of 3300 m (Dhyani *et al*., 2010;Gopal *et al*., 2014).

Ayurvedic properties and formulations of *Kâkoli*

Kâkolî is one of the most profligate plants of the nature. It is grouped among *Ashtawarga* plants, *Jîvanîya*, *Sukrajanana* and *Bramhaniyagana* in different ayurvedic scriptures (Mishra, 2020). The plant is sweet in taste, heavy and mucilaginous in attribute and cold in potency. In Ayurveda, it is considered *Vatapittasamaka, slesmataka*; *roghanatâ* and is a part of different Ayurvedic preparations namely, *Chyavanprashrasayan* (a class of rejuvenating tonics), *Mahakalyan* and *Jivaniyaghrita* (Herbal medicines with *ghee* as a base), *Vachadi* and *Chitrakadi* taila (herbal oil, having properties to treat enlarged lymph nodes, fistula and sinus), *Vrahinigutika* (useful against erectile dysfunction) and *Astawarga* and *Jivaniya* gunachurna (Marde and Mishra, 2019).

Phytoconstituents of *Roscoea purpurea*

Several bioactive compounds from different chemical categories such as diterpenoids, flavonoids, and phenolic acids have been reported from *R. purpurea* till now (Table 1). Sitostanol caffeate, a compound claimed to lower the risk of coronary heart diseases has been isolated from the roots by Kaur *et al*., (2020a), which can be further utilized as a marker to identify and differentiate the species from its homotypes. In Nepal, the boiled tubers are eaten and used in veterinary medicine. A research conducted by Misra *et al*., (2015) have

proved the nutraceutical value of these tubers and showed that they are highly rich in fibre (28%), proteins (3.5%) and oil (3.5%). Total phenolic and flavonoid content were found to be ranged between 7.10 to 6.10 %, respectively. Phytochemical screening of the powdered tubers showed the presence of alkaloids, carbohydrates, flavonoids, glycosides, phenolics, proteins, saponins, and tannins (Misra *et al*., 2015, Devkota and Timalsina, 2021). The nutritional components of the rhizomes constitute fiber, oil, protein, sugar, and starch (Owolabi *et al*., 2012; Misra *et al*., 2015). Rawat *et al*. (2014) reported the presence of riboflavin, thiamine, minerals, fat and fibers in rhizomes. The methanol extract having blackish-brown composition displayed the presence of two compounds belonging to catechin-type of class such as epigallocatechin and epicatechin along with other compounds in the roots (Kaur *et al*., 2020a).

Singamaneni*et al*. (2021) reported two new compounds coronarin K and L from the class labdanediterpenes along with other known compounds as coronarin A, kaempferol 3-O-methyl ether, kaempferol, fenozan acid, 3-(3-methoxy,4-hydroxyphenyl)-2-propenoic acid ferulic acid, caffeic acid, bisdemethoxycurcumin and gallic acid from its rhizomes. Coronarin K demonstrated to have anti-cancer potency when evaluated against lung cancer cell line (A-549).

Miyazaki *et al*., (2014) isolated flavonoids as kaempferide, kaempferide 3-O-β-D-glucuronopyranoside, kaempferol 3-O-β-D-glucuronopyranoside and (Z)-3-hexen-l-ol-β-D-glucopyranoside from aerial parts and kaempferide, kaempferol 3-O-methyl ether from the rhizomes of *R. purpurea*. Kaur *et al.* (2020b) reported the triterpenoid, lupenone for the first time from *Kakoli* roots. Similarly, stigmasterol was also obtained from powdered leaves of *R. purpurea* by Barai and Bag (2019) which is known to possess various pharmacological activities including anti-osteoarthritic, antioxidant, anti-inflammatory, anti-mutagenic, hypoglycemic, anti-genotoxicity and anticancerous (Kaur *et al*., 2011; Ali *et al*., 2015). They have also reported the presence of monoterpenoids in ethanolic extract of leaves by GC-MS study.

Medicinal uses

Roscoea pupurea is an important species contributing to the Ayurvedic members of *Ashtawarga* plants, considered as prime *rasayana* group possessing health rejuvenating properties. Due to its anti-oxidant, anti-anging and cell regenerative properties, it is an essential ingredient of several poly herbal formulations, specially '*Chayawanprash*', which is widely used in India as an energy and immunity booster supplement (Misra *et al*., 2015; Raval *et al*., 2015). Its regular use also enhances the intellect, long life and memory (Tripathi, 2013). Traditionally, its roots and other parts in the form of various preparations are used medicinally (Table 2). Roots and rhizomes are considered as good appetizer, aphrodisiac, and tonic (Miyazaki *et al*., 2014). Therapeutically, they are reported to be anti-rheumatic, diuretic, expectorant, febrifuge, galactagogue, haemostatic and *Sukrajanana* (semen count enhancer) in nature (Balkrishna *et al.,* 2012; Sharma and Sharma 2008). In Ayurveda, it roots are considered beneficial in *Vishamajvara* (high fever), *Pittaroga, Raktapitta* (Bleeding disorders)*, Netraroga* (Eye disorders)*, Hridayaroga* (Heart disorders)*, Daha* (Burning sensation) and *Swasaroga* (Respiratory disorders) (Ayurvedic Pharmacoepia of India, 2001). According to Saheli *et al*. (2019), the plant is known to possess anti-diabetic and hypoglycemic effects. The various medicinal properties of the plant along with its parts have been summarized (Fig. 2).

Anti-oxidant activity

Tubers and rhizomes of *R. purpurea* are commonly used for various Ayurvedic formulations, so some of the researchers have tried to summarize pharmacological aspects of these parts. The methanolic extract derived from the tubers of *R. purpurea* was tested for anti-oxidant activity by using ferric reducing power, DPPH and α carotene linoleate assays. The extract has shown to up regulate the reducing power with an increase in concentration, similar to positive control (ascorbic acid, quercetin, rutin and BHT). In DPPH and carotene bleaching assay, the extract showed IC_{50} 810.66±1.154 & 600.66±1.154 μg/ml, which can be justified by the presence of polyphenolic content of the tubers which are well validated for their potent bioactivities (Misra, *et al*., 2015). In a

similar study, the ethanolic extract and its petroleum ether, chloroform, acetone, ethanolic and aqueous fractions prepared from tubers of *R. purpurea* were evaluated for anti-oxidant activity by using DPPH assay. The extract and petroleum ether, chloroform, acetone, ethanolic and aqueous fractions exhibited anti-oxidant effects with IC_{50} of 0.925±0.005, 1.25±0.005, 0.25±0.005, 0.48±0.005, 0.77±0.011 & 1.04±0.005 mg/ml, respectively (Srivastava *et al*., 2015).

Anti-cancer activity

According to WHO, cancer is the leading source of death globally with a record 10 million deaths in 2020. Lung cancer and breast cancer are among the most common cancers and the search for plant bioactives as anticancerous aid is progressing. In his research study, Singamaneni, *et al*. (2021) has evaluated the potential of methanolic extract, its chloroform fraction and compounds, coronarin K and L, firstly isolated from rhizomes of *R. purpurea*, along with other known compounds; coronarin A, bisdemethoxycurcumin and kaempferol 3-O-methyl for anti-cancer activity against human lung (A549), colon (HCT-116), breast (MCF-7) and pancreas (Bxpc-3) cancer cell lines by using MTT assay. Paclitaxel was used as standard (IC_{50} 6.2±0.20, 8.6±0.04 5.46±0.74 & 3.81±0.32 µM, respectively for A549, HCT-116, Bxpc-3 & MCF-7 cells). It was found that extracts and fraction exhibited anti-cancer effects against all tested cell lines with IC_{50} 21.35±0.83 to >100 µM when compared with paclitaxel. Moreover, coronarin K showed potent anti-cancer effect against A549, HCT-116, Bxpc-3 & MCF-7 cells with IC_{50} 13.49±0.62, 26.03±1.46, 56.70±2.17 & 56.24±0.83 µM, respectively when compared with paclitaxel. Furthermore, coronarin L and coronarin A displayed anti-cancer effects against A 549 cells with IC_{50} 33.78±1.37 & 61.80±2.82 µM, respectively while coronarin L against MCF-7 cells with IC_{50} 49.84±2.61 µM when compared with paclitaxel. Additionally, coronarin L, coronarin A and bisdemethoxycurcumin showed effects against Bxpc-3 cells with IC_{50} 56.83±1.92, 22.83±1.47 & 68.15±2.41 µM, respectively when compared with paclitaxel. These results revealed that *R. purpurea* possessed prominent anti-cancer effects. (Singamaneni *et al*., 2021). In a previous study, the ethanolic extract and its petroleum ether, chloroform, acetone, ethanolic and aqueous fractions from rhizomes of *R. purpurea* were also evaluated for anti-cancer potential against human lung carcinoma (A549), cervical cancer (SiHa), chinese hamster ovary cells (CHOK1) and rat glioma (C6) cell lines using SRB assay where vinblastine was established as control. It is noticed that the extract and fractions exhibited cytotoxic effect against all cell lines with IC_{50}<10 to >150 µg/ml (Srivastava *et al*., 2015).

Immunomodulatory activity

The ethanolic extract (300 & 600 mg/kg, p.o.) from the rhizomes was investigated for immunomodulatory activity by analyzing delayed type hypersensitivity (DTH) response and macrophage phagocytosis using carbon clearance assay in Swiss Albino mice. Cyclophosphamide (30 mg/kg) utilized as standard. It was observed that, the extract at both doses significantly ($p<0.05$) increased the foot pad thickness, WBC and total platelet count of anti-genically (SRBCs suspension) challenged mice when compared with control group. Moreover, extract at both doses significantly ($p<0.05$) increased the phagocytic index using carbon clearance method when compared with control group. Therefore, these results justified that the extract possessed immunostimulant properties (Sahu *et al*., 2010).

Why do we need to conserve *Roscoea purpurea* (Kakoli)?

India has ample resource of medicinal plants comprises of 8,000 diverse species. In the 21st century, there is a remarkably focus among individuals on health and environmental preservation, resulting in a significant increase in the utilization of medicinal plants. The substantial amount of raw material is derived from forest area. Therefore, the forest area has been exacerbated by the pharmaceutical and associated industries. Consequently, myriad number of plants have been threatened and some of them have been entailed in Red Data Book. *Roscoea purpurea* (Kakoli) is also among one of them, which is needed to be conserved and cultivated due to its excessive utilization (Kumar and Jnanesha, 2016; Virk *et al*., 2017).

There are two main strategies for the conservation: *in situ* and *ex situ* conservation. *In-*

Table 1: Chemical constituents of *Roscoea purpurea* in different plant parts

Chemical Constituents	Plant Parts	Extract	References
1. Phenolic compounds			
Protocatechuic acid	Tubers	Methanol/ Ethanol	(Mishra *et al*., 2015; Srivastava *et al*., 2015)
Ferulic acid	Tubers	Methanol/ Ethanol	(Mishra *et al*., 2015; Srivastava *et al*., 2015)
Syringic acid	Tubers	Methanol/Ethanol	(Mishra *et al*., 2015; Srivastava *et al*., 2015)
Apigenin	Tubers	Ethanol	(Srivastava *et al*., 2015)
Vanillic acid	Tubers	Methanol	(Mishra *et al*., 2015)
Gallic acid	Rhizomes	Methanol-Chloroform fraction	(Singamaneni *et al*., 2021)
Caffeic acid	Rhizomes	Methanol-Chloroform fraction	(Singamaneni *et al*., 2021)
Ellagic acid	Rhizomes	Methanol-Chloroform fraction	(Giri *et al*., 2017)
Ferulic acid	Rhizomes	Methanol-Chloroform fraction	(Singamaneni *et al*., 2021)
Fenozan acid	Rhizomes	Methanol-Chloroform fraction	(Singamaneni *et al*., 2021)
3-(3-methoxy,4-hydroxyphenyl) -2-propenoic acid ferulic acid	Rhizomes	Methanol-Chloroform fraction	(Singamaneni *et al*., 2021)
2. Flavonoids	Aerial parts/	Methanol	(Miyazaki *et al*., 2014)
Kaempferide	Rhizomes		
Kaempferol	Rhizomes/ Tubers	Methanol	(Singamaneni *et al*., 2021; Mishra *et al*., 2015)
Kaempferide 3-O-â-D-glucuronopyranoside	Aerial parts	Methanol	(Miyazaki *et al*., 2014)
Kaempferol 3-O-â-D-glucuronopyranoside	Aerial parts	Methanol	(Miyazaki *et al*., 2014)
Kaempferol 3-O-methyl ether	Rhizomes	Methanol	(Miyazaki *et al*., 2014; Singamaneni *et al*., 2021)
Quercetin	Rhizomes	Methanol	(Misra *et al*., 2015, Devkota and Timalsina, 2021)
Epicatechin	Roots	Methanol	(Kaur *et al*., 2020a)
Epigallocatechin	Roots	Methanol	(Kaur *et al*., 2020a)
Rutin	Rhizomes/ Tubers	Ethanol	(Giri *et al*., 2017; Srivastava *et al*., 2015)
6-methyl pinocembrin	Tubers	Ethanol	(Chaudhari, 1988)
3. Curcuminoid			
Bisdemethoxycurcumin	Rhizomes	Methanol-Chloroform fraction	(Singamaneni *et al*., 2021)
4. Terpenoids			
Coronarin A (Labdanediterpenes)	Rhizomes	Methanol-Chloroform fraction	(Singamaneni *et al*., 2021)
Coronarin K	Rhizomes	Methanol-Chloroform fraction	(Singamaneni *et al*., 2021)
Coronarin L	Rhizomes	Methanol-Chloroform fraction	(Singamaneni *et al*., 2021)
Sterols			
Stigmasterol	Leaves	Ethyl acetate	(Barai and Bag, 2019)
Beta-sitosterol	Tubers	Ethanol	(Chaudhari, 1988)
Beta-sitosterol-beta-D-glucoside	Tubers	Ethanol	(Chaudhari, , 1988)
Triterpenoids			
Lupenone	Roots	Methanol	(Kaur *et al*., 2020b)
Ursolic acid	Tubers	Ethanol	(Chaudhari, 1988)
5. Miscellaneous			
Adenosine	Rhizomes	Methanol	(Miyazaki *et al*., 2014)
(Z)-3-hexen-l-ol-â-D-glucopyranoside	Aerial parts	Methanol	(Miyazaki *et al*., 2014)
n-hentriacontane	Tubers	Ethanol	(Chaudhari, 1988)

Table 2: Medicinal uses and formulations of *Roscoea purpurea*

Traditional Uses	Preparation/Ayurvedic formulations	References
Abdominal disorders	Whole plant*:* Processed with *Vacâdi* oil and other herbs is used as an *anuvâsanavasti* to treat distension, dyspepsia and *gulma.*	(Rao, 2011)
Cardio-vascular disorders Cardiac diseases Chest injury	Roots: Powder with other herbs as *Crepidium acuminatum*, *Malaxis muscifera*, *Pueraria tuberosa*, *Crepidium acuminatum*, *Malaxis muscifera*, *Polygonatum verticillatum*, *Pueraria tuberosa*, *Medâ*, *Mahâmedâ* mixed with honey. *Nâgabalâsarpi* processed with *Roscoea purpurea* and other herbs are useful in chest injury	(Balkrishna, 2014)
Cuts and wounds	Leaves and roots: Dried powder	(Pande, Tiwari, and Pande, 2006; Bisht, 2017).
Diabetes	Leaves, roots, rhizomes and flowers: Infusion/ paste.	(Misra *et al*., 2015; Singamaneni, 2021; Seth and Kondal, 2020).
Diarrhea and dysentry	Leaves, roots, rhizomes and flowers: Infusion/ Decoction.	(Seth and Kondal, 2020; Misra *et al*., 2015; Singamaneni, *et al*., 2021)
Gout	Whole plant: Decoction with *Prunus cerasoides*, *Glycyrrhiza glabra*, *Curcuma longa*, *Vetiveria zizanioides*, *Rubia cordifolia*, *Lilium polyphyllum*, is prepared as a paste, processed with oil and made into a *taila* known as *Khuâkapadmataila,* useful in treating gout and burning sensation.	(Tripathi and Pandey, 2012)
Hemorrhage	Roots: Powder helps to treat bleeding disorders. Whole plant: Powder processed with *Nâgabalâsarpi* and other herbs are useful in intrinsic haemorrhage.	(Balkrishna, 2014)
Paediatric diseases	Whole plant: powder, processed with *Ghrta* (clarified butter) and *Cedrus deodara, Lilium polyphyllum*, *Crepidium acuminatum*, *Malaxis muscifera*, *Vigna trilobata* and *Teramnus labialis* is useful in appropriate doses in case of emaciation in children.	(Balkrishna, 2014)
Respiratory disorders	Tubers: Paste.	(Swar, 2014)
Cold and cough	Whole Plant: Powder with other herbs and honey**.**	(Balkrishna, 2014)
Bronchitis and asthma	Rhizome: Powder mixed with orange rind powder	(Balkrishna, 2014)
Reproductive disorders Impotency	Roots and Rhizomes: Decoction	(Kaur *et al*., 2017; Seth and Kondal, 2020)
Sinusitis	Whole plant : Powder processed with *Ghrta* and herbs as *Glycyrrhiza-glabra, Curcuma longa*, *Polygonatum verticillatum*, *Nelumbo nucifera*, used as *parishechana* (irrigative therapy) in *pittaja* associated erysipelas and sinus.	(Rao, 2011).
Skin disorders	Roots: powder, mixed with black pepper and rat dropping, applied to boils.	(Singh, 2009; Quattrocchi, 2012).
Tuberculosis	Whole plant: Powder processed with *Nâgabalâsarpi* and other herbs.Bulbs are used to treat phthisis.	(Balkrishna, 2014; Saroya, 2013).
Urinary disorders	Roots: Extract as a tonic; ingested to treat urinary infection and polyuria	(Misra *et al*., 2015; Miyazaki *et al*., 2014)

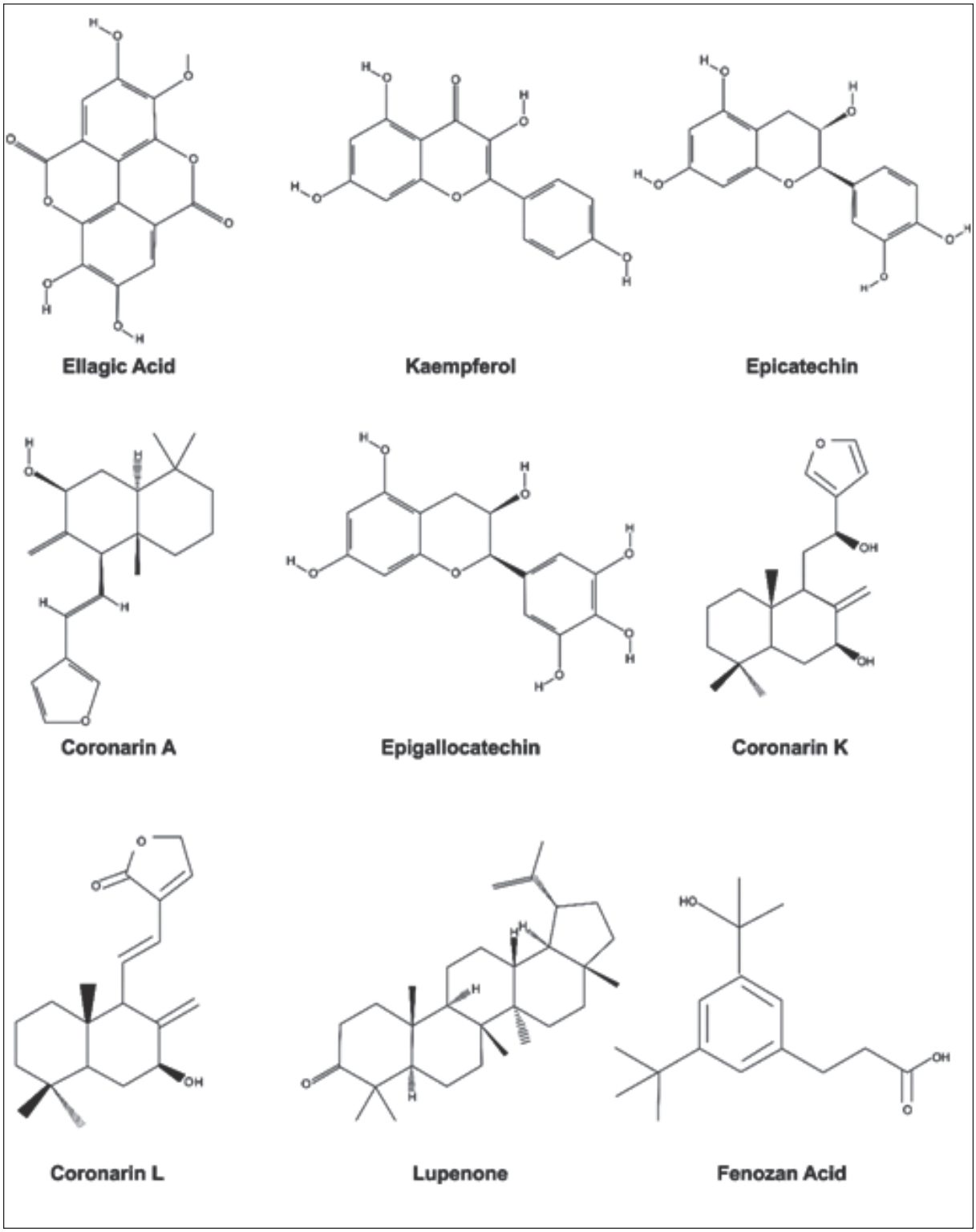

Fig. 1: Chemical structures of *R. purpurea* compounds

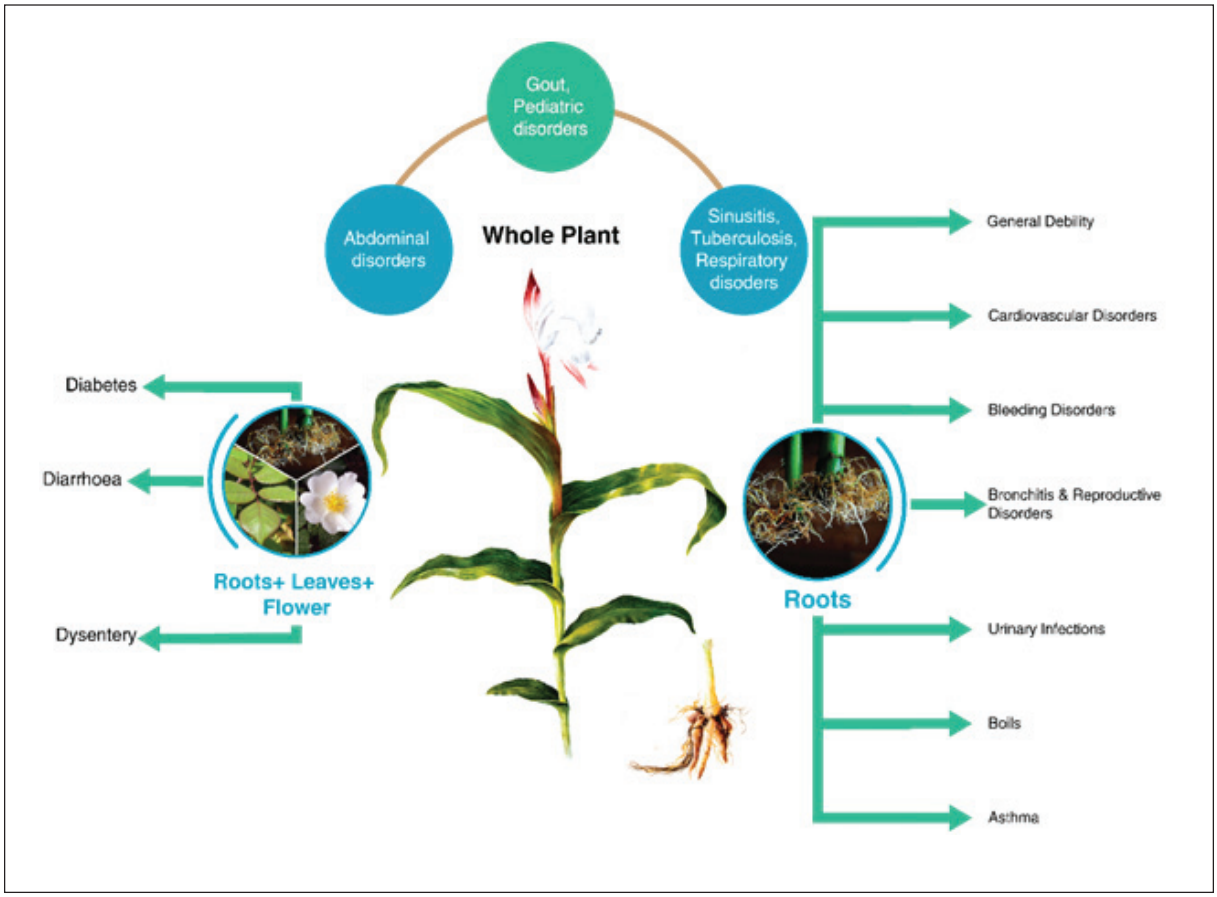

Fig. 2 : Medicinal Properties of Kakoli (*R. purpurea*).

situ strategy of conservation focuses on the "on-site conservation" of the wild genetic diversity in natural environment**.** It is a methodological technique that entails to conserve such endangered plant species with indispensable therapeutic value (Chandra, 2016). The involvement of community is to preserve both threatened plant species as well as their natural communities. This procedure adopts an eco-centric strategy for conservation, where preserving endangered plant species is prioritized while minimizing human intervention (Akshay *et al*., 2014). There are two essential segment of this technique, firstly, is to explore for the geographical location of these plant species based on their therapeutic properties and industrial value. Secondly, to monitor, manage, and preserve such plant species by forming a network and zone of forest habitat of a specified size that must be established (Dhama *et al*., 2018). In addition to this, the significant aspect of this method is to investigate and survey of such plant after a regular interval of time that may leads to contribute better perception of natural biodiversity and various methods of preservation of such plants. This type of well-organised conservation strategy can evoke recovery of endangered plant species.

There are several *in-situ* conservation methods for medicinal plants such as biosphere reserve, national park, sacred groves and others**.** The Ministry of Environment and Forest, Government of India had identified 13 biosphere reserve for the conservation of medicinal plants in different states of India (Kadam and Pawar, 2020). In West Bengal and Tamil Nadu have established biospheres named Sunderban and Gulf of Mannar respectively for the protection medicinal plant species (Chandra, 2016). In India, there are 91 national parks, 2 out of them namely,Pin Valley National Park and Great Himalayan National Park in Himachal Pradesh have been established. Unluckily, a substantial population live in proximity to these forests (Chandra, 2016). The establishment of national park

create the involvement of local people which generate responsibility towards them to protect medicinal plant species (Huang *et al*., 2002). People who live near to forests area can help in ensuring the survival of endangered medicinal plant species only if they are made aware of conservation methods.

Sacred groves are said to be small or substantial areas of vegetation that are protected on the basis of tradition and religious beliefs. In India there are 13,720 small scale sacred groves through which medicinal plant species is protected (Murtem and Chaudhry, 2014). Manipur is the state reported over 365 sacred groves. These groves persist significant ecologically value because they link ritual behavior to ecology, especially in order to protect threatened plant species. Traditional agricultural methods are eco-friendly and long-lasting approach which supports to conserve the diversity of medicinal plant species (Ahmad *et al*., 2021). The native farmers who live near the forest region have gathered more knowledge about medicinal plant (Benniamin, 2011) and their expertise should be organized through appropriate documentation. We can disseminate the garden conservation for the protection of medicinal plants in small apartments, ûats, house and bungalows. For the conservation of medicinal plant species proper monitoring is needed in above mentioned protected areas. People may be driven to support conservation efforts *via* financial incentives.

Ex-situ strategy of conservation focuses on the "off-site conservation" of the wild genetic diversity in natural environment. It is a pivotal procedure for the protection of medicinal plant species that are high risk of extinction (Werden *et al*., 2020). *Ex situ* conservation objective to reintroduce the endangered species into their native habitat in order to secure their survival and periodically to produce substantial amount of planting resources used in the formation of therapeutic drugs. In recent years, the government promotes long-term plant species conservation through parks and botanical gardens. When the medicinal plant species were grown away from their natural habitat, not only they retain their high efficacy but their reproductive parts are extracted and maintained in the repository in seed banks for future regeneration (Kumar and Jnanesha, 2016). There are several *ex-situ* conservation methods for medicinal plants such as seed bank, botanical garden etc.

In India, one of the Institute named Tropical Botanical Garden and Research Institute (TBGRI) located in Kerala has taken step towards the conservation and sustainable use of medicinal plant wealth of peninsular India. The TBGRI performs major activities such as seed gene bank, *in-vitro* gene bank and field gene bank to protect medicinal plant species (Bhattacharyya *et al*., 2006). Botanical garden also plays a vital role sustaining the ecosystem to escalate the survival of rare and endangered plant species. They include a large diverse variety of plant species that share common condition. Botanical garden act as center for release, revive and rehabilitation of rare, endangered and extinct prone species of medicinal plants and other significant plant genetic resources. Apart from this, it plays significant function in education and training center sector such as horticulture, gardening, landscaping, *ex-situ* conservation and environmental awareness. A case study was done to elucidate the medicinal plant conservation through botanical garden where in Malabar Botanic Garden of India is used to train local farmers for the cultivation and repository methods of medicinal plants moreover giving market guidance. In 2 years, they have trained more than 200 farmers, who later established a farmers' association to exchange knowledge (Hawkins, 2007). Botanical gardens have been built in several regional communities. Example: Government Botanical Gardens in Tamil Nadu, Garden of Medicinal Plants in West Bengal and many more across the country (Kadam and Pawar, 2020).

CONCLUSION

The rhizomes, tubers and other parts of *R. purpurea* are well described in various Ayurvedic texts and literatures as an analeptic, widely used for treating various health ailments in Nepal and India. Despite the wealth of historical data supporting its traditional applications, there exists a shortage of comprehensive studies validating these assertions. While existing publications have demonstrated the plant's potential as a natural antioxidant and cytostatic agent, there remains a dearth of empirically grounded literature designating it as a promising candidate for future

drug discovery initiatives. The phytochemical profiling of the plant has revealed the presence of flavonoids and phenolic acids, known for their robust bioactive attributes. As such, it is advisable to investigate the individual compounds and their synergistic effects to determine optimal formulations and dosages. Notably, compounds like stigmasterol, due to their nanoscale dimensions, hold promise for advancement in nanobiotechnology research. To lay the groundwork for forthcoming research endeavors, an increased number of both *in-vivo* and *in-vitro* studies are imperative. Rigorous clinical trials are essential to substantiate the therapeutic efficacy of this traditional medicine within the conventional medical framework. Addressing these research gaps should be a focal point for future investigations, guided by the imperative to conserve these valuable botanical resources.

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Gas exchange parameters and chlorophyll content as influenced by different chemicals and planting materials in pomegranate (*Punica granatum* L.) cv. Bhagwa

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ABSTRACT

Soil drenching with paclobutrazol @ 0.375 g a.i. m^{-1} canopy diameter 60 days after bahar treatment to tissue culture plants improved gas exchange parameters viz., photosynthetic rate (P_N) at both flowering (17.15 µmol m^{-2} s^{-1}) and fruit set stages (11.62 µmol m^{-2} s^{-1}), transpiration rate (E) at both flowering (7.16 mmol m^{-2} s^{-1}) and fruit set stages (4.27 mmol m^{-2} s^{-1}), stomatal conductance (g_s) at both flowering (0.53 mol m^{-2} s^{-1}) and fruit set stages (0.28 mol m^{-2} s^{-1}). High total chlorophyll content was observed due to soil drenching of paclobutrazol @ 0.375 g a.i. m^{-1} canopy diameter 60 days after bahar treatment to tissue culture plants at both flowering (3.03 mg g^{-1}) and fruit set (2.00 mg g^{-1}) stages. Foliar spray of nitrobenzene @ 2.0 ml $litre^{-1}$ to tissue culture plants improved gas exchange parameters viz., photosynthetic rate (P_N) at both flowering (17.52 µmol m^{-2} s^{-1}) and fruit set (14.05 µmol m^{-2} s^{-1}) stages. While transpiration rate (E) was high in tissue culture plants due to soil drenching of paclobutrazol @ 0.375 g a.i. m^{-1} canopy diameter 45 days after bahar treatment at flowering stage (8.01 mmol m^{-2} s^{-1}) while at fruit set stage, soil drenching of paclobutrazol @ 0.375 g a.i. m^{-1} canopy diameter 60 days after bahar treatment to tissue culture plants registered highest transpiration rate (E) (4.07 mmol m^{-2} s^{-1}). Highest stomatal conductance (g_s) (0.46 mol m^{-2} s^{-1}) was observed due to foliar spray of nitrobenzene @ 2.0 ml $litre^{-1}$ to tissue culture plants at flowering stage while at fruit set stage, the same chemical registered higher value (0.26 mol m^{-2} s^{-1}) in grafted plants. High total chlorophyll content was observed due to foliar spray of nitrobenzene @ 2.0 ml $litre^{-1}$ to tissue culture plants at both flowering (3.07 mg g^{-1}) and fruit set stages (2.94 mg g^{-1}).

***Keywords*:** Gas exchange parameters, nitrobenzene, paclobutrazol, photosynthesis, pomegranate

INTRODUCTION

Sunlight is the primary source of energy for photosynthesis, although air temperature and humidity have an impact on transpiration. Transpiration is caused by the drying power of the atmosphere, which depends on wind speed and relative humidity, as well as the evaporative demand from net radiation absorbed by leaves (Elanchezian *et al*., 2015). By allowing CO_2 and water vapour to pass into and out of the leaf, stomata play a significant role in controlling transpiration and photosynthesis (Mokhles *et al*., 2019). According to Monerri *et al*. (2011), stomatal aperture control is influenced by state variables (such as leaf water potential and intercellular carbon dioxide concentration), the interaction of processes (transpiration and photosynthetic rates), and environmental factors (specifically, the difference in water vapour concentration between the leaf surface and the bulk air).

The amount of chlorophyll in the plant, together with the photosynthetic rate (P_N), transpiration (E), stomatal conductance (g_s), and other factors, all affect the metabolic activity of the plant and its overall growth and development. There have been extensive studies conducted on a variety of fruit

crops, including mango, citrus, apple, sweet cherry, etc. (Jones, 1992). In an experiment with olive, Arun *et al*. (2017) looked at how plant growth regulators affected gas exchange and chlorophyll content.

For the first time in pomegranate, studies on gas exchange parameters—photosynthetic rate (P_N), transpiration (E), and stomatal conductance (g_s)—as well as chlorophyll content were carried out to examine the metabolic activity of the plant using chemicals like methyl jasmonate, nitrobenzene, and paclobutrazol at different concentrations.

MATERIALS AND METHODS

The current study was carried out at the research farm of ICAR-Indian Institute of Horticultural Research (IIHR) in Hesaraghatta, Bengaluru during the *ambe bahar* (January–February) and *hastha bahar* (September–October) seasons of 2020 and 2021. As noted before, plants were multiplied from three different sources: air layers (P_3), grafted plants (P_2), and tissue culture plants (P_1). During *Ambe bahar,* the average maximum and minimum temperatures of 33.08^{0}C and 20.43^{0}C, were recorded respectively, and the relative humidity was 75.04 percent and 59.06 percent and the total rainfall was 74.95 mm of 12.10 mm, respectively. During *Hastha bahar,* the average maximum and minimum temperatures of 26.13^{0}C and 18.94^{0}C were recorded respectively and relative humidity of 59.06 percent and the total rainfall was 12.10 mm, respectively. A ten-tree factorial randomized block statistical design was employed. The study included three distinct concentrations of MeJA (Methyl Jasmonate), NB (Nitrobenzene), and one concentration of PBZ (Paclobutrazol) delivered 30 days, 45 days, or 60 days after the 'bahar treatment'. The treatments were expressed as follows according to the concentration of a certain growth regulator: T_1: 100 ppm MeJA, T_2: 150 ppm MeJA, T_3: 200 ppm MeJA, T_4: 1.0 ml NB, T_5: 1.5 ml NB, T_6: 2.0 ml NB, T_7: Soil drenching of paclobutrazol @ 0.375g a.i. m^{-1} canopy diameter 30 days after bahar treatment, T_8: Soil drenching of paclobutrazol @ 0.375g a.i. m^{-1} canopy diameter 45 days after bahar treatment, T_9: Soil drenching of paclobutrazol @ 0.375g a.i. m^{-1} canopy diameter 60 days after bahar treatment, T_{10}: Water spray.

Gas exchange parameters

The Portable Photosynthesis System (LCpro+, ADC BioScientific Limited, UK) was used to assess the rates of photosynthetic activity (P_N), transpiration (E), and stomatal conductance (g_s). Fully developed leaves were measured at least three times between 9:30 and 11:30 am while they were exposed to ambient light and CO_2 levels.

Total chlorophyll content

With the aid of acetone and dimethyl sulphoxide (DMSO), the complete chlorophyll content was removed. A 0.1 g leaf sample was dissolved in 10 ml of DMSO: Acetone reagent (1:1) and let to sit for 72 hours in the dark. Using a UV-VIS spectrophotometer (T80+ UV/VIS spectrometer, PG Instrument Ltd., UK), the extract was collected and the absorbance measured at 663 nm and 645 nm for quantification of chlorophyll a, chlorophyll b, and total chlorophyll. Additionally, using the equations put out by Lichtenthaler and Buschmann (2001), the pigment contents were computed and given in mg g^{-1}.

$$\text{Chlorophyll } a = [12.7\ (A_{663}) - 2.69\ (A_{645})]\ \frac{V}{1000 \times W \times a}$$

$$\text{Chlorophyll } b = [22.9(A_{645}) - 4.68\ (A_{663})]\ \frac{V}{1000 \times W \times a}$$

$$\text{Total chlorophyll} = [20.2\ (A_{645}) + 8.02\ (A_{663})]\ \frac{V}{1000 \times W \times a}$$

Where,

A = Absorbance at specific wave lengths (645 nm and 663 nm)

V = Volume of the extract (10 ml)

W = Fresh weight of the sample (100 mg)

a = path length of light in cuvette (1 cm)

Statistical analysis

The method of evaluating variance developed by Panse and Sukhatme (2005) was used to analyze the data. The F value at a 5% level of significance was used to determine the statistical significance. Critical differences were calculated at the 0.05 level to determine if there were any significant effects.

RESULTS AND DISCUSSION

Rate of photosynthesis

The rate of photosynthesis (P_N) in tissue-culture plants was considerably impacted by the flowering stage of *ambe bahar* and reached its peak (14.24 mol m^{-2} s^{-1}) in P_1 (i.e., in tissue-culture plants) (Table 1). Paclobutrazol, one of the chemicals, produced a P_N of 14.71 mol m^{-2} s^{-1} 60 days after the bahar treatment (T_9), which was comparable to those obtained from the same treatment 45 days after the bahar treatment (T_8) (13.89 mol m^{-2} s^{-1}) and nitrobenzene 1.5 ml $litre^{-1}$ (T_5) or 2.0 ml $litre^{-1}$ (T_6). P_1 with paclobutrazol 45 days (T_8) or 60 days (T_9) after the bahar treatment registered comparable values of P_N, whereas higher values of P_N (15.46 mol m^{-2} s^{-1}) were recorded in grafted plants (P_2) sprayed with nitrobenzene 2.0 ml $litre^{-1}$ (T_6). These differences in PN were also significant between the types of propagules and chemical combinations.

In comparison to tissue-culture plants or grafts, air layers in ambe bahar displayed significantly higher P_N values at the fruit set stage. Paclobutrazol among the chemicals showed equivalent values 45 or 60 days after the bahar treatment, which were considerably different from those observed in the other treatments. In the combinations, air layers treated with paclobutrazol 45 or 60 days after the bahar treatment or with nitrobenzene 2.0 ml $litre^{-1}$ produced outcomes that were equivalent and had higher P_N values than nitrobenzene 2.0 ml $litre^{-1}$ sprayed grafted plants (P_2) did. (Table 1)

According to the data in Table 1, all of the propagules at the flowering stage of *hastha bahar* varied significantly in terms of P_N, whose values in tissue-culture plants (P_1) (14.33 mol m^{-2} s^{-1}) and in air layers were comparable but different from those in the grafted plants. Nitrobenzene 2.0 ml $litre^{-1}$ (T_6) was one of the compounds that produced results that were noticeably greater than those from the other treatments (16.40 mol m^{-2} s^{-1}). Tissue-culture plants (P_1) treated with paclobutrazol or nitrobenzene 2.0 ml $litre^{-1}$ 60 days after the bahar treatment, or both, reported comparable P_N values according to the propagule and chemical treatment combination. During the fruit set stage in *hastha bahar*, neither the different types of propagules nor their combinations with the chemicals showed any significant differences in terms of P_N. Among the chemicals, nitrobenzene 1.5 ml $litre^{-1}$ or 2.0 ml $litre^{-1}$ $plant^{-1}$showed comparable values of P_N significantly different from those seen with any of the other chemical treatments. (Table 1)

Regardless of the form of propagule or season, we observed a higher rate of photosynthesis, i.e. P_N, during the initial stage (flowering stage), which gradually declined at the fruit set stage. The higher rate was most likely due to the plant's reproductive structures' increased demand for assimilates, which is consistent with Drogoudi *et al*., (2012) findings in pomegranate. In the current study, (P_N) was high in all propagules during flowering and gradually decreased during fruit set, owing to strong sinks such as developing fruits, which deplete carbohydrates in the source, primarily leaves (Mokhles *et al*., 2019). During flowering, increased rates of photosynthesis were also observed in mango (Laurent *et al.* 2008). Increased demand for photosynthates during flower initiation and growth (Wunsche *et al*., 2005) was reported to increase photosynthesis rates in supporting leaves of a branch in apples (Wahl *et al*., 2013), which was also attributed to increased demand from sinks in the form of developing flowers. Treatment with PBZ could also have resulted in a higher photosynthetic rate. Treatment with PBZ increases the chlorophyll content in wheat and potato, which is a crucial component for photosynthesis, and this may be one mechanism through which PBZ improved photosynthesis in pomegranates (Kishore *et al*. 2006; Nouriyani *et al*. 2012). Tissue culture plants treated with PBZ also had higher transpiration rates and stomatal conductance.

Rate of transpiration

At the flowering stage in *ambe bahar*, as can be seen from the data presented in Table 2, plants raised from different types of propagules differed

Table 1:Rate of photosynthesis (µmol m^{-2} s^{-1}) in pomegranate as influenced by different planting materials and chemicals

	Flowering stage								Fruit-set stage							
	ambe bahar				*hastha bahar*				*ambe bahar*				*hastha bahar*			
Propagules	**P_1**	**P_2**	**P_3**	**Mean**	**P_1**	**P_2**	**P_3**	**Mean**	**P_1**	**P_2**	**P_3**	**Mean**	**P_1**	**P_2**	**P_3**	**Mean**
T_1	12.76	10.75	10.81	**11.44**	13.68	13.38	12.67	**13.24**	9.59	9.36	11.07	**10.00**	10.25	11.62	12.45	**11.30**
T_2	13.08	11.32	13.47	**12.62**	14.49	12.96	15.65	**14.36**	9.72	9.47	11.66	**10.28**	11.01	11.13	9.69	**10.61**
T_3	13.32	11.61	13.76	**12.89**	13.15	14.07	15.39	**14.20**	9.83	9.92	11.45	**10.40**	9.43	10.67	11.64	**10.58**
T_4	14.15	14.03	9.13	**12.44**	13.86	14.38	14.25	**14.16**	8.74	10.75	11.51	**10.03**	10.26	12.29	13.57	**12.04**
T_5	14.43	14.83	12.87	**14.04**	13.11	13.95	15.52	**14.19**	10.61	12.53	10.53	**10.48**	12.99	13.41	13.79	**13.39**
T_6	14.50	15.46	12.41	**14.12**	17.52	15.29	16.41	**16.40**	11.22	13.22	13.14	**11.74**	14.05	15.43	14.64	**14.71**
T_7	15.10	11.73	11.40	**12.74**	14.30	15.17	13.97	**14.48**	11.32	9.85	12.38	**11.45**	11.09	10.95	10.90	**10.98**
T_8	16.09	12.69	12.88	**13.89**	14.94	12.48	12.86	**13.43**	11.37	10.32	13.07	**12.27**	12.05	11.65	9.03	**10.91**
T_9	17.15	13.16	13.83	**14.71**	16.15	13.98	12.04	**14.06**	11.62	10.61	13.77	**12.87**	9.48	11.74	11.33	**10.85**
T_{10}	11.82	10.92	11.32	**11.35**	12.10	11.69	11.73	**11.84**	8.44	9.06	10.01	**9.17**	9.37	8.54	10.18	**9.37**
Mean	**14.24**	**12.65**	**12.19**		**14.33**	**13.73**	**14.05**		**10.24**	**10.51**	**11.86**		**11.00**	**11.74**	**11.72**	
	P	T	P × T		P	T	P × T		P	T	P × T		P	T	P × T	
SE(m)	0.19	0.36	0.62		0.16	0.30	0.52		0.12	0.23	0.39		0.26	0.47	0.82	
C.D. (5%)	0.56	1.03	1.78		0.47	0.86	1.49		0.35	0.65	1.12		N.S	1.35	N.S	

P1: tissue-culture plants; **P2**: grafted plants; **P3**: Air layer plants

T1: Methyl jasmonate (MeJA) 100 ppm; **T2**: MeJA 150 ppm; **T3**: MeJA 200 ppm; **T4**: Nitrobenzene (NB) 1.0 ml l^{-1}; **T5**: NB 1.5 ml l^{-1}, **T6**: NB 2.0 ml l^{-1}; **T7**: paclobutrazol (PBZ) 0.375 g of active ingredient per metre of canopy diameter applied 30 days after the 'bahar' treatment; **T8**: same as T7 except applied 45 days after the treatment; **T9**: same as T7 except applied 60 days after the treatment; **T10**: Control; sprayed with water after the 'bahar' treatment

Table 2: Rate of transpiration (mmol m^{-2} s^{-1}) in pomegranate as influenced by different planting materials and chemicals

	Flowering stage								Fruit-set stage							
	ambe bahar				*hastha bahar*				*ambe bahar*				*hastha bahar*			
Propagules	**P_1**	**P_2**	**P_3**	**Mean**	**P_1**	**P_2**	**P_3**	**Mean**	**P_1**	**P_2**	**P_3**	**Mean**	**P_1**	**P_2**	**P_3**	**Mean**
T_1	6.52	4.57	2.50	**4.53**	3.83	4.89	4.39	**5.10**	3.70	3.95	2.96	**3.65**	2.68	2.89	3.18	**2.91**
T_2	5.21	5.88	2.84	**4.64**	4.28	4.01	4.14	**5.06**	3.82	4.01	3.38	**3.82**	3.23	3.54	2.52	**3.09**
T_3	4.66	5.91	2.98	**4.51**	4.56	3.91	4.52	**5.21**	3.84	4.13	3.48	**3.93**	2.89	2.83	2.76	**2.83**
T_4	7.01	5.59	3.40	**5.33**	3.94	4.17	5.96	**5.42**	3.52	4.61	3.02	**3.48**	3.82	3.1	3.34	**3.42**
T_5	6.45	5.67	3.58	**5.23**	4.70	6.71	5.90	**5.54**	3.57	5.10	3.26	**3.44**	3.27	3.49	3.54	**3.43**
T_6	5.89	6.10	5.47	**5.82**	5.05	6.28	6.95	**5.80**	3.86	5.40	3.51	**4.08**	3.87	4.18	3.8	**3.95**
T_7	7.16	6.55	3.51	**5.74**	4.54	5.07	4.38	**5.93**	3.92	4.02	3.99	**4.29**	3.48	3.66	3.01	**3.38**
T_8	7.85	6.66	3.50	**6.00**	5.07	8.01	5.43	**5.09**	4.02	4.24	4.35	**4.36**	3.47	3.71	2.43	**3.2**
T_9	8.01	7.16	3.52	**6.23**	5.41	7.62	4.80	**5.51**	4.27	4.31	4.59	**4.37**	4.07	3.61	3.26	**3.65**
T_{10}	4.29	3.71	2.48	**3.49**	3.48	4.90	3.73	**4.13**	3.22	3.90	2.85	**3.50**	3.03	2.46	2.48	**2.65**
Mean	**6.30**	**3.38**	**4.30**		**5.56**	**5.02**	**5.28**		**3.77**	**4.37**	**3.54**		**3.38**	**3.35**	**3.03**	
	P	T	P × T		P	T	P × T		P	T	P × T		P	T	P × T	
SE(m)	0.10	0.18	0.31		0.10	0.18	0.32		0.07	0.13	0.23		0.10	0.18	0.32	
C.D. (5%)	0.28	0.52	0.90		0.28	0.52	0.9		0.21	0.39	0.67		0.28	0.52	N.S	

P_1: tissue-culture plants; **P_2**: grafted plants; **P_3**: Air layer plants

T_1: Methyl jasmonate(MeJA) 100 ppm; **T_2**: MeJA 150 ppm; **T_3**: MeJA 200 ppm; **T_4**: Nitrobenzene (NB) 1.0 ml $litre^{-1}$; **T_5**: NB 1.5 ml $litre^{-1}$, **T_6**: NB 2.0 ml $litre^{-1}$; **T_7**: paclobutrazol (PBZ) 0.375 g of active ingredient per metre of canopy diameter applied 30 days after the 'bahar' treatment; **T_8**: same as T_7 except applied 45 days after the treatment; **T_9**: same as T_7 except applied 60 days after the treatment; **T_{10}**: Control; sprayed with water after the 'bahar' treatment

Table 3: Stomatal conductance (mol m^{-2} s^{-1}) in pomegranate as influenced by different planting materials and chemicals

	Flowering stage								Fruit set stage							
	ambe bahar				*hastha bahar*				*ambe bahar*				*hastha bahar*			
Propagules	**P_1**	**P_2**	**P_3**	**Mean**	**P_1**	**P_2**	**P_3**	**Mean**	**P_1**	**P_2**	**P_3**	**Mean**	**P_1**	**P_2**	**P_3**	**Mean**
T_1	0.25	0.18	0.18	**0.20**	0.29	0.27	0.26	**0.27**	0.19	0.19	0.13	**0.20**	0.14	0.17	0.19	**0.17**
T_2	0.27	0.23	0.21	**0.23**	0.25	0.31	0.24	**0.26**	0.24	0.21	0.14	**0.23**	0.14	0.16	0.17	**0.15**
T_3	0.28	0.27	0.23	**0.26**	0.26	0.24	0.27	**0.26**	0.26	0.24	0.17	**0.25**	0.16	0.12	0.17	**0.15**
T_4	0.34	0.30	0.25	**0.30**	0.23	0.31	0.33	**0.29**	0.21	0.28	0.18	**0.21**	0.17	0.16	0.17	**0.16**
T_5	0.35	0.32	0.27	**0.31**	0.32	0.34	0.36	**0.34**	0.22	0.30	0.21	**0.24**	0.17	0.16	0.23	**0.19**
T_6	0.37	0.34	0.28	**0.33**	0.46	0.40	0.38	**0.41**	0.23	0.32	0.22	**0.24**	0.24	0.26	0.26	**0.25**
T_7	0.42	0.19	0.30	**0.30**	0.28	0.18	0.28	**0.25**	0.26	0.26	0.22	**0.23**	0.16	0.12	0.14	**0.14**
T_8	0.44	0.20	0.31	**0.32**	0.31	0.25	0.26	**0.27**	0.26	0.27	0.25	**0.24**	0.19	0.15	0.13	**0.16**
T_9	0.53	0.24	0.32	**0.36**	0.37	0.24	0.23	**0.28**	0.28	0.29	0.26	**0.25**	0.19	0.14	0.15	**0.16**
T_{10}	0.20	0.16	0.13	**0.16**	0.21	0.21	0.22	**0.21**	0.16	0.21	0.13	**0.17**	0.11	0.12	0.15	**0.13**
Mean	**0.34**	**0.24**	**0.25**		**0.30**	**0.27**	**0.28**		**0.23**	**0.26**	**0.19**		**0.16**	**0.16**	**0.17**	
	P	T	P × T		P	T	P × T		P	T	P × T		P	T	P × T	
SE(m)	0.01	0.01	0.03		0.08	0.01	0.02		0.07	0.01	0.02		0.07	0.01	0.02	
C.D. (5%)	0.02	0.05	0.08		N.S	0.04	0.07		0.01	0.03	0.06		N.S	0.01	N.S	

P_1: Tissue-culture plants; **P_2**: grafted plants; **P_3**: Air layer plants

T_1: Methyl jasmonate (MeJA) 100 ppm; **T_2**: MeJA 150 ppm; **T_3**: MeJA 200 ppm; **T_4**: Nitrobenzene (NB) 1.0 ml $litre^{-1}$; **T_5**: NB 1.5 ml $litre^{-1}$, **T_6**: NB 2.0 ml $litre^{-1}$;**T_7**, paclobutrazol (PBZ) 0.375 g of active ingredient per metre of canopy diameter applied 30 days after the 'bahar' treatment; **T_8**, same as T_7 except applied 45 days after the treatment; **T_9**, same as T_7 except applied 60 days after the treatment; **T_{10}**, Control; sprayed with water after the 'bahar' treatment

Table 4: Total chlorophyll content (mg g^{-1} fresh leaves) in pomegranate as influenced by different planting materials and chemicals

	Flowering stage								Fruit set stage							
	ambe bahar				*hastha bahar*				*ambe bahar*				*hastha bahar*			
Propagules	**P_1**	**P_2**	**P_3**	**Mean**	**P_1**	**P_2**	**P_3**	**Mean**	**P_1**	**P_2**	**P_3**	**Mean**	**P_1**	**P_2**	**P_3**	**Mean**
T_1	1.37	1.44	1.73	**1.51**	1.75	1.53	1.88	**1.72**	1.54	1.35	1.51	**1.46**	1.37	1.25	1.65	**1.42**
T_2	1.66	1.58	1.95	**1.73**	1.86	1.72	1.52	**1.70**	1.60	1.26	1.60	**1.48**	1.65	1.49	1.29	**1.47**
T_3	1.96	1.65	1.81	**1.81**	1.93	2.63	1.47	**2.01**	1.65	1.16	1.55	**1.45**	1.70	2.29	1.26	**1.75**
T_4	2.36	1.94	1.83	**2.04**	2.82	2.84	2.12	**2.59**	1.68	1.43	1.67	**1.59**	2.55	2.58	1.58	**2.23**
T_5	2.20	2.16	2.15	**2.17**	2.64	3.00	2.49	**2.71**	1.60	1.44	1.68	**1.57**	2.48	2.68	2.10	**2.42**
T_6	2.17	2.23	2.35	**2.25**	3.07	3.24	2.75	**3.02**	1.91	1.54	1.69	**1.71**	2.94	2.95	2.06	**2.65**
T_7	2.40	1.69	1.73	**1.94**	2.04	2.48	1.79	**2.11**	1.96	1.13	1.70	**1.59**	2.04	2.05	1.31	**1.80**
T_8	2.62	1.77	1.60	**1.99**	2.32	2.07	2.45	**2.28**	1.97	1.19	1.80	**1.65**	1.96	1.77	1.19	**1.64**
T_9	3.03	1.81	2.47	**2.43**	3.02	2.57	1.95	**2.51**	2.00	1.40	1.90	**1.76**	2.81	2.14	1.60	**2.18**
T_{10}	1.64	1.19	1.27	**1.36**	1.59	1.57	1.52	**1.56**	1.26	0.92	1.43	**1.20**	1.42	1.30	1.10	**1.27**
Mean	**2.14**	**1.74**	**1.89**		**2.30**	**2.36**	**1.99**		**1.72**	**1.28**	**1.65**		**2.09**	**2.05**	**1.51**	
	P	T	P × T		P	T	P × T		P	T	P × T		P	T	P × T	
SE(m)	0.05	0.1	0.17		0.05	0.10	0.18		0.02	0.04	0.07		0.05	0.09	0.16	
C.D. (5%)	0.15	0.28	0.49		0.16	0.3	0.52		0.06	0.12	N.S		0.14	0.26	0.45	

P_1: tissue-culture plants; **P_2**: grafted plants; **P_3**: Air layer plants

T_1: Methyl jasmonate (MeJA) 100 ppm; **T_2**: MeJA 150 ppm; **T_3**: MeJA 200 ppm; **T_4**: Nitrobenzene (NB) 1.0 ml litre^{-1}; **T_5**: NB 1.5 ml litre^{-1}, **T_6**: NB 2.0 ml litre^{-1}; **T_7**: paclobutrazol (PBZ) 0.375 g of active ingredient per metre of canopy diameter applied 30 days after the 'bahar' treatment; **T_8**: same as T_7 except applied 45 days after the treatment; **T_9**: same as T_7 except applied 60 days after the treatment; **T_{10}**: Control; sprayed with water after the 'bahar' treatment

significantly in their rate of transpiration (E; mmol m^{-1} s^{-1}), with tissue-culture plants (P_1) showing a significantly higher rate (5.78 mmol m^{-2} s^{-1}) than that shown by either of the other two propagules. Among the chemicals, paclobutrazol 45 days or 60 days after the bahar treatment and nitrobenzene 2.0 ml $litre^{-1}$ showed comparable rates of transpiration. As to the combinations of propagules and chemicals, tissue-culture plants treated with paclobutrazol irrespective of the time of its application showed a higher rate of transpiration than that seen in any of the other combinations.

Regardless of when paclobutrazol was applied, grafted plants among the propagules and *ambe bahar* showed higher rates of transpiration during fruit set, whereas among the combinations, grafted plants sprayed with nitrobenzene at 1.5 ml $litre^{-1}$ or 2.0 ml $litre^{-1}$ recorded higher rates than those in any of the other combinations (Table 2).

The rate of transpiration varied greatly according on the propagule at the blooming stage in hastha bahar, with tissue-culture plants and air layers (P_3) displaying significantly higher rates than grafted plants. 45 days following the bahar treatment, findings with the substances nitrobenzene 2.0 ml $litre^{-1}$ and paclobutrazol were comparable. In comparison to other combinations, paclobutrazol given to tissue-culture plants 45 or 60 days after the bahar treatment caused transpiration rates to be higher (Table 2).

During fruit set in *hastha bahar*, tissue-culture plants among the propagules showed significantly higher rates of transpiration than grafts or air layers. Among the chemicals, nitrobenzene at 1.5 ml $litre^{-1}$ or 2.0 ml $litre^{-1}$ showed comparable results. As to the combinations, none of them differed significantly from any of the rest in terms of the rate of transpiration (Table 2).

Stomatal conductance

Stomatal conductance (g_s) varied significantly across the propagules at the blooming stage in *ambe bahar* (Table 3), with tissue-culture plants exhibiting higher g_s than air layers or grafts. High g_s, which was comparable to either of the two doses of nitrobenzene (1.5 ml $litre^{-1}$ or 2.0 ml $litre^{-1}$), was observed in plants treated with paclobutrazol 45 or 60 days after the bahar treatment. Paclobutrazol-treated tissue-culture plants showed high gs among the combinations 60 days after the bahar treatment.

The propagules varied greatly throughout fruit set in *ambe bahar*, with grafts displaying greater values than either tissue-culture plants or air layers. Among the compounds, methyl jasmonate 150 ppm and paclobutrazol, regardless of the date of administration, demonstrated noticeably higher stomatal conductance than the other treatments. In comparison to the other combinations, the one containing nitrobenzene, regardless of dose, or paclobutrazol 60 days after the bahar treatment and grafted plants displayed increased stomatal conductance (Table 3).

Grafts displayed greater values than either tissue-culture plants or air layers during fruit set in *ambe bahar*, where propagules varied greatly in terms of g_s. Paclobutrazol, regardless of the date it was applied, and methyl jasmonate 150 ppm among the compounds had noticeably higher stomatal conductivity than the other treatments. The combinations with nitrobenzene, regardless of dose, or paclobutrazol 60 days after the bahar treatment and grafted plants had greater stomatal conductance than the others (Table 3).

The propagules and the combinations of propagules and chemicals did not exhibit any discernible variations in conductance during fruit setting in *hastha bahar*. But when compared to the other chemical treatments, nitrobenzene 2.0 ml $litre^{-1}$ displayed noticeably higher values (Table 3).

Plants treated with PBZ have also shown increased transpiration rates in previous studies. In peonies, PBZ application increased photosynthetic rate and transpiration rate significantly (Xing *et al.,* 2018). Photosynthesis has been studied as a significant limitation due to stomatal opening. In peony, both transpiration rate and stomatal conductance were found to be higher in PBZ treated plants than in control plants, similar to our findings. Another research by Berova and Zlatev (2003) which showed a high transpiration rate and stomatal conductance after PBZ treatment was also in agreement with our findings. This may be explained by the fact that a large stomatal opening and conductance are conducive to CO_2 entry into the intracellular space, allowing PBZ to improve photosynthesis.

Total chlorophyll content

The data in Table 4 show that all three propagules significantly influenced the total chlorophyll content of leaves at the flowering stage of *ambe bahar*, with tissue-culture plants showing the highest levels (2.14 mg g^{-1}), which were significantly higher than those in the other two kinds of propagules. Paclobutrazol, applied 60 days after the bahar treatment, and nitrobenzene, applied to 1.5 ml $litre^{-1}$ or 2.0 ml $litre^{-1}$ plants, respectively, both reported chemical treatments with significantly greater total chlorophyll contents than any of the others. When compared to the other combinations, tissue-culture plants treated with paclobutrazol 45 or 60 days following the bahar treatment displayed the highest levels of total chlorophyll.

The total chlorophyll content of leaves varied significantly between all propagules during fruit set stage of *ambe bahar*; tissue-culture plants had the highest levels (1.72 mg g^{-1}), and the highest chemical concentrations were found for paclobutrazol 45 or 60 days after the bahar treatment and nitrobenzene at 2.0 ml $litre^{-1}$. However, none of the combinations really stood out from the others (Table 4).

At the flowering stage in *hastha bahar*, each of the three propagule differed significantly from the other two in terms of chlorophyll content, with the grafted plants showing the highest levels. Among the chemicals, nitrobenzene at 2.0 ml $litre^{-1}$ led to significantly higher levels than those seen in any of the other chemical treatments. The combinations also differed significantly among themselves, with the combination of nitrobenzene at any of the three doses and grafted plants recording the highest levels of chlorophyll (Table 4).

During fruit set in *hastha bahar*, both tissue-culture plants and grafted plants showed significantly higher levels of chlorophyll; among the chemicals, significantly higher levels of chlorophyll were seen in plants treated with nitrobenzene at 2.0 ml $litre^{-1}$ than those in any of the other treatments; among the combinations, that of grafted plants and nitrobenzene, whether at 1.0 ml $litre^{-1}$ or 1.5 ml $litre^{-1}$ or 2.0 ml $litre^{-1}$, recorded the highest levels of total chlorophyll, significantly higher than those in any of the other combinations (Table 4).

Chlorophyll content was high in tissue culture and grafted plants treated with paclobutrazol or nitrobenzene at the flowering stage in both *ambe bahar* and *hastha bahar*.

Chlorophyll plays a dual role in photosynthesis as an essential component of the primary photosynthetic reaction. It captures light and also acts as a medium for charge separation and electron transport caused by light (Zhao *et al*., 2011). Paclobutrazol had an important impact on the biosynthesis of chloroplast pigments. The increased chlorophyll content in paclobutrazol-treated plants may be due to reduced reactive oxygen damage and improvements in carotenoids, ascorbate, and ascorbate peroxidase levels. Plants treated with paclobutrazol synthesized more cytokinin, which enhanced chloroplast differentiation and chlorophyll biosynthesis, and prevented chlorophyll degradation, according to Niveditha Devi and Somasundaram (2012). When compared to untreated controls, data suggested that Pomegranate plants treated with plant growth regulators, especially PBZ, produced more fruit and had improved photosynthetic characteristics.

Conflict of interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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Variability in morphological parameters of Jamun (*Syzygium cumini* Skeels) genotypes

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ABSTRACT

The study was conducted at the Experimental Farm, Division of Fruit crops, ICAR-IIHR, Bengaluru to assess the variability in morphological traits of Jamun genotypes. The experiment was laid out in a randomized block design with three replications. Result showed that all the genotypes expressed considerable variability with respect to the morphological characters. The genotype Dharwad market sample-2 was showing the highest plant height (618.3 cm). The genotype Andaman collection-4 recorded highest (21.56 cm) leaf length and lowest (9.83 cm) value recorded in the genotype Kaveri pattnam-2. The inter nodal length of the genotype Dharwad -2 recorded the highest value of 7.2 cm and the Dharwad-13 recorded the lowest value of 2.66 cm. The petiole length of the genotype Patna recorded the highest value of 3.00 cm and the Andaman -4 recorded the lowest value of 0.43 cm. Among the genotypes, Dharwad-6 was showing the distinctive from other genotypes in cluster analysis.

***Keywords*:** *Syzygium cumini* Skeels, Genotypes, Morphological and Variability

INTRODUCTION

Jamun botanically called as *Syzygium cumini* Skeels, belongs to the family Myrtaceae (Chase *et al.,* 2009). The jamun also known as Indian blackberry, Java plum, Jambu, black plum and Jambul, Kalajam, Phalinda and Rajamun, damson plum, duhat plum*, etc.* (Sharma *et al*., 2012). *S. cumini* is native to India, Burma, Ceylon and to the Andaman Islands and it is available throughout Indian plains as well as in Kumaon hills up to 1,600 m. It is found grown as a wild and semi-wild in tropical and subtropical parts of India *viz*., Punjab, Haryana, Uttar Pradesh, Maharashtra, Rajasthan, Gujarat, Madhya Pradesh and Bihar. It is a multipurpose tree of both food and medicinal values (Inamdar *et al.,* 2000). All parts of the tree such as fruits, leaves, seeds, and bark are used in Indian medicine system like Ayurveda, Homeopathy, Sidda and Unani (AYUSH) *etc*. Different parts of the jamun were also reported for its antioxidant, anti-inflammatory, anti-microbial, and anti-ulcerogenic (Ghosh *et al*., 2017 and Ayyanar *et al.,* 2012). Before the discovery of insulin, in the treatment of diabetes *S. cumini* was used either alone or in combination with other hypoglycaemic plants even in Europe (Helmstadter, 2008). Seeds contain an alkaloid 'jambosin' and glycoside 'jambolin' which can reduce diastatic conversion of starch into sugars (Yamini *et al*., 2022).

There are no major varieties in jamun; there exist a large number of local seedling strains of this crop which provide great scope for the selection of better types. A lot of variations is available with respect to plant and fruit parameters. These variations can be useful to evolve quality genotype. Hence, the present study was aimed to characterize the jamun genotypes to know the existing variability.

MATERIALS AND METHODS

The study was conducted at Research field, Division of Fruit Crops, ICAR-IIHR, Bengaluru on five years old Jamun genotypes. Different morphological attributes like plant height, canopy spread, leaf characters, new flush colour, intermodal length, petiole length, leaf anthocyanin and phenol

were recorded as per jamun DUS guidelines. The observations were recorded among three trees of each genotype and each tree was considered as a replication. It was analyzed as randomized complete block design (RCBD).

RESULTS AND DISCUSSION

Results presented in Table-1 showed significant variability in morphological parameters of all the genotypes studied. As per jamun DUS guidelines plant showing three types of spreading nature, *i.e.,* spreading, semi-spreading and upright. Most of the genotypes were grouped under upright growth habit. The plant height of the accessions was highly variable. Dharwad market sample-2 recorded the highest plant height (618.3 cm) and KHA-32 genotype was showed the lowest (155 cm) plant height (Table 1). The stem girth of the genotype Kaveripattnam- 4(a) showing highest value (80.43 cm) and lowest was recorded be in genotype KHA -32 (19.66 cm). In present study variations in plant height and stem girth was influenced by the age of the plant. The existence variation in morphological characters of jamun was reported by Inamdar *et al*. (2000) also reported similar results. The canopy spread in North-South direction was highest in Dharwad market sample-2 (513.33 cm) and lowest (161.66 cm) in the genotype KHA-32. The genotype Dharwad market sample-2 showed the highest (498.33 cm) canopy spread in E-W direction and lowest in KHA-32 (19.66 cm). Anushma and Anuradha (2018) reported a similar report on jamun. The leaf length of the genotypes showed more variability. The genotype Andaman-4 recorded highest (21.56 cm) leaf length value and lowest (9.83 cm) value recorded in the genotype Kaveri pattnam-2. Anushma and Anuradha (2018), reported the mean leaf lamina length ranged from 11.63 cm (IIHRJ-14) to 15.53 cm (IIHRJ-10). The genotype Chinnapalli recorded the highest mean value of leaf width 8.23 cm and it was on par with Andaman-4 (7.9cm) and the genotype Madhya Pradesh-2 and Madhya Pradesh-5 recorded the lowest value of 3.80 cm (Table 1). The variation between the genotypes for different morphological characters may be attributed to the differences in the genetic makeup of these genotypes.

The internodal length of the genotypes Dharwad -2 recorded the highest value of 7.2 cm and the Dharwad-13 recorded the lowest value of 2.66 cm. The petiole length of the genotype Patna recorded the highest value of 3.00 cm and the Andaman-4 recorded the lowest value of 0.43 cm. The new shoot length of the genotype Savadatti recorded the highest value of 28.33 cm and the Khanapur-32 recorded the lowest value of 10.33 cm. The genotype Srisailam-18 recorded the highest number of leaves/new shoot 17 and the Dharwad market sample-3 recorded the lowest value of 6. Similar findings were reported by Swamy *et al*. (2017) and Kumar *et al*. (2022) in Jamun.

The lowest tender leaf anthocyanin content (11.26 mg/100g) was recorded in Dharwad -7, whereas the highest leaf anthocyanin content (69.26 mg/100g) in Madhya Pradesh-3. The lowest leaf phenols content (138.4 mg/100g) was recorded in Kaveri pattanam-4, whereas the highest leaf phenols content (3538 mg/100g) in Kaveri pattanam-2.

Grouping of genotypes based on plant characters were done which resulted in 5 non-overlapping clusters. Cluster wise listing of germplasm according to plant characters are given in Table 3 and Fig.1. Cluster-I had maximum number of genotypes (22) and Cluster II had the minimum number of genotypes (1) and this genotype seems to be morphologically distinctive from other clusters with reference to morphological parameters. Cluster wise summary mean of plant characters (Table 4) will indicate the mean range of different traits and genotypes was grouped based on which similar parameters.

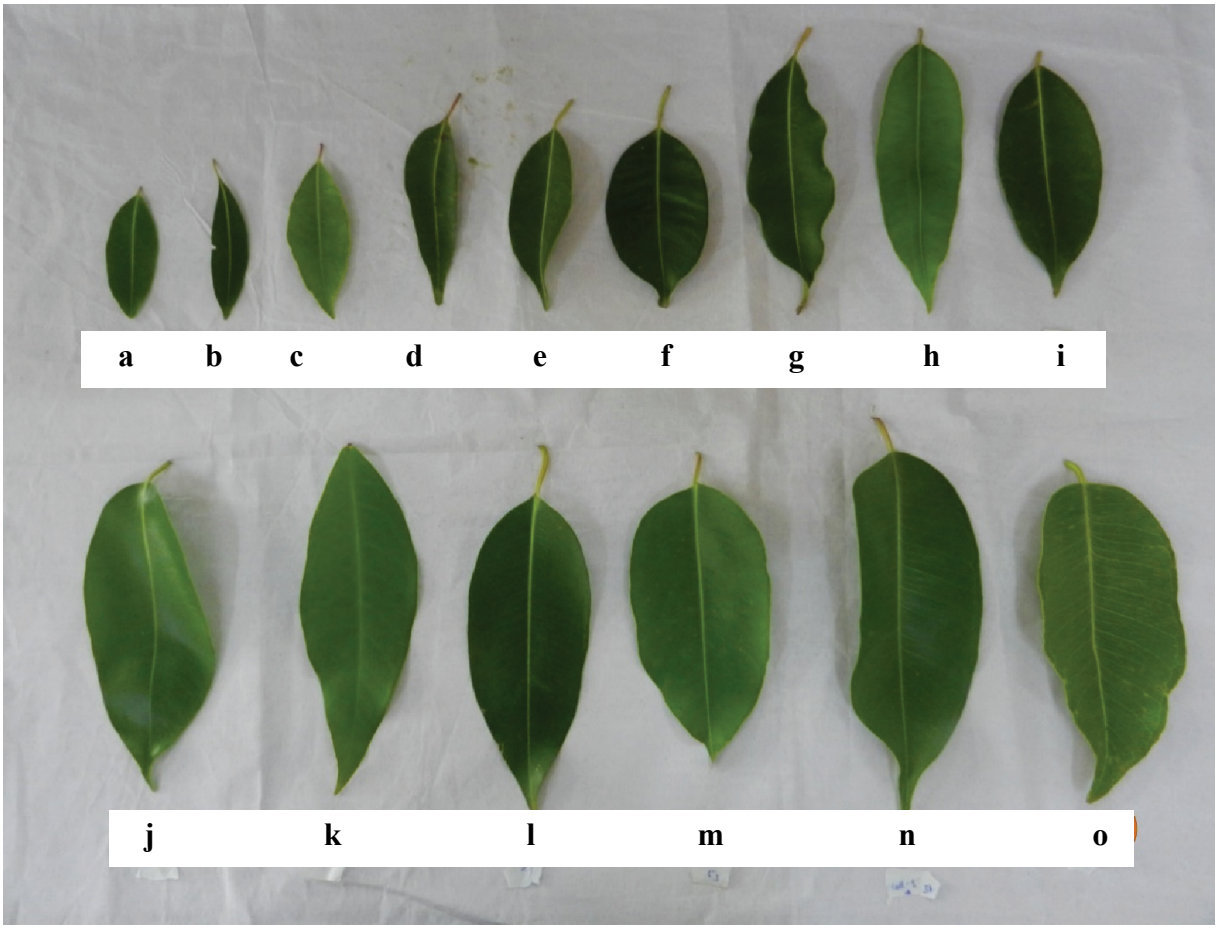
The cluster mean of plant height ranged from 284.4 cm to 591.67 cm Cluster IV (591.67 cm) recorded the highest plant height and the Cluster V (284.4 cm) recorded the lowest plant height (Table 4). The cluster mean value of stem girth was ranged from 32.71cm to 74.33cm. The Cluster IV has the maximum girth of 74.33cm and the minimum girth of 32.71cm was recorded in Cluster V (Table 4). The cluster mean value of N-S ranged from 219.17 cm to 481.67 cm. The Cluster IV has the maximum north-south canopy of 481.67cm and the minimum of 219.17cm was recorded in Cluster V (Table 4). The cluster mean value of E-W ranged from 227.07 cm to 480.42 cm. The Cluster IV has the maximum east-west direction of 480.42cm and the minimum of 227.07 cm was recorded in Cluster V (Table

Upright growth

Semi-spreading growth

Spreading growth

Plate 1. Nature jamun plants



a. Kaveri pattnam-4(a); **b**. Mp-5; **c**. Kaveripattnam-1; **d**. Hurulichikkinahalli; **e**. Kaithnal; **f**. IC-715; **g**. Collection-4a; **h**. Dharwad market sample-4; **i**. JNR-2; **j**. Dharwad market sample-2; **k**. Andaman collection-4; **l**. Collection-9; **m**. JNR-1; **n**. Collection-8; **o**. CHK.

Plate 2. Variability of jamun leaves for shape and size

Table 1: Variation of plant characters of jamun genotypes

Genotype	IC Number	Plant height (cm)	North - South (cm)	East –West (cm)	Stem girth (cm)	Spreading nature
Dhoopdal	IC-0621955	423.33	423.33	420.00	56.33	semi spreading
Selection-45	IC-0621954	426.66	393.33	388.33	49.56	semi spreading
Selection-58	IC-0621956	435.00	420.00	416.66	50.16	Spreading
Savadatti	IC-0621957	393.33	376.66	393.33	55.66	semi spreading
Kaithnal	IC-0621952	455.00	425.00	452.50	62.83	semi spreading
AJG-85	IC-0621953	465.00	441.66	428.33	58.53	semi spreading
IC-715	IC-0587715	421.66	445.00	465.00	59.06	Upright
Konkan Bahadoli	IC-0621958	467.50	395.00	407.50	59.93	semi spreading
Dharwad -2	IC-0621961	425.00	395.00	410.00	61.70	semi spreading
Dharwad -2a	NA	347.50	287.50	287.50	47.13	semi spreading
Dharwad -3a	NA	428.33	425.00	420.00	63.56	semi spreading
Dharwad -4a	NA	365.00	348.33	306.66	43.63	Upright
Dharwad -7	NA	471.66	455.00	443.33	70.96	semi spreading
Dharwad -12	IC-0631356	475.00	450.00	465.00	65.10	semi spreading
Chinnapalli	IC-0621967	470.00	455.00	438.33	67.93	semi spreading
Goma priyanka	IC-0621959	476.66	418.33	401.66	52.50	semi spreading
Paiyur -4	IC-0621969	457.50	405.00	415.00	61.33	Upright
Kaveri pattanam -4	IC-0621971	471.66	368.33	388.33	61.93	Upright
Dharwad -5	NA	418.33	408.33	428.33	50.16	semi spreading
Andaman -4	IC-0621973	503.33	371.66	366.66	40.56	Upright
Dharwad -3	NA	530.00	335.00	350.00	40.43	Upright
Dharwad -4	IC-0631354	235.00	176.66	178.33	70.56	Upright
Dharwad -6	IC-0621963	450.00	443.33	445.00	56.86	Upright
Dharwad -8	NA	447.50	402.50	420.00	56.86	Upright
Dharwad -9	NA	360.00	331.66	315.00	50.40	Upright
Dharwad -10	IC-0631355	416.66	353.33	353.33	50.36	semi spreading
Dharwad -11	IC-0621965	422.50	420.00	415.00	50.60	Upright
Dharwad -13	IC-0621966	480.00	423.33	371.66	60.23	Upright
Kaveri patnam-1	IC-0621970	515.00	391.66	420.00	60.03	Upright
Kaveri pattanam-2	IC-0631357	361.66	220.00	246.66	36.96	Upright
Hirehally	IC-0621968	448.33	335.00	335.00	51.43	Upright
Huruli chikkanahally	IC-0621972	500.00	385.00	391.66	62.16	Upright
Dharwad market sample -1	IC-0621960	480.00	416.66	416.66	57.10	Upright
Dharwad market sample -2		618.33	513.33	498.33	74.33	Upright
Dharwad market sample -3	IC-0621962	546.66	463.33	488.33	75.33	Upright
Dharwad market sample -4	NA	488.33	460.00	445.00	63.63	Upright
Patna	IC-0621975	528.33	478.33	466.66	70.66	semi spreading
Lucknow	IC-0621976	458.33	421.66	396.66	57.23	Upright
Jayanagar-1	IC-0621977	493.33	443.33	431.66	53.83	Upright
Jayanagar-2	IC-0621978	673.33	471.66	468.33	76.96	semi spreading
Chikkodi	IC-0621979	476.66	315.00	338.33	53.96	Upright
Madhya Pradesh-1	IC-0621980	476.66	401.66	386.66	53.66	Upright
Madhya Pradesh-2	IC-0621981	481.66	446.66	430.00	57.23	Upright
Madhya Pradesh-3	IC-0621982	453.33	365.00	358.33	53.76	Upright
Madhya Pradesh-5	IC-0621983	496.66	490.00	476.66	73.63	Upright
Kaveri pattanam -4 (a)	IC-0621971	522.50	507.50	470.00	80.43	Upright
Khanapur-1	IC-0631358	401.66	306.66	371.66	36.00	Upright
Khanapur -24	IC-0631365	241.66	208.33	190.00	30.50	semi spreading
Khanapur -32	IC-0631366	155.00	161.66	145.00	19.66	Spreading
Srisailam-18	IC-0631370	248.33	173.33	170.33	25.83	Upright
Range		155 to 618.3	161.6 to 513.3	19.6 to 498.3	19.6 to 80.43	
SEm±		**28.59**	**34.79**	**36.78**	**5.43**	-
C.D@5%		**80.38**	**97.79**	**103.38**	**15.26**	-

Table 2: Variation in leaf characters of Jamun genotypes

Genotype	IC Number	Leaf length (cm)	Leaf breadth (cm)	Internodal length (cm)	Petiole length (cm)	New shoot length (cm)	No. of leaves/ new shoot	Tender leaf anthocyanin (mg/100g)	Leaf phenols (mg/100g GAE)
Dhoopdal	IC-0621955	14.56	6.86	5.76	2.70	22.33	13.00	23.40	972.17
Selection-45	IC-0621954	13.26	6.33	4.26	2.20	15.00	9.33	12.03	791.55
Selection-58	IC-0621956	13.76	6.60	5.03	2.30	22.33	11.00	12.46	844.55
Savadatti	IC-0621957	14.73	6.23	4.60	2.13	28.33	9.33	15.00	589.90
Kaithnal	IC-0621952	14.90	5.93	4.06	2.20	22.00	6.66	21.27	983.44
AJG-85	IC-0621953	12.83	5.40	4.46	2.33	22.00	11.33	12.10	664.11
IC-715	IC-0587715	12.16	5.76	5.13	1.66	20.66	8.33	31.53	812.39
Konkan Bahadoli	IC-0621958	11.73	6.03	3.73	2.00	23.66	8.00	23.35	1,055.07
Dharwad -2	IC-0621961	16.10	7.06	7.20	2.50	28.00	10.33	18.61	767.61
Dharwad -2a	NA	13.33	5.40	4.00	2.60	26.00	10.33	27.73	990.22
Dharwad -3a	NA	12.90	6.56	3.53	1.76	24.33	10.33	24.80	949.52
Dharwad -4a	NA	14.26	6.36	4.40	2.13	26.00	7.66	26.61	764.37
Dharwad -7	NA	14.56	6.73	4.83	2.00	21.66	9.66	11.26	1,590.48
Dharwad -12	IC-0631356	15.06	6.50	3.40	2.63	25.66	12.00	29.88	1,646.87
Chinnapalli	IC-0621967	13.10	8.23	4.93	2.00	25.00	9.00	21.16	463.92
Gomapriyanka	IC-0621959	12.23	6.80	4.60	2.30	22.33	9.00	26.41	389.80
Paiyur -4	IC-0621969	14.83	6.83	3.83	2.06	25.33	8.67	35.52	401.20
Kaveri pattanam -4	IC-0621971	14.23	7.53	5.00	2.30	21.66	8.33	33.69	138.40
Dharwad -5	NA	14.13	5.50	2.93	1.26	27.00	10.66	17.27	588.88
Andaman -4	IC-0621973	21.56	7.90	5.33	0.43	28.00	10.33	15.88	477.88
Dharwad -3	NA	17.40	7.53	3.50	1.90	26.00	10.00	13.87	632.80
Dharwad -4	IC-0631354	15.13	5.16	3.80	2.00	25.66	10.00	16.80	817.89
Dharwad -6	IC-0621963	14.90	5.93	5.63	2.56	23.66	9.66	16.77	722.40
Dharwad -8	NA	14.33	6.83	4.66	2.08	26.00	8.00	13.82	631.54
Dharwad -9	NA	15.93	7.43	5.00	2.73	23.33	8.33	40.76	717.47
Dharwad -10	IC-0631355	14.60	7.10	4.26	2.60	22.33	7.33	38.35	1,154.52
Dharwad -11	IC-0621965	12.66	5.83	5.06	2.33	19.33	8.33	31.47	2,285.62
Dharwad -13	IC-0621966	10.83	6.06	2.66	2.26	18.33	7.66	20.53	2,447.44
Kaveri patnam-1	IC-0621970	10.00	3.96	2.96	1.43	21.33	9.33	18.37	1,165.27
Kaveri pattanam-2	IC-0631357	9.83	4.75	3.43	1.80	20.00	9.33	34.65	3,538.37
Hirehally	IC-0621968	13.33	6.90	3.96	2.46	25.00	14.00	30.52	840.08
Huruli chikkanahally	IC-0621972	11.26	4.83	3.63	2.66	20.00	11.33	30.10	2,611.91
Dharwad market sample -1	IC-0621960	15.66	6.16	5.40	1.86	19.66	8.33	44.03	2,806.36
Dharwad market sample -2		14.50	6.06	6.73	1.53	15.33	10.00	26.54	2,306.23
Dharwad market sample -3	IC-0621962	17.73	7.40	4.33	1.76	16.66	6.00	38.50	2,229.93
Dharwad market sample -4	NA	14.83	6.10	3.66	1.40	18.46	11.00	47.02	2,319.20
Patna	IC-0621975	14.50	6.80	5.13	3.00	17.26	8.00	33.71	2,209.48
Lucknow	IC-0621976	15.83	6.36	4.33	1.60	22.33	8.00	35.36	2,521.02
Jayanagar-1	IC-0621977	11.66	5.70	3.00	1.56	18.30	9.66	34.70	2,415.22
Jayanagar-2	IC-0621978	15.83	6.83	5.13	1.86	19.16	9.00	40.81	2,187.43
Chikkodi	IC-0621979	15.03	5.70	5.46	1.43	20.10	10.33	33.27	2,312.52
Madhya Pradesh-1	IC-0621980	12.96	5.16	3.16	1.56	17.36	8.00	45.88	2,250.16
Madhya Pradesh-2	IC-0621981	10.80	3.80	3.06	1.06	17.66	10.66	16.86	1,341.85
Madhya Pradesh-3	IC-0621982	14.16	7.43	5.00	1.93	22.20	11.66	69.26	1,893.60
Madhya Pradesh-5	IC-0621983	12.83	3.80	4.60	1.46	20.00	8.66	50.79	1,777.13
Kaveri pattanam -4 (a)	IC-0621971	10.76	4.13	3.13	1.10	21.40	10.33	17.32	1,837.67
Khanapur-1	IC-0631358	14.00	5.83	4.23	1.14	17.33	15.33	45.24	2,287.80
Khanapur -24	IC-0631365	13.80	6.33	4.86	2.26	14.00	12.00	26.49	2,100.01
Khanapur -32	IC-0631366	14.60	6.80	5.90	1.66	10.33	10.66	38.20	2,299.87
Srisailam-18	IC-0631370	13.33	5.33	5.70	1.60	12.50	17.66	33.57	2,403.66
Range		9.8-21.5	3.8-8.23	2.6-7.2	0.43-3	10.3-28.3	6.0-17.6	11.2-69.2	138.4-3538
SEm±		**1.09**	**0.62**	**0.81**	**0.33**	**2.32**	**1.33**	**3.03**	**159.39**
C.D@5%		**3.06**	**1.74**	**NS**	**0.95**	**6.53**	**3.74**	**8.65**	**454.36**

Table 3: Cluster wise grouping of genotypes

Clusters	Genotypes
Cluster-I	Dhoopdal, Selection-58, Savadatti, Kaithnal, Dharwad -2, Dharwad -3aDharwad -4a, Dharwad -7, Chinnapalli, Paiyur -4, Kaveripattanam -4,Dharwad -5, Andaman collection -4, Dharwad -3, Dharwad -8, Dharwad -9, Dharwad -10, Dharwad market sample -1, Dharwad market sample -4Lucknow, Chikkodi and Madhya Pradesh-3.
Cluster-II	Dharwad-6
Cluster-III	Selection-45, AJG-85, Konkan Bahadoli, Goma priyanka, Dharwad -11, Dharwad -13, Huruli chikkanahally, Jayanagar-1, Madhya Pradesh -1, Madhya Pradesh -2, Madhya Pradesh -5 and Kaveri pattanam -4 (a).
Cluster-IV	Dharwad market sample -2, Dharwad market sample -3, Patna and Jayanagar-2.
Cluster-V	Dharwad -2a, Dharwad -4, Kaveri pattanam-2, Khanapur-1, Khanapur-24,Khanapur-32 and Srisailam-18.

Table 4: Cluster wise summary mean of plant characters

Characters	Cluster -1	Cluster -2	Cluster -3	Cluster -4	Cluster -5
Plant height (cm)	446.74	450.00	475.76	591.67	284.40
North-South (cm)	394.43	443.33	430.49	481.67	219.17
East-West (cm)	395.11	445.00	416.6	480.42	227.07
Stem girth (cm)	55.94	56.87	59.36	74.33	32.71
Leaf length (cm)	15.07	14.90	11.99	15.64	13.13
Leaf breadth (cm)	6.77	5.93	5.33	6.78	5.66
Internodal length (cm)	4.67	5.63	3.78	5.33	4.56
Petiole length (cm)	1.97	2.57	1.91	2.04	1.87
New shoot length (cm)	23.75	23.67	19.62	17.11	17.98
Number of leaves / new shoot	9.45	9.67	9.36	8.25	12.19

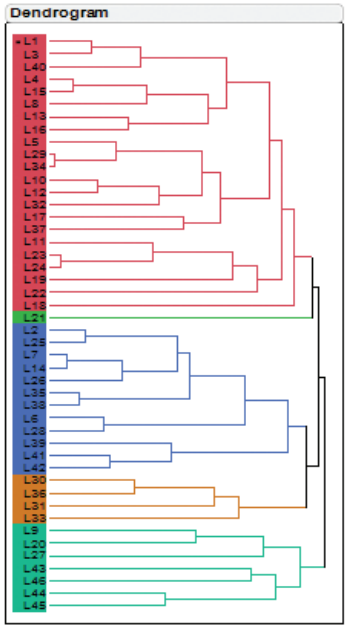


Fig. 1: Showing grouping with reference to morphological characters of jamun genotypes

4).The cluster IV genotypes (Dharwad market sample -2, Dharwad market sample -3, Patna and Jayanagar-2) were highly vigorous and cluster V had least vigorous types.

The cluster mean of leaf length ranged from 11.99 cm to 15.64 cm. The Cluster IV recorded the highest mean value of 15.64 cm and the Cluster III recorded the lowest mean value of 11.99 cm (Table 4). The cluster mean value of leaf breadth ranged from 5.33 cm to 6.78 cm. Cluster IV recorded the highest mean value of 6.78 cm and the Cluster III recorded the lowest value of 5.33 cm (Table 4). The cluster mean value of intermodal length was ranged from 3.78 cm to 5.63 cm. The Cluster III recorded lowest value of 3.78 cm and the Cluster II recorded the highest value of 5.63 cm (Table 4). The cluster mean value of petiole length ranged from 1.87 cm to 2.57 cm. The Cluster V recorded lowest value of 1.87 cm and the Cluster II recorded the highest value of 2.57 cm (Table 4). The cluster mean value of new shoot length was ranged from

Table 5: Correlation analysis of different morphological and leaf biochemical characters of Jamun genotypes

Characters	Plant height (cm)	North - South (cm)	East – West (cm)	Stem girth (cm)	Leaf length (cm)	Leaf breadth (cm)	Internodal length (cm)	Petiole length (cm)	New shoot length (cm)	Number of leaves/ new shoot	Anthocyanin in tender leavesmg/ 100g	Phenol in leaves mg/100g
Plant height (cm)	1											
North - South (cm)	0.831**	1										
East –West (cm)	0.846**	0.970**	1									
Stem girth (cm)	0.679**	0.757**	0.742**	1								
Leaf length (cm)	0.068	-0.041	-0.009	-0.048	1							
Leaf breadth (cm)	0.047	-0.007	-0.006	-0.090	0.648**	1						
Internodal length (cm)	-0.101	-0.080	-0.075	-0.095	0.454**	0.410**	1					
Petiol length (cm)	-0.131	-0.039	-0.056	0.076	-0.076	0.297*	0.151	1				
New shoot length (cm)	0.126	0.149	0.193	0.217	0.267	0.246	-0.123	0.162	1			
Number of leaves/ new shoot	-0.323**	-0.417**	-0.387**	-0.434**	-0.079	-0.155	0.088	-0.181	-0.177	1		
Anthocyanin in tender leaves mg/100g	0.019	-0.077	-0.071	-0.056	0.039	0.056	0.092	-0.064	-0.347*	0.041	1	
Phenol in leaves mg/100g	0.044	-0.094	-0.118	-0.074	-0.208	-0.346	-0.033	-0.187	-0.663**	0.125	0.505**	1

**. Correlation is significant at the 0.01 level.

*. Correlation is significant at the 0.05 level.

17.11 cm to 23.75 cm. The Cluster IV recorded lowest value of 17.11 cm and the Cluster I recorded the highest value of 23.75 cm (Table 4). The cluster mean value of number of leaves/new shoot ranged from 8.25 to 12.19. The Cluster IV recorded lowest value of 8.25 and the Cluster V recorded the highest value of 12.19 (Table 4).

The plant height was highly positively correlated with North-South (0.831), East-West (0.846) and stem girth (0.679). But negatively correlated with number of leaves per new shoot (-0.323) (Table-5). The plant canopy North-South was significantly highly positively correlated with East-West (0.970), stem girth (0.757) and negatively correlated with number of leaves per new shoot (-0.417). The plant canopy East-West was highly positively correlated with stem girth (0.742) and negatively correlated with number of leaves per new shoot (-0.387) (Table-5). Leaf length was highly positively correlated with leaf breath (0.648) and positively correlated with internodal length (0.454). Leaf breadth was positively correlated with internodal length (0.410) and petiole length (0.297). New shoot length was negatively correlated with anthocyanin content in tender leaf (-0.347) and highly negatively correlated with phenol in leaves (-0.663) (table-5). Anthocyanin in tender leaf was positively correlated with phenol in leaves (0.505).

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Cluster analysis and principal component analysis studies among different accessions of Grape accessions in Leh district of Ladakh UT

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ABSTRACT

Evaluation of grape accessions was carried out from different villages of Leh district on the basis of several morphological characters. Observations were recorded on the basis of growth, foliage and yield of vine using cluster analysis and principle component analysis. The analysis of variance revealed significant differences among accessions for each character under study. Based on Mahalanobis D^2 values the accessions were grouped into five clusters. Cluster-II comprised of maximum of 27 accessions followed by cluster III and cluster V having eight accessions. Mean values of clusters for various growth and yield parameters revealed that cluster-I possessed maximum values for cane diameter (2.03 cm), yield (20.99 kg/vine), length (21.36 cm), breadth (11.12 cm), weight (150.42 g) of bunch, number of bunches/vine (148.75), number of berries/bunch (98.25), length (1.53 cm), breadth (1.33 cm) of berry, acidity (0.23 %) and juice content (89.99 %). Per cent variation (28.17) and eigen root (5.634) were registered maximum values for first component. Cluster I accessions and cluster IV accessions were observed more different from each other and provide better segregants through hybridization and can be taken as one of parent in future breeding programmes.

***Keywords*:** Accessions, cluster, evaluation, grape, PCA**,** selection, survey.

INTRODUCTION

Grape (*Vitis vinifera* L.) is now fully adapted in the subtropical and tropical climatic conditions of India however it is considered as temperate fruit crop. Grape species are highly heterozygous and offsprings produced through seedlings show wide genetic variability with respect to vine vigour and berry quality due to which seeds are not preferred for propagation. More than 9,600 grape cultivars are present across the world (Galet, 2000) and almost 16,000 prime names appear in the *Vitis* International Variety Catalogue (Maul and Eibach, 2003). For efficient utilization of germplasm, facts related to variability among plant species is of prime importance. Selection and multiplication of elite accessions of any fruit crop from existing heritable variability is important to increase its adoption and production for domestic consumption and from commercial point of view. Presence of genetic divergence in any population helps in the selection of desirable parents used in breeding programme which leads in the reduction of crosses made (Vanavermaete *et al.,* 2020). In any hybridization programme parents are selected on the basis of their earlier performance and the objectives of research programme. For purposeful hybridization in any heterosis breeding, degree and nature about the genetic divergence is important for the breeders in choosing the right parents (Farhad *et al.,* 2010, Khodadabi *et al*., 2011). To take advantages form the transgressive segregation, the information of genetic gap among parents is essential (Khodadadi *et al.,* 2011). The standardization of variables is also necessary towards defining the genetic distance so that all variables are of similar significance in defining the distance. Among various methods, Tocher's method is mostly used for the estimation of genetic diversity through cluster analysis. Euclidean distance can theoretically evaluate the genetic distance among parents to maximize the transgressive segregation (Hoque and Rahman, 2006). Identification of elite grapes accessions through survey, their adoption and multiplication, hence upsurge grapes productivity. Till date no

recognized effort has been conducted and documented on this aspect in Ladakh region. Due to economic importance of grape and for the enhancement of its cultivation in Ladakh region, the present study was conducted in five villages of Leh district to produce vivacious figures on grape vine germplasm using cluster analysis and Principle Component Analysis.

MATERIALS AND METHODS

Area surveyed and experimental material details

After a systematic survey from five different villages (Achinathang, Warseedo, Dha, Hanuthang and Yokmathang) of Leh district in the UT of Ladakh region during 2014, fifty grape accessions were selected. The surveyed area lies between 34°80'N latitude and 77°34'E longitude at an altitude of 3414 m MSL however weather during the study period was mostly warmer with highest temperature upto 38°C in summers and lowest -37°C in winters. Considering vigour, health, bearing habit and desirable berry physio-chemical parameters of berries the vines were selected and marked which are of seedling origin having age between 20-60 years.

Observations recorded

Data was recorded on various growths and yield parameters viz. length (m) and diameter (cm) of cane, internodal length (cm), leaf area (cm^2), yield (kg/vine), yield efficiency (kg/cm^2) was calculated as per Westwood (1993). Ten bunches and ten berries were randomly taken for physical parameters and both bunch (length and breadth) and berry (length and breadth) were measured using vernier caliper and expressed in centimeter.Weight of bunch (g) and berry (g) were also taken using digital weighing balance from the randomly selected samples. Number of bunches/vine, berries/bunches (randomly ten bunches/vine) and seeds/berry were counted and expressed in numbers. Bio-chemical analysis of berries on various parameters *viz.* total soluble solids (°B), acidity (%), TSS/acid ratio, total sugars (%) and juice content (%) were recorded as per the standard procedure as given in AOAC (2000).

Data recorded on all the studied parameters were statistically analyzed using standard procedures (Panse and Sukhatme, 1995). Non- hierarchical Euclidean cluster method was used for the analysis of genetic divergence (Spark, 1973) among the accessions and for the classification of accessions into uniform groups.

RESULTS AND DISCUSSION

Significant differences were observed among the accessions for all the studied characters which depict large extent of genetic variability. The computation from covariance matrix gave non-hierarchical clustering based on Mahalanobis D^2 values among fifty accessions and grouped them into five clusters. The clusters occupied by fifty accessions of grape are presented in Table 1and Fig. 1 and elucidated that maximum number of accessions was registered in cluster II having 27 accessions followed by cluster III, cluster V, Cluster I and cluster IV having eight, eight, four and three accessions, respectively. The clustering pattern revealed that accessions from the same locations did not fall in the same cluster depicting that genetic diversity was not necessarily associated with graphical location. The configuration of grouping indicated that the geographical diversity was not the only criteria to group the accessions of a specific source or area which means that accessions originating from a single locality were grouped in different clusters which could be due to factors like, genetic architecture, heterogeneity, history of selection and mutation in the existing population (Martínez *et al.*, 2023). A similar trend of clustering patterns has been reported by Barua and Sharma (2003), Kaushal and Sharma (2005), Thakur *et al.* (2005),Sharma *et al*. (2014 and 2015) and Bhowmick *et al.* (2016) in different fruit crops *viz.* apple, almond, pecan, walnut and Burmese grape, respectively.

The mean value on various characters of cluster in grape accessions is presented in Table 2. Cluster I had the highest mean values for characters cane diameter (2.03cm), internodal length (29.17cm), leaf area (170.75cm^2), yield (20.99kg/vine), bunch length (21.36 cm), bunch breadth (11.12cm), bunch weight (150.42g), number of bunches/vine (148.75), number of berries/bunch (98.25), length of berry (1.53cm), breadth of berry (1.33cm), number of seeds/berry (1.75), acidity (0.23 %) and juice content (89.99 %) while cluster II exhibited the highest yield efficiency (0.09) and total sugar

Table 1: Clustering pattern of fifty grape accessions on the basis of genetic divergence

Cluster	Number of accessions	Accessions number
I	4	LG-1, LG-15, LG-29, LG-42
II	27	LG-2, LG-4, LG-5, LG-6, LG-7, LG-8, LG-9, LG-10, LG-11, LG-12, LG-18, LG-19, LG-20, LG-21, LG-22, LG-24, LG-25, LG-26, LG-27, LG-30, LG-31, LG-32, LG-33, LG-37, LG-38, LG-41, LG-45
III	8	LG-3, LG-16, LG-17, LG-34, LG-36, LG-43, LG-44, LG-46
IV	3	LG-13, LG-40, LG-47
V	8	LG-14, LG-23, LG-28, LG-35, LG-39, LG-48, LG-49, LG-50

Table 2: Cluster means for differentparameters of grape accessions in Leh district

Characters	Clusters				
	I	II	III	IV	V
Cane length (cm)	186.76	136.98	171.22	127.62	201.77
Cane diameter (cm)	2.03	1.54	1.71	2.02	1.72
Internodal length (cm)	29.17	23.13	24.58	23.13	22.54
Leaf area (cm^2)	170.75	141.44	150.37	164.78	154.16
Yield (kg/vine)	20.99	12.10	14.68	13.47	12.20
Yield efficiency (kg/cm^2)	0.08	0.09	0.03	0.07	0.05
Bunch length (cm)	21.36	17.13	20.25	19.26	18.36
Bunch breadth (cm)	11.12	9.21	10.55	9.95	9.59
Bunch weight (g)	150.42	90.19	113.22	100.65	90.28
No. of bunches/vine	148.75	135.29	129.12	102.96	94.82
No. of berries/bunch	98.25	62.77	76.50	66.66	56.37
Berry length (cm)	1.53	1.08	1.10	1.17	1.03
Berry breadth (cm)	1.33	1.02	1.04	1.05	1.03
Berry weight (g)	1.38	1.33	1.32	1.41	1.54
Number of seeds/berry	1.75	1.00	1.00	1.00	1.00
TSS (^oB)	19.10	20.18	20.53	20.66	20.08
Acidity (%)	0.23	0.21	0.20	0.15	0.22
Total sugars (%)	9.16	11.56	10.75	9.73	10.29
TSS/acid ratio	82.75	94.85	101.83	131.83	92.02
Juice content (%)	89.99	74.80	79.99	77.71	79.00

(11.56 %). Maximum mean values for TSS (20.66^{o}B) and TSS/acid ratio (131.83) was recorded in cluster IV whereas cane length (201.77 cm) and berry weight (1.54 g) was registered maximum in cluster V. Mean values of Cluster III and I were bestowed with desirable features which is required for desirable recombination's in segregating generations. Kanavi *et al.* (2020) reported that mean of clusters depicts the inner diversity in the material taken under investigation. Priority should be given to those clusters for further selection and choosing parents in hybridization programme, in which characters contribute more towards the D^2 values (Bose and Pradhan, 2005).

The principal component analysis revealed prominent differences among the accessions studied. Table 3 depicts that in principal component analysis, maximum contribution of diversity (28.17 %) and maximum eigen roots (5.634) values was observed in first component with major contributions from cane diameter, fruit yield, length, breadth and weight of bunch whereas minimum contribution towards diversity (0.04 %) and minimum eigen root value (0.007) was reported

Table 3: Eigen vectors, eigen roots and per cent variation elucidated by different characters of grape accessions

Characters	Eigen roots	Variation (%)	Eigen vectors																			
			1	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**	**10**	**11**	**12**	**13**	**14**	**15**	**16**	**17**	18	19	20
Cane length (cm)	5.634	28.17	0.228	0.201	0.167	0.198	0.349	-0.101	0.303	0.286	0.377	0.005	0.234	0.242	0.267	0.102	0.256	-0.005	0.075	-0.165	-0.063	0.308
Cane diameter (cm)	2.415	12.08	-0.021	0.041	-0.055	-0.015	0.007	0.087	-0.236	-0.206	-0.081	0.320	0.210	0.128	0.090	-0.336	0.046	-0.434	0.356	-0.094	-0.519	0.050
Internodal length (cm)	2.165	10.83	-0.296	-0.009	0.117	-0.015	0.250	0.447	0.111	0.055	0.135	0.304	0.402	-0.061	-0.097	-0.351	0.047	0.062	-0.266	0.228	0.242	-0.155
Leaf area (cm^2)	1.737	8.69	0.036	-0.398	0.094	-0.124	0.090	0.223	0.326	0.358	0.082	-0.153	0.003	-0.379	0.336	0.087	-0.097	-0.084	0.330	-0.031	-0.317	-0.043
Yield (kg/vine)	1.404	7.02	-0.080	-0.349	0.389	-0.181	-0.041	-0.026	-0.065	0.077	-0.089	0.106	-0.117	0.341	0.337	0.041	0.114	0.330	0.287	0.438	-0.076	-0.094
Yield efficiency (kg/cm^2)	1.181	5.91	0.080	0.183	0.419	0.400	0.125	-0.068	-0.076	-0.112	-0.023	0.429	-0.261	-0.219	-0.281	0.318	0.009	-0.161	-0.021	0.276	-0.050	0.014
Bunch length (cm)	1.108	5.54	-0.311	-0.219	0.071	0.132	-0.022	0.352	0.057	0.137	-0.029	0.132	-0.269	0.321	0.237	0.296	-0.248	-0.304	-0.217	-0.381	0.014	-0.059
Bunch breadth (cm)	0.881	4.41	0.012	-0.060	0.458	0.542	-0.278	-0.110	0.070	0.024	-0.160	-0.335	0.172	0.005	0.024	-0.373	-0.083	-0.003	0.003	-0.173	0.007	0.232
Bunch weight (g)	0.797	3.99	-0.441	0.120	-0.177	0.327	-0.092	0.265	-0.092	0.025	-0.114	-0.024	-0.116	-0.066	-0.113	0.064	0.459	0.366	0.317	-0.179	-0.006	0.201
No. of bunches/vine	0.702	3.51	0.304	0.063	-0.326	0.349	0.070	0.234	0.092	0.004	-0.051	0.166	-0.014	0.128	0.055	-0.071	-0.591	0.349	0.255	0.118	0.019	0.037
No. of berries/ bunch	0.528	2.64	-0.399	0.401	0.068	-0.140	-0.180	-0.175	0.244	0.361	-0.227	-0.008	-0.073	-0.016	0.037	-0.162	-0.293	-0.128	0.012	0.248	-0.052	0.388
Berry length (cm)	0.468	2.34	0.023	0.119	-0.186	-0.043	-0.072	-0.329	0.234	0.353	-0.146	0.468	-0.084	0.011	-0.019	-0.074	0.161	0.114	0.039	-0.235	-0.015	-0.553
Berry breadth (cm)	0.321	1.61	0.099	-0.561	-0.312	0.300	-0.114	-0.187	0.141	0.093	-0.147	0.194	-0.037	-0.043	0.041	-0.179	0.225	-0.171	-0.231	0.201	0.085	0.366
Berry weight (g)	0.296	1.48	0.516	0.512	0.237	-0.250	-0.287	0.452	0.074	0.085	-0.323	0.134	-0.133	-0.023	0.002	-0.125	0.204	0.050	-0.110	-0.166	0.083	0.214
Number of seeds/berry	0.211	1.06	-0.117	-0.241	0.273	-0.180	0.122	-0.293	-0.066	-0.167	0.020	0.306	0.040	-0.047	-0.191	-0.142	-0.249	0.312	0.043	-0.479	0.127	0.357
TSS (°B)	0.074	0.37	0.068	-0.016	0.012	0.055	0.274	-0.001	-0.642	0.510	-0.082	-0.033	-0.055	-0.353	0.290	-0.106	-0.067	0.027	-0.071	-0.058	-0.025	-0.034
Acidity (%)	0.045	0.22	-0.082	0.014	0.022	-0.011	0.076	0.017	0.337	-0.357	-0.133	0.061	0.005	-0.567	0.624	0.072	-0.011	0.042	0.039	-0.058	-0.025	-0.034
Total sugars (%)	0.013	0.07	-0.014	0.004	0.005	0.024	-0.642	0.036	-0.129	0.090	0.609	0.208	0.087	-0.186	0.123	0.060	-0.075	0.151	-0.130	0.021	-0.196	0.044
TSS/acid ratio	0.010	0.05	0.001	-0.040	-0.011	0.012	-0.131	-0.037	-0.089	0.085	-0.414	0.066	0.690	0.016	-0.034	0.527	-0.053	0.087	-0.101	-0.004	-0.104	0.040
Juice content (%)	0.007	0.04	0.025	-0.028	0.010	-0.029	-0.202	-0.008	-0.064	0.066	0.097	0.088	0.148	-0.067	0.056	0.100	-0.017	-0.356	0.538	-0.005	0.689	-0.006

Percent variation explained by first 12 components = 95.1068

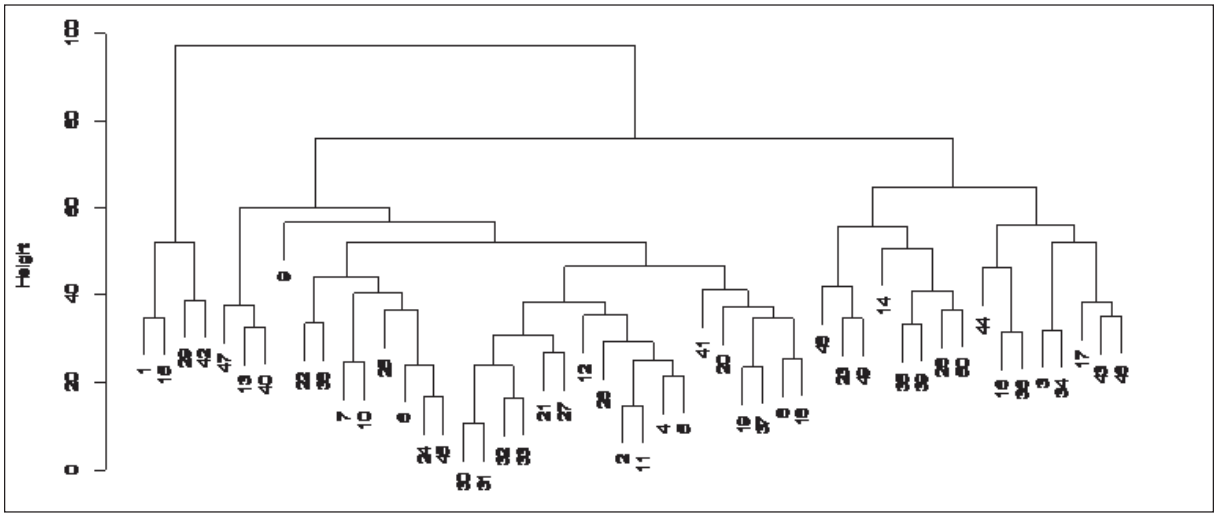

Fig. 1: Dendogram of cluster analysis of various grapes accessions of Leh district

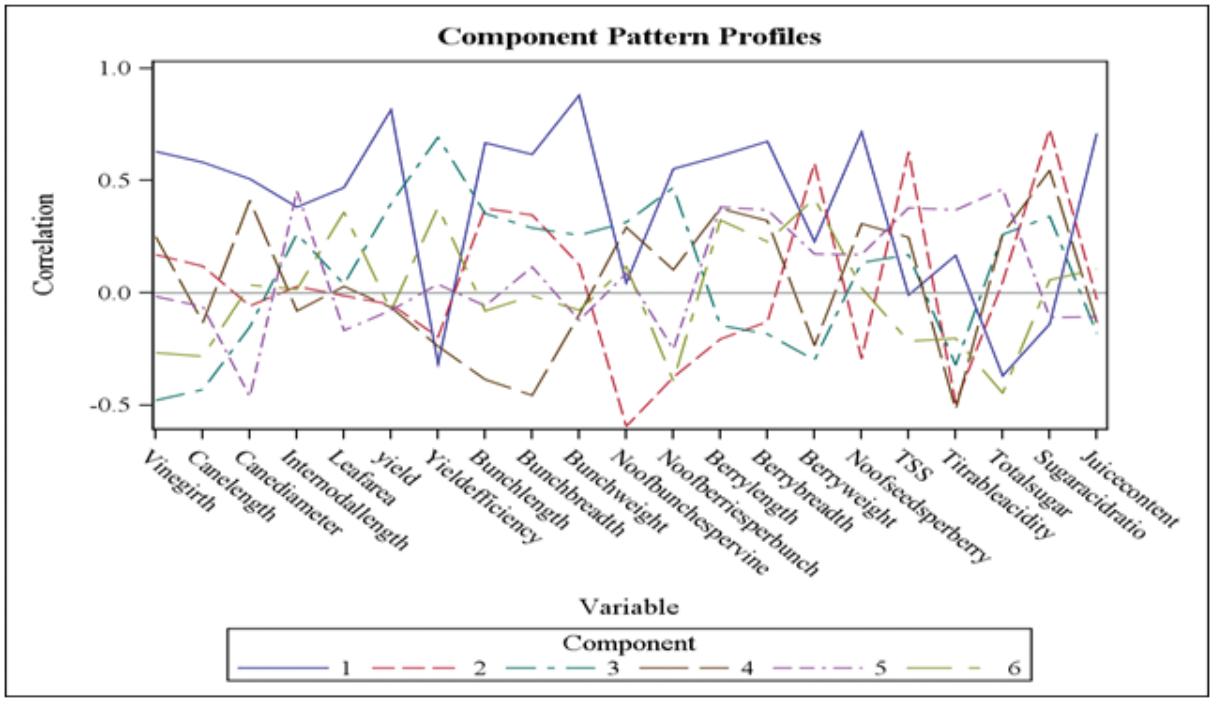

Fig. 2: Scatter plot of Principal component of various grape accessions ofLeh district

in last component with major role from number of berries/bunch, berry weight, acidity and TSS/acid ratio. Cane diameter which is second component exhibited the eigen root value and total variation of 2.415 and 12.08 per cent, respectivelywith mainly contribution of number of bunches/vine, number of berries/bunch and cane diameter. An eigen root value of 2.165 with total variation of 10.83 per cent was registered by the third component i.e. intermodal length and the major contribution for this component is of yield, yield efficiency, number of bunches/vine, number of berries/bunch and total sugars whereas 4th vector i.e. leaf area has the eigen root value and total variation of 1.737 and 8.69 per cent, respectively and the contribution was from length and breadth

of bunch, breadth of berry and acidity. Yield was considered as fifth component with eigen root value of 1.404 and the total variation was 7.02 per cent with maximum and contributed mainly by internodal length, berry length, berry breadth, total soluble solids and total sugars, however, sixth component i.e. yield efficiency showed eigen root value and total variation of 1.181 and 5.91 per cent respectively and mainly contribution was of internodal length, leaf area, number of bunches/ vine, berry weight and total sugars. First twelve components registered 95.11 per cent of total variation.The scatter plot was generated for grape accessions selected from five different villages of Leh district for the first two principal components from a principal component analysis of twenty agro-morphological characters as depicted in Fig. 2. Such separation of genotypes may be due to distinct and diverse nature of the varieties for different agro-morphological traits (Kadu *et al*., 2007 and Viana *et al.,* 2011). Characters with largest absolute value closer to unity within the first principal component influenced the clustering more than those with lower absolute value closer to zero (Kumar *et al.,* 2015).

Rao *et al.* (2003) also reported that for any crop species there is direct correlation between geographical distribution and genetic diversity of that crop and also concluded that eco-geographically different cultivars/accessions also differ from each other genetically. A wide range of variation for almost all the economically important traits is present in this crop. This implies a great potential for breeding through hybridization programme.

From current investigations, with respect to genetic divergence and its component analysis of grape accessions this is inferred that hybridization among genetically different accessions will be helpful for obtaining desirable segregates.

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Effect of dates of transplanting on bulb yield and quality of *kharif* onion cultivars

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ABSTRACT

An experiment was performed at Horticulture Research Farm of Babasaheb Bhimrao Ambedkar University, Lucknow, Uttar Pradesh, to study the influence of different dates of transplanting on bulb quality of two cultivars for off-season kharif onion, in the year 2018-19 and 2019-20. Two Kharif onion cultivars viz., Agrifound Dark Red and L-883 were selected and laid out in factorial randomized block design (two factors) with eight different dates of transplanting and each treatment combination replicated thrice. It was found that onion bulb quality was significantly affected by transplanting on different dates for both cultivars. Among them, transplanting on 20th September produced bulb with maximum total soluble solids, total sugars and ascorbic acid. Among the two varieties, L-883 had maximum titratable acidity. However, maximum bulb yield was obtained when it was transplanted on 30th September resulting the highest yield. The results of the above study indicate that L-883 bulbs can be transplanted on 30th September for profitable off season (kharif) production, and on 20th September for better quality bulbs at Lucknow region.

***Keywords*:** onion, kharif onion, yield, quality

INTRODUCTION

Onion (*Allium cepa* L.) is one of the most important and commercial vegetable bulb crop cultivated extensively in India. It belongs to family Alliaceae, having chromosome number 16 (2n = 2 x = 16). This bulb vegetable is grown mainly during the *rabi* season and included in the daily diet of people around the globe. Allyl propyl disulphide is a volatile compound present in onion, responsible for pungency. This plant has various therapeutic effects (Ouzid *et al*., 2021) and is used for treating loss of appetite, prevention of atherosclerosis, age related changes in blood vessels as well as reduces the risk of gastric cancer. In terms of area and production of onions, India ranks second to China. In India, major onion growing states are Maharashtra, Karnataka, Madhya Pradesh, Bihar, Rajasthan, Gujarat, Andhra Pradesh, Haryana, Uttar Pradesh and West Bengal. In India, onion is cultivated in an area of about 1320130 hectares with a production of 20931250 MT and productivity of 15.86 tonnes per hectare. In Uttar Pradesh state of India, it is grown in about 30.00 ha area with a production of 508.90 MT and productivity of 16.94 tonnes/ha. (NHB Data based 2021-22) The production of *kharif* onion has numerous advantages over the *rabi* (winter) onion *i.e.* increases total production to fulfill the demand for fresh onion in the market. *Kharif* onion also provides a higher price as than *rabi* season onion crop. There are very few *kharif* season varieties known to farmers (Dewangan *et al*., 2012). However, there are many onion varieties available in the local market, but their performances have not yet been evaluated under Lucknow conditions, and is unclear which onion variety is to be chosen for *kharif* season. Due to lack of technical knowledge on this aspect, the onion growers of these regions are suffering up to a great extent, not only due to low bulb production but also problems in keeping quality of bulb in *kharif* season cultivation. The *rabi* season onion is harvested in April-May is stored all over the country and slowly available for domestic supply and export until September-October. There is a critical gap in onions supply from October to March and, as a result, prices shoot up in the country. In this period of dearth, a successful harvest in the *kharif* season can fill the shortage between demand and supply of onions. Keeping this view, the present experiment

was conducted to assess the influence of various dates of transplanting on *kharif* onion production with two onion varieties.

MATERIALS AND METHODS

The experiment was conducted during the year 2018-19 and 2019-20 at the Horticulture Research Farm of the Department of Horticulture, School of Agricultural Sciences and Technology, Babasaheb Bhimrao Ambedkar University, Lucknow, Uttar Pradesh (26°552 North latitude and 80°592 East longitude, 123 m from MSL). The climate is subtropical with a dry, hot summer (maximum 45° C) and cool winter (3° C), and the soil is somewhat alkaline. The meteorological observation collected from Indian Institute of Sugarcane Research (ICAR-IISR) Lucknow near to experimental area, during the experimental period showed that it received maximum rainfall during August-September period of both the years and some erratic rainfall during winter months of January (Table 2). The present experiment consisted of eight different transplanting dates and two cultivars of *kharif* onion were used namely, Agrifound Dark Red (V_1) and L-883 (V_2). The transplanting of onion seedlings was done on 1.95 m x 2 m plots at 15 cm x 10 cm spacing on respective dates. Seeds of selected onion cultivars were collected from the National Horticultural Research and Development Foundation, Deoria Centre, Uttar Pradesh, India. Seeds were sown in the nursery on 5th June for transplantation on 30th August and on subsequent dates of transplanting, as per the experimental design. The soil was mixed with finely crumbled and fully decomposed farmyard manures (FYM) at 3-4 kg/m^2 ten days before seeds were sown. To prevent damping off, the seeds were treated with thiram at a rate of 2g per kg of seeds before sowing. Seeds were sown in lines spacing at 5 cm distance and covered with finely sieved compost. There were eight dates of transplanting (D_1-30th August, D_2-10th September, D_3-20th September, D_4-30th September, D_5-10th October, D_6-20th October, D_7-30th October, D_8-10th November) for both selected onion cultivars, laid out in factorial randomized block design. To raise a healthy crop, all recommended practices were uniformly adapted to all cultivars and treatments. Plots were irrigated frequently at 7-10 days intervals until maturity, depending on weather conditions. At maturity, when two thirds of the leaves turned yellow with neck fall, the onion bulbs were harvested and cured for a short period of five days. The observations were taken following standard procedure (Ranganna, 1991) on bulb yield and quality parameters. Treatment specific input and output costs were computed using on farm market price and other economic indicators such as net return and benefit : cost ratio. The cost of cultivation (Rs. per ha) was computed using the local market price of various inputs used in production. Using the on farm market price, the monetary return of onion bulb yield was calculated in rupees. The gross income (Rs. per ha) was calculated by adding the monetary value of the onion bulb by multiplying bulb yield (q per ha) with price (Rs per quintal). Net return (Rs. per ha) of each treatment was computed by subtracting the cost of cultivation from the specific treatment's gross income. Subsequently, benefit: cost ratio (B: C) was worked out by dividing the net return of a treatment by the expenditure incurred. The observed data were analyzed statistically and treatment mean was compared at 5% level of significance.

RESULTS AND DISCUSSION

Effect of different dates of transplanting on bulb yield (kg per plot and q/ha) on onion cultivars (*kharif*)

Recorded observations were analysed and presented in Table 1. The bulb yield (kg/plot) of variety V_2(L-883) was found as the highest (10.32 kg/plot) when transplanted in D_4 - 30thSeptember transplanting as compared to variety V_1(Agrifound Dark Red) at D_6 transplanting date i.e. 20th October transplanting (9.97 kg/plot), while, the minimum yield was reported at D_1, 30th August transplanting (7.81 kg/plot) in variety V_1. Similar result was found in the case of yield per ha (q/ha). The maximum yield/ha was observed on D_4 *i.e.*30thSeptember transplanting (393 q/ha) in variety V_2, while the minimum was reported on D_1 i.e.30th August transplanting (7.81 kg/plot) in variety V_1. The similar findings were also noted by Sharma *et al*. (2003), Mahanthesh (2009) and Prasad *et al*. (2017) in onion.

Table 1: Effect of date of transplanting on *kharif* onion production.

D-Date of transplanting; V- Variety; * SEm and CD values are based on mean of V_1 and V_2

Variety/Date of transplanting	Yield (kg/plot)		Yield (q/ha)		Reducing sugar (%)		Non Reducing sugar (%)		Total sugar (%)		TSS (%)		Ascorbic acid (mg/100g)		Acidity (%)	
	V_1	V_2	V_1	V_2	V_1	V_2	V_1	V_2	V_1	V_2	V_1	V_2	V_1	V_2	V_1	V_2
D_1-30th August	7.81	8.19	300.22	314.75	4.42	5.19	3.57	4.43	7.99	9.62	8.81	9.68	8.23	8.43	0.54	0.50
D_2-10th September	8.48	8.67	325.9	331.75	4.78	5.27	4.19	4.22	8.97	9.45	10.22	10.52	8.64	9.34	0.51	0.47
D_3-20th September	9.08	9.54	349.09	362.52	4.97	6.35	4.29	4.89	9.26	11.07	12.14	12.07	9.92	10.39	0.45	0.26
D_4-30th September	9.72	10.32	371.46	393.91	5.34	5.41	3.53	4.33	8.87	9.75	11.87	11.52	9.08	9.33	0.35	0.42
D_5-10th October	9.25	9.47	363.19	361.49	4.90	5.65	4.07	4.14	8.97	9.75	12.49	11.09	9.68	9.09	0.34	0.42
D_6-20th October	9.97	8.95	378.43	353.37	5.66	5.39	4.21	3.92	9.87	9.31	12.21	11.24	9.24	8.80	0.39	0.36
D_7-30th October	9.38	8.83	359.51	342.03	5.73	5.44	4.49	4.33	10.23	9.77	12.82	11.31	10.24	9.38	0.32	0.45
D_8-10th November	8.66	9.87	332.91	379.47	4.88	5.15	3.96	3.80	8.84	8.93	10.73	10.93	8.69	9.45	0.48	0.42
SEm (±)	D	0.024		5.42		0.12		0.14		0.18		0.61		0.025		0.005
	V	0.012		2.71		0.06		0.07		0.09		0.08		0.012		0.002
	D×V	0.033		7.66		0.18		0.2		0.26		0.23		0.035		0.007
CD(P=0.05)	D	0.068		15.73		0.37		0.42		0.52		0.47		0.072		0.014
	V	NS		NS		0.18		0.21		0.26		0.23		0.036		0.007
	D×V	0.097		22.24		0.52		0.59		0.74		0.66		0.102		0.020

Table 2: Weekly meteorological observation during the experimental period.

Period		Mean temperature (°C)		Relative humidity (%)		Wind	Rainfall
Weekly data	Date	Max.	Min.	Max.	Min.	velocity (km/hr)	(mm)/ annum
1	27/08/18 to 02/09/18	33.1	25.3	95	78	1.6	22.45
2	03/09/18 to 09/09/18	30.8	24.2	96	85	2.1	20.91
3	10/09/18 to 16/09/18	33.3	24.6	86	61	5.9	0
4	17/09/18 to 23/09/18	33.9	23.4	89	58	3.2	0.65
5	24/09/18 to 30/09/18	34.2	23.1	87	50	3.1	0.28
6	01/10/18 to 07/10/18	33.3	24.6	86	61	5.9	0
7	08/10/18 to 14/10/18	33.9	23.4	89	58	3.2	0
8	15/10/18 to 21/10/18	34.2	23.1	87	50	3.1	0
9	22/10/18 to 28/10/18	33.1	21.1	85	54	2.8	0
10	29/11/18 to 04/11/18	32.2	20.2	86	52	2.5	0
11	05/11/18 to 11/11/18	34.3	22.7	85	53	3.1	0
12	12/11/18 to 18/11/18	29.5	12.7	97	43	2.8	0
13	19/11/18 to 25/11/18	26.5	9.6	83	35	2.5	0
14	26/11/18 to 02/12/18	26.3	7.0	96	34	1.3	0
15	03/12/18 to 09/12/18	27.2	6.5	89	32	1.1	0
16	10/12/18 to 16/12/18	25.1	5.7	85	33	2.2	0
17	17/12/18 to23/12/18	24.2	5.4	78	32	1.2	0
18	24/12/18 to 30/12/18	23.8	4.6	75	31	1.1	0
19	31/12/18 to 07/01/19	22.7	4.9	97	45	1.5	0
20	08/01/19to 14/01/19	22.6	5.8	93	37	1.8	0
21	15/01/19to 21/01/19	22.9	4.5	96	40	2.3	0
22	22/01/19 to 28/01/19	21.8	10.3	90	65	2.0	0.78
23	29/01/19 to 04/02/19	22.3	7.0	94	45	2.7	0
24	05/02/19 to 11/02/19	22.5	9.5	97	58	2.5	2.65
25	12/02/19 to 18/02/19	23.6	10.4	94	53	2.3	0.2
26	19/02/19 to 25/02/19	26.4	11.3	93	42	3.6	0
27	26/02/19 to 04/03/19	23.6	9.5	91	51	2.9	0.82
28	05/03/19 to 11/03/19	27.5	10.9	88	38	4.4	0
29	12/03/10 to 18/03/19	30.5	13.1	78	30	4.1	0
30	19/03/19 to 25/03/19	32.1	15	71	27	5.5	0
31	27/08/19 to 02/09/19	34.7	24.8	92	65	1.4	0.77
32	03/09/19 to 09/09/19	35.1	24.9	90	67	1.5	0.8
33	10/09/19 to 16/09/19	33.8	23.6	94	77	1.7	6.6
34	17/09/19 to 23/09/19	30.2	22.2	96	90	1.3	15.06
35	24/09/19 to 30/09/19	27.8	19.6	97	89	2.0	26.97
36	01/10/19 to 07/10/19	31.0	19.2	95	63	1.3	0.71
37	08/10/19 to 14/10/19	33.1	17.1	95	55	1.3	0
38	15/10/19 to 21/10/19	32.0	16.8	95	58	0.6	0
39	22/10/19 to 28/10/19	30.0	11.6	94	53	1.3	0
40	29/10/19 to 04/11/19	30.0	15.2	94	55	1.2	0
41	05/11/19 to 11/11/19	29.5	14.9	92	53	1.8	0
42	12/11/19 to 18/11/19	29.4	13.0	90	38	1.9	0
43	19/11/19 to 25/11/19	27.7	11.9	94	44	2.1	0

Contd.

Table 2 Contd.

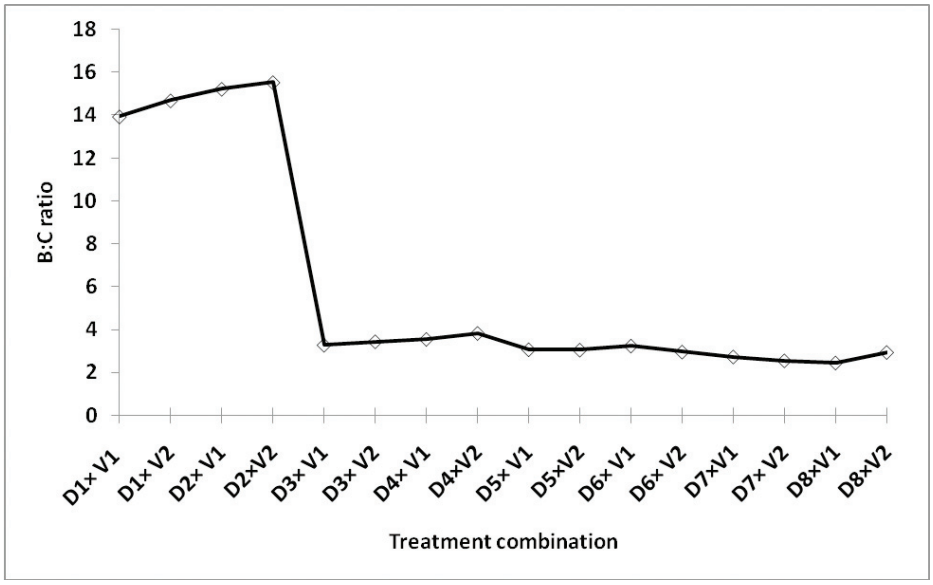
Period		Mean temperature (°C)		Relative humidity (%)		Wind velocity (km/hr)	Rainfall (mm)/ annum
Weekly data	Date	Max.	Min.	Max.	Min.		
44	26/11/19 to 02/12/19	26.5	12.5	97	55	1.4	0
45	03/12 19 to 09/12/19	24.9	8.2	96	44	1.2	0
46	10/12/19 to 16/12/19	22.2	10.4	96	63	1.5	3.08
47	17/12/19 to 23/12/19	17.9	7.9	93	67	3.2	0
48	24/12/19 to 30/12/19	15.2	5.7	92	66	2.1	0
49	01/01/20 to 07/01/20	20.4	8.3	95.4	55.3	1.9	0.65
50	08/01/20 to 14/01/20	17.7	7.1	96.7	72.3	2.5	1.74
51	15/01/20 to 21/01/20	17.7	10.2	98.7	87.6	2.0	9.68
52	22/01/20 to 28/01/20	20.8	6.6	93	49.7	3.1	0
53	29/01/20 to 04/01/20	22.3	7.2	92.1	50.4	3.6	0
54	05/02/20 to 11/02/20	22.9	5.3	92.9	73.6	2.1	0
55	12/02/20 to 18/02/20	25.6	9.5	88.1	36.1	4.3	0
56	19/02/20 to 25/02/20	26.5	12.1	93.4	57.6	2.5	1.31
57	26.02/20 to 04/03/20	26.7	12.7	95.8	51.5	1.9	0.2
58	05/03/20 to 11/03/20	26.3	13.2	89	55.3	4.1	3.54
59	2/03/20 to 18/03/20	27.0	14.0	89.4	57.9	2.5	2.57
60	19/03/20 to 25/03/20	31.2	16.5	87.7	36.9	2.7	0
61	26/03/20 to 01/04/20	31.8	18.1	74.9	35.4	7.4	0.68

Effect of different dates of transplanting on total soluble solids in bulb of onion cultivars

The perusal of data of the study significantly revealed that transplanting on 20th September (D_3) had highest TSS content in onion bulb followed by 30th October transplanting (D_7). Among the two cultivars, TSS was higher in L-883 (V_2) compared to the variety Agrifound Dark Red (V_1). The laboratory analysis recorded that maximum TSS was found with D_3xV_2 (6.35^{0}Brix 20th September transplanting x L-883 variety), followed by D_7 x V_1 (5.73 ^{0}Brix 30th October x Agrifound Dark Red), and the minimum TSS (4.42^{0}Brix) was observed in the bulb with D_1 x V_1 (30th August x Agrifound Dark Red). Total sugars was also obtained highest in V_2 when transplanted on 20th September where as V_1 showed the highest total sugars when transplanted on 30 October. Similar trend was also observed in case of reducing and non-reducing sugars. Ascorbic acid content was estimated and seen that it was very close among the two varieties, however, maximum ascorbic acid was found in variety L883 and transplanting date was 20th September. Whereas, variety V_1 (Agrifound Dark Red) showed maximum ascorbic acid when transplanted on 30 October. Likewise, very close difference was observed in acid content. These results are in close conformity with the findings of (Singh *et al*., 2000, Crowther *et al*., 2002) Mahanthesh *et al.* (2009), Patil *et al.* (2012) and Tripathy *et al.* (2014).

Economic analysis

The economic study (Fig. 1) revealed that different dates of transplanting had significant effect on gross return and net return for *kharif* onion production. It was also varied with varieties. Maximum gross and net return was obtained from variety V_2 (L- 883) when transplanted on 10th September (D_2) followed by V_1 (Agrifound Dark Red) transplanted on same day (D_2). The higher net return caused are marketable increase in Benefit: cost ratio of (18.58) and (12.47) followed by (18.11) and (12.32) as compared to late transplanting. The highest B:C ratio obtained in early transplanting also boosted up early harvesting when there was very high price for onion due to unavailability on onion in market. The high demand caused high return as well as high B:C ratio. However, at later



D_1-30th August, D_2-10th September, D_3-20th September, D_4-30th September, D_5-10th October, D_6-20th October, D_7-30th October, D_8-10th November; V_1- Agrifound Dark Red and V_2- L-883

Fig. 1: Benefit : cost ratio of onion production

stage of harvesting the market price was lower than early harvesting time and resulted less return and lower B: C ratio as compared to other treatments. It was also observed that in the second year of trial the market price was not so high all over the country and therefore, the net return and B: C ratio was comparatively lower than first year trial. Overall performance of *kharif* onion suggested that more return might be granted for *off season* production as was also stated by several scientist similar results like Nandal and Singh (2002), Kalhapure and Shete (2013) and Patel *et al.* (2011).

CONCLUSION

In the present investigation, it was noticed that maximum values for yield and bulb quality characters were observed in Variety L 883 when it was transplanted on 20th September (D_3 x V_2). However, it was also clear that all the varieties (V_1, V_2) produced higher yield when transplanted on 30th September (D_4) which exhibited maximum income and high B: C ratio (Fig.1). Although, there was sudden rise of price during August and mid September transplanting, but they had low yield than D_4. Therefore, it can be concluded that cultivar L-883 transplanting on 30th September can be suggested for off-season onion production in the subtropical agro-climatic region of Central Uttar Pradesh for good yield and quality but 10th September transplanting for more profit during this period of study

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Optimization of fertilizer for common fig (*Ficus carica*) cultivation in South western Bangladesh

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ABSTRACT

Egyptian common fig (Ficus carica) showed cultivation potential with year round bearing in Bangladesh. As fertilizer dose is a prerequisite for proper cultivation, an experiment was conducted in Batiaghata, Khulna, Bangladesh to recommend a fertilizer dose for successful fig cultivation. Two manures (farm yard manure, vermi compost) and different doses of NPK were applied. Different growth and yield contributing parameters were observed and benefit cost ratio was calculated. In response to applied fertilizer and manure, T_7 (Vermicompost (VC) @ 7.5kg + N @ 160g + P_2O_5 @ 105g + K_2O @ 105g plant^{-1}) showed superiority over other doses in both growth and yield parameters and also showed viable benefit cost ratio (3.95). T_7 produced 18.33 cm long and 16.52 cm wide leaves; 4.33 shoots (3.00 fruiting) and 65.44 cm plant height with 9.30 cm plant girth in an average. Results of all these parameters were about 1.5 folds than the control. Earliest fruit set (at 66.00 days), maximum number of fruits per plant (31.67) with the largest size and the highest weight (57 g fruit^{-1}),the earliest fruit maturity (4 days earlier than control) along with maximum yield (4.53 ton ha^{-1}) were recorded in T_7. The research concludes that, common fig (Ficus carica) can successfully and commercially be cultivated in south-western Bangladesh using 7.5 kg vermicompost with N @ 160 g + P_2O_5 @ 105 g + K_2O @ 105 g plant^{-1}.

***Keywords*:** Benefit cost ratio**,** Common fig, farm yard manure, fertilizer, vermicompost

INTRODUCTION

Figs (*Ficus carica* L.) are believed to be one of the oldest cultivated plants. For centuries, these fruits have been used fresh or dried as food for humans and animals. Figs can supply a lot of vitamins and minerals and can combat the hidden hunger caused by the micronutrient deficiency (Ashrafuzzaman *et al*., 2021). Figs have been popular not only because of their pleasant taste but possibly also because of their medicinal properties (Veberic and Petkovsek, 2016). But this fruit is not prevalent in Bangladesh, though some wild figs such as *Ficus pumila, Ficus recemosa*, *Ficus hispida* etc. grow spontaneously which are mostly used as animal food in the wild. In some specific locality, these figs are used as vegetables. In the world market, there is an increasing demand for fresh and dried figs for its high nutrient, mineral and polyphenol content (Salimpour *et al*., 2019).

Figs are deciduous subtropical species, native to arid, semi-desert regions and can be cultivated under rain fed conditions. Excellent table figs may grow where the temperature is moderate but it can be cultivated under a wider range of ecological conditions (Botti *et al*., 2003). Bangladesh is blessed with magnificent fertile land with a variety of soil type along with copious irrigation water and zestful human resources which may grant fig cultivation as a fruit crop. The soil and climatic conditions of Bangladesh was suggested to be suitable for commercial fig cultivation (Mehraj *et al.,* 2013). So, to mitigate the malnutrition problem year round fruits like figs are of immense importance and can successfully be inserted in the fruit production schedule of Bangladesh.

An optimum combination of fertilizers impacts a lot to determine the total growth. For successful cultivation of fig (*Ficus carica*), determination of fertilizer dose is essential. However, no effort has been taken for the optimization of fertilizer for fig cultivation in Bangladesh. To determine an optimum fertilizer dose, the influence of different

combination of organic (Farm yard manure and vermicompost) and inorganic (NPK) fertilizers on growth and yield of fig was studied.

MATERIALS AND METHODS

The experiment was conducted in Kismat Fultola village under Batiaghata Upazilla, Khulna, Bangladesh during August, 2020 to June, 2021. Weather data of the experimental site during experiment period, were presented in the Table 1. Two composite soil samples (on January and April) were collected from the experimental plot and analyzed in the laboratory of Soil Resource Development Institute, Daulatpur, Khulna under the Ministry of Agriculture, Peoples Republic of Bangladesh. The nutritional status of the soil has been shown in Table 2.

Egyptian variety of *Ficus carica* was used as plant material in this study. Saplings were grown from cuttings by using coco-peat as rooting media in the month of August. The saplings became ready and planted in the main field on mid-October. Land of the experimental site was prepared through weeding followed by 4-5 ploughing with power tiller and the soil of top 15 cm depth was made friable. Except nitrogen (applied at 10 days after planting) all of the fertilizers *viz.,* TSP and MOP along with 20g furadan (Carbofuran/carbamate pesticide) was applied as basal dose in 0.5m × 0.5 m× 0.5m planting holes. After receiving the assigned fertilizers (according to different treatments) pits were covered with soil and left for six weeks. After three weeks, the soil of the pits was mixed well and kept for another three weeks as before.

Healthy saplings of 30 days old were transplanted in the field at a distance of 2m×2 m on October 19, 2020, in the afternoon. Different intercultural operations were performed as per requirement. Mancer 75 WP (fungicide with Carbendazim 12% + Mancozeb 63%) at 2g per liter and Intrepid 10 SC (insecticide) at 1 ml per liter were sprayed on the plants at 10 days interval with the beginning of nitrogenous fertilizer application (10 Days after planting) and ended at 70 days after transplanting. On 30 days after planting and 60 days after planting, Thiovit 80 WG (Sulphur fungicide) was applied with Intrepid instead of Mancer 75 WP.

The fruits were harvested when they were fully matured and turned to pinkish red in color. Harvesting of fruits was started at 121 days after transplanting and continued up to 165 days with an interval of 3 days. Harvesting was usually done by hands. Fruits immerged after 20 days of the first fruit set were considered for yield count.

Design of experiment and treatments

Randomized Complete Block Design (RCBD) was followed with three replications for the treatments containing one plant in each replication. The treatments were as follows:

T_0 = Control
T_1 = Farm Yard Manure @ 12.5kg + N @ 160g + P_2O_5 @ 105 g + K_2O @ 105g plant^{-1}
T_2 = FYM @ 12.5kg + N @ 200 g + P_2O_5 @ 130 g + K_2O @ 130g plant^{-1}
T_3 = FYM @ 12.5kg + N @ 240 g + P_2O_5 @ 160 g + K_2O @ 160 g plant^{-1}
T_4 = FYM @ 25 kg + N @ 160 g + P_2O_5 @ 105 g + K_2O @ 105 g plant^{-1}
T_5 = FYM @ 25 kg + N @ 200 g + P_2O_5 @ 130 g + K_2O @ 130 g plant^{-1}
T_6 = FYM @ 25 kg + N @ 240 g + P_2O_5 @ 160 g + K_2O @ 160 g plant^{-1}
T_7 = Vermicompost (VC) @ 7.5kg + N @ 160g + P_2O_5 @ 105g + K_2O @ 105g plant^{-1}
T_8 = VC @ 7.5 kg + N @ 200 g + P_2O_5 @ 130 g + K_2O @ 130 g plant^{-1}
T_9 = VC @ 7.5 kg + N @ 240 g + P_2O_5 @ 160 g + K_2O @ 160 g plant^{-1}
T_{10} = VC @ 15 kg + N @ 160 g + P_2O_5 @ 105 g + K_2O @ 105 g plant^{-1}
T_{11} = VC @ 15 kg + N @ 200 g + P_2O_5 @ 130 g + K_2O @ 130 g plant^{-1}
T_{12} = VC @ 15 kg + N @ 240 g + P_2O_5 @ 160 g + K_2O @ 160 g plant^{-1}

Nitrogenous fertilizer (urea) was applied in three splits viz. at 10, 35 and 60 days after transplanting. P_2O_5 was applied in the form of Triple Super Phosphate (TSP) and K_2O was through Muriate of Potash (MoP).

Observations recorded

Growth parameters

For growth parameters, data were recorded at 30 days interval up to 90 days after planting and for yield contributing parameters from fruit set to harvest. Data were collected on growth parameters like days required for first leaf emergence, number of leaves (total leaves which were fully opened, matured and not senescent at the day of data recording including the leaves prevailing during planting), number of nodes, leaf length (cm) (from the base of the petiole to the leaf apex along the midrib), leaf width (cm) (the most spacious part of leaf blades), number of shoot, plant height (cm) (from the collar of the plant to the terminal growing point of the main stem) and shoot girth (measured at the base of the plant). From each plant, 5 leaves were taken and the average length and width were expressed in centimeters.

Yield contributing parameters

Recorded yield parameters were - days required for fruit set (the day when it was clear that the bud developed on the leaf axis is a fruit not shoot), number of fruiting shoot, number of fruits per plant, average fruit length (mm), average fruit diameter (mm), average fruit girth (mm), average fruit weight (g), total fruit per plant (kg) and yield per hectare (ton). Fruit length and diameter were measured with a digital slide calipers and girth was measured with a centimeter tape. Data of each fruit was recorded and the average of all the treatments was calculated. The fruits were harvested when they reached at their desired shape and color. From bud initiation to harvesting of the fruits, the days were calculated for each fruit and the average days needed to mature were determined for each treatment.

Fruit yield

Fruit yield per plant (kg) was determined by adding the total fruit weight over all of the pickings from each treatment and was expressed in kilograms (kg). The fruit yield plant^{-1} of *Ficus carica* was converted into per hectare and has been expressed in tonnes (t). It was measured by using the following formula :

$$\text{Yield (ton) per hectare} = \frac{\text{Fruit yield per plant (kg)} \times 10000\text{m}^2}{\text{Area of plot in square meter (m}^2\text{)} \times 1000(kg)}$$

Benefit cost ratio (BCR)

Production cost includes all expenditures including land rent, labor, power tiller rent, cost for sapling, fertilizer, transportation and all other management costs up to marketing and sales. The economic indicator BCR was calculated using following formula for each treatment.

$$\text{Benefit cost ratio} = \frac{\text{Gross income(Tk)}}{\text{Total cost of production(Tk)}}$$

Here, "Gross income" means the total selling price without deducting the production cost.

Statistical analysis

Collected data were subject to two way analysis of variance (ANOVA) by Statistical Tool for Agricultural Research (STAR) (IRRI, 2013). The effects of various treatments were assessed within ANOVA and the level of significance was tested by Tukeys's Honest Significant Difference (HSD) following significant ($P \leq 0.01$) F test. The assumptions on normality of data and homogeneity of variance were checked to ensure the validity of analysis.

RESULTS AND DISCUSSION

Effect of manures and fertilizers on leaf parameters

Earliest leaf initiation (9.67 days) was observed in T_{12} (VC @ 15 kg + N @ 240 g + P_2O_5 @ 160 g + K_2O @ 160 g plant^{-1}) followed by T_{11} (11.67 days) which is statistically similar to T_6, T_8 and T_9 (13.00, 13.00 and 13.33 days respectively). Most delayed first leaf initiation (18.67 days) was witnessed in control (T_0) (Table 3).

Similarly, T_{12} resulted in maximum number of leaf production (47.78) that was followed by T_{11} (45.44), T_9 (43.22), T_{10} (42.33), T_6 (42.11)and T_8 (41.22). Control produced lowest number of leaves (30.00).Though lower dosed of VC and combination of NPK in T_7 (VC @ 7.5 kg + N @ 160 g + P_2O_5 @ 105 g + K_2O @ 105 g plant^{-1}) exhibited best performance for leaf length and width (18.33 cm and 16.52 cm, respectively) followed

Table 1: Weather conditions of study area from August 2020 to June 2021 (BMD, 2020 and 2021)

Month	Monthly average max. temperature (°C)	Monthly average min. temperature (°C)	Monthly average relative humidity (%)	Monthly total rainfall (mm)
August 2020	32.8	27.0	86	265
September 2020	33.6	27.0	85	172
October 2020	33.2	26.2	83	90
November 2020	30.7	20.0	76	06
December 2020	26.1	15.0	80	00
January 2021	26.1	14.2	79	00
February 2021	29.5	15.9	73	03
March 2021	34.4	22.4	67	00
April 2021	36.4	24.9	67	02
May 2021	35.4	26.0	73	124
June 2021	32.6	26.3	85	468

Table 2: Nutritional status of the experimental plot

Sl. no	Soil property	Field soil -1 (January)	Field soil-2 (April)
01	P^H	8.2	8.1
02	Salinity (dSm^1)	3.9	4.7
03	Organic matter (%)	1.57	2.04
04	Total Nitrogen (%)	0.091	0.119
05	Phosphorus µgm gm^1 soil	25.35	15.05
06	Potassium mleq. gm^1 soil	0.19	0.20
07	Sulphur µgm gm^1 soil	76.01	105.34
08	Zinc µgm gm^1 soil or %	3.17	1.19
09	Boron µgm gm^1 soil	0.50	10.2

by higher doses of FYM and combinations of NPK in T_4(13.55 cm and 11.76 cm) and T_5 (13.22 cm and 11.82 cm) without any significant difference between them. On the other hand, shortest (10.43 cm) and narrowest (9.03 cm) leaves were found in the plants treated with higher doses of VC and NPK combinations (Table 3).

The leaf initiation started before the application of nitrogenous fertilizer that means leaf initiation was influenced by initial application of fertilizers as FYM, Vermicompost, TSP and MoP. This result was in conformity to Mordogan *et al. (*2013). Different fertilizer combination displayed significant effect on leaf characters of fig. With the higher rate of fertilizers, all leaf parameters showed better performance initially (on 30 DAT. But with the course of time (on 90 DAS) they decreased gradually except leaf number and leaves became smaller, thicker, dark green and leathery in higher doses. This result is in accordance with Khan *et al*. (2020) Pal *et al*. (2015) and Blouin *et al*. (2019). They reported small and thick leaves caused by excessive nitrogen applied and good impact of FYM and vermicompost in combination with recommended dose of fertilizer on nutrient availability and nitrogen dynamics which ultimately influences crop growth and yield.

Shoot response of *F. carica* to different combination of manures and fertilizers

Like leaf parameters, shoot parameters were also significantly influenced by different manure and fertilizer combinations. Node number was highest in T_{12} (50.33) followed by T_{11} (48),T_9 (46.11) and $T_{10.}$ (45.11) while maximum shoot number was noted in T_7 (4.13) followed by T_1 (3.45), T_2 (3.22) and T_4 (3.22) (Table 4) which are statistically similar. Again, highest plant height (65.44 cm) and plant girth (9.3 cm) were also obtained from T_7

Table 3: Response of the *Ficus carica* leaf to different combination of manure and fertilizer

Treatment	days to leaf initiation	Leaf number			Date mean	Leaf length (cm)			Date mean	Leaf width (cm)			Date mean
		30 DAT	60 DAT	90 DAT		30 DAT	60 DAT	90 DAT		30 DAT	60 DAT	90 DAT	
T_0	18.67 a	11.67 g	29.33 h	49.00 g	30.00 H	7.67 cd	13.33 bc	14.33 cd	11.78 BC	6.67 c-e	11.33 cd	12.97 b-d	10.32 B
T_1	16.67 b	13.33 fg	34.33 g	54.33 f	34.00 G	7.00 d	15.33 b	17.00 b	13.11 B	5.17 e	15.20 b	15.67 b	12.01 B
T_2	15.00 c	15.33 ef	36.00 g	56.33 ef	35.89 F	9.33 b-d	11.00 cd	12.67 de	11.00 C	6.67 c-e	9.27 de	11.00 c-e	8.98 C
T_3	14.67cd	18.00 cd	39.67 ef	58.33 de	38.67 E	9.67 bc	10.00 d	11.67 ef	10.45 C	6.83 b-e	7.73 e	10.03 de	8.20 C
T_4	13.67 c-e	16.33 d-f	44.33 bc	55.33 ef	38.67 E	9.33 b-d	15.00 b	16.33 bc	13.55 B	6.50 de	13.27 bc	15.50 b	11.76 B
T_5	13.33 de	17.67 c-e	45.67 a-c	58.00 de	40.45 D	10.00 a-c	14.33 b	15.33 bc	13.22 B	8.23 a-e	13.60 bc	13.63 bc	11.82 B
T_6	13.00 ef	18.67 cd	47.33 ab	60.33 d	42.11 CD	10.67 ab	10.00 d	11.67 ef	10.78 C	8.07 a-e	8.67 cd	9.93 de	8.89 C
T_7	13.33 de	15.00 ef	37.33 fg	65.00 c	39.11 E	11.67 ab	21.33 a	22.00 a	18.33 A	9.8 a-c	19.33 a	20.43 a	16.52 A
T_8	13.00 ef	16.33 d-f	41.00 de	66.33 bc	41.22 D	11.00 ab	13.33 bc	14.33 cd	12.89 B	8.7 a-d	11.67 cd	12.90 b-d	11.09 B
T_9	13.00 ef	20.33 bc	43.33 cd	66.00 bc	43.22 C	10.00 a-c	11.00 cd	12.67 de	11.22 C	9.03 a-d	9.00 de	11.5 c-e	9.84 C
T_{10}	13.33 de	18.67 cd	41.00 de	67.33 bc	42.33 C	11.33 ab	13.33 bc	14.67 b-d	13.11 B	10.00 ab	11.33 cd	13.83 bc	11.72 B
T_{11}	11.67 f	22.00 ab	45.33 a-c	69.00 ab	45.44 B	12.33 a	13.00 bc	14.00 c-e	13.11 B	10.93 a	11.33 cd	12.53 b-d	11.60 B
T_{12}	9.67 g	23.67 a	48.00 a	71.67 a	47.78 A	12.33 a	9.33 d	9.67 f	10.44 C	10.00 ab	8.43 de	8.67 e	9.03 C
Treatment mean			17.46 *C*	40.97 *B*	61.31 *A*		10.18 *C*	13.10 *B*	14.33 *A*		8.2 *C*	11.55 *B*	12.97 *A*

HSD (*P*d"0.01)for days to leaf initiation=1.57; for leaf number: comparison of date at each level of treatment=1.77, comparison of treatment at each level of date = 3.05; for leaf length: comparison of date at each level of treatment= 1.40, comparison of treatment at each level of date = 2.40; for leaf width: comparison of date at each level of treatment =1.90, comparison of treatment at each level of date =3.27* Means with the same lower-case or upper-case letter in rows or columns are not significantly different at $P \leq 0.01$ by the Tukeys's Honest Significant Difference (HSD)Test.T_0 = Control, T_1 = Farm Yard Manure (FYM) @ 12.5kg + N @ 160g + P_2O_5 @ 105 g + K_2O @ 105 g plant^{-1}, T_2 = FYM @ 12.5kg + N @ 200 g + P_2O_5 @ 130 g + K_2O @ 130g plant^{-1}, T_3 =FYM @ 12.5kg + N @ 240 g + P_2O_5 @ 160 g + K_2O @ 160 g plant^{-1}, T_4 = FYM @ 25 kg + N @ 160 g + P_2O_5 @ 105 g + K_2O @ 105 g plant^{-1}, T_5 = FYM @ 25 kg + N @ 200 g + P_2O_5 @ 130 g + K_2O @ 130 g plant^{-1}, T_6 = FYM @ 25 kg + N @ 240 g + P_2O_5 @ 160 g + K_2O @ 160 g plant^{-1}, T_7 = Vermicompost (VC) @ 7.5kg + N @ 160g + P_2O_5 @ 105g + K_2O @ 105g plant^{-1}, T_8 =VC @ 7.5 kg + N @ 200 g + P_2O_5 @ 130 g + K_2O @ 130 g plant^{-1}, T_9 = VC @ 7.5 kg + N @ 240 g + P_2O_5 @ 160 g + K_2O @ 160 g plant^{-1}, T_{10}=VC @ 15 kg + N @ 160 g + P_2O_5 @ 105 g + K_2O @ 105 g plant^{-1}, T_{11} =VC @ 15 kg + N @ 200 g + P_2O_5 @ 130 g + K_2O @ 130 g plant^{-1}, T_{12} =VC @ 15 kg + N @ 240 g + P_2O_5 @ 160 g + K_2O @ 160 g plant^{-1}, DAT = Days after transplanting

Table 4: Response of the *Ficus carica* node and shoot number to different combination of manure and fertilizer

Treatment	Node number			Date mean	Shoot number			Date mean
	30 DAT	60 DAT	90DAT		30 DAT	60 DAT	90 DAT	
T_0	11.67 h	29.67 h	52.67 g	31.34 G	1.33 b	3.00 b-d	4.00 b-d	2.78 B
T_1	13.67 gh	36.33 g	58.00 f	36 F	2.00 ab	3.67 ab	4.67 ab	3.45 B
T_2	16.33 e-g	38.33 fg	59.00 ef	37.89 F	1.33 b	3.67 ab	4.67 ab	3.22 B
T_3	17.67 d-f	42.33 de	61.33 de	40.44 E	1.33 b	2.00 d	3.00 d	2.11C
T_4	17.00 d-f	47.00 bc	59.00 ef	41D E	2.00 ab	3.33 bc	4.33 bc	3.22 B
T_5	18.00 d-f	48.67 ab	62.00 de	42.89 D	1.67 ab	2.33 cd	3.33 cd	2.44 C
T_6	19.00 c-e	49.67 ab	64.00 d	44.22 C	2.00 ab	2.33 cd	3.33 cd	2.55 C
T_7	15.00 fg	40.00 ef	69.00 c	41.33 D	2.67 a	4.67 a	5.67 a	4.33 A
T_8	16.67 e-g	44.00 cd	70.33 bc	43.67 C	2.00 ab	3.33 bc	3.67 b-d	3.00 B
T_9	21.33 a-c	47.00 bc	70.00 bc	46.11 BC	1.67 ab	3.00 b-d	4.00 b-d	2.89 B
T_{10}	20.00 b-d	44.00 cd	71.33 bc	45.11 C	1.67 ab	2.67 b-d	3.67 b-d	2.67 C
T_{11}	22.67 ab	48.33 ab	73.00 ab	48 B	1.00 b	2.00 d	3.00 d	2.00 C
T_{12}	24.33 a	51.00 a	75.67 a	50.33 A	1.00 b	2.00 d	3.00 d	2.00 C
Treatment mean	**17.95 *C***	**43.56 *B***	**65.03 *A***		**1.67 *C***	**2.92 *B***	**3.87 *A***	

* Means with the same lower-case or upper-case letter in rows or columns are not significantly different at $P \leq 0.01$ by the Tukeys's Honest Significant Difference (HSD) Test. HSD (*P*d"0.01) for node number: comparison of date at each level of treatment=1.92, comparison of treatment at each level of date = 3.30; for shoot number: comparison of date at each level of treatment =0.73, comparison of treatment at each level of date =1.25T_0 = Control, T_1 = Farm Yard Manure (FYM) @ 12.5kg + N @ 160g + P_2O_5 @ 105 g + K_2O @ 105 g plant^{-1}, T_2 = FYM @ 12.5kg + N @ 200 g + P_2O_5 @ 130 g + K_2O @ 130g plant^{-1}, T_3 =FYM @ 12.5kg + N @ 240 g + P_2O_5 @ 160 g + K_2O @ 160 g plant^{-1}, T_4 = FYM @ 25 kg + N @ 160 g + P_2O_5 @ 105 g + K_2O @ 105 g plant^{-1}, T_5 = FYM @ 25 kg + N @ 200 g + P_2O_5 @ 130 g + K_2O @ 130 g plant^{-1}, T_6 = FYM @ 25 kg + N @ 240 g + P_2O_5 @ 160 g + K_2O @ 160 g plant^{-1}, T_7 = Vermicompost (VC) @ 7.5kg + N @ 160g + P_2O_5 @ 105g + K_2O @ 105g plant^{-1}, T_8 =VC @ 7.5 kg + N @ 200 g + P_2O_5 @ 130 g + K_2O @ 130 g plant^{-1}, T_9 = VC @ 7.5 kg + N @ 240 g + P_2O_5 @ 160 g + K_2O @ 160 g plant^{-1}, T_{10} =VC @ 15 kg + N @ 160 g + P_2O_5 @ 105 g + K_2O @ 105 g plant^{-1}, T_{11} =VC @ 15 kg + N @ 200 g + P_2O_5 @ 130 g + K_2O @ 130 g plant^{-1}, T_{12} =VC @ 15 kg + N @ 240 g + P_2O_5 @ 160 g + K_2O @ 160 g plant^{-1}, DAT = Days after transplanting

(Table 5). In all growth parameters control showed lowest values.

Plots, where higher doses of NPK[@ 200, 130 and 160 g plant^{-1}were applied (T_{12})], resulted about 0.93 time lower height and 0.71 time lower girth than the plants receiving the lower doses of these fertilizers in T_7 (Table 5). Similarly, lower number of nodes and shoots were also recorded in the plants receiving higher doses of fertilizers (Table 4).Excessive P significantly affects leaf growth and chlorophyll concentration (Shi *et al*., 2020) and excess K found to be inhibitory to growth and root development by Xu *et al*.(2020).

The leaf and node formation might be increased by high dose of fertilizer but shorter internodes lowered the plant height. Similar findings were reported by Sun *et al*. (2020); Shi *et al*. (2020) and Xu, *et al*. (2020). High P in high soil p^H (>6.5; Table 2) induces micronutrient deficiencies which results leaf dwarfing. Besides, high rate of nitrogen significantly decreases phosphorus and potassium availability along with lowering glutamine synthesized activity (Sun *et al*., 2020). The applied NPK and nitrogen fixing bacteria from vermicompost, might have improved the fertility, aeration, water holding capacity, mineral uptake and synthesis of growth hormones (Mengel *et al.,* 2001).

Yield performance of *F. carica* to different combination of manure and fertilize

Earlier fruit set occurred in plants (6.33 to 27.33 days earlier) receiving lower doses of VC and NPK in T_7 (VC @ 7.5 kg + N @ 160 g + P_2O_5 @ 105 g + K_2O @ 105 g) than rest of the fertilizer combinations followed by T_1 (72.33 DAT) where lowest dose of NPK (N160g+$P_2O_5$105g+K_2O105g plant^{-1}) was applied with only 12.5 kg FYM. Most delayed fruit set (93.33 DAT) was resulted from

Table 5: Response of the *Ficus carica* plant height and plant girth to different combination of manure and fertilizer

Treatment	Plant height (cm)			Date mean	Plant girth (cm)			Date mean
	30 DAT	60 DAT	90 DAT		30 DAT	60 DAT	90 DAT	
T_0	16.33 g	41.33 f	81.67 e	46.44 E	5.17	7.17	9.57	7.30 CDE
T_1	18.33 fg	43.33 ef	83.33 de	48.33 E	5.97	7.73	10.07	7.92 C
T_2	19.33 e-g	44.67 d-f	84.67 de	49.56 D	5.50	7.57	9.77	7.61CD
T_3	20.67 d-f	45.67 c-e	85.93 cd	50.76 D	5.27	7.30	9.57	7.38 CDE
T_4	22.00 c-e	47.00 cd	86.67 cd	51.89 CD	5.87	7.90	9.90	7.89 C
T_5	23.00 cd	49.00 c	89.23 c	53.74 C	5.83	7.90	10.53	8.09 BC
T_6	24.67 c	48.33 c	88.33 c	53.78 C	5.23	7.30	9.50	7.34 CDE
T_7	33.00 ab	58.33 ab	105.00 a	65.44 A	7.00	9.27	11.63	9.30 A
T_8	35.00 a	60.00 a	98.67 b	64.56 A	6.80	8.86	11.07	8.91 AB
T_9	32.00 ab	57.00 ab	97.13 b	62.04 B	6.87	8.83	11.07	8.92 AB
T_{10}	34.00 ab	59.00 ab	98.83 b	63.94 AB	5.43	7.70	10.10	7.74 C
T_{11}	31.67 ab	56.67 ab	96.17 b	61.50 B	4.77	6.57	8.80	6.71 DE
T_{12}	31.00 b	56.00 b	96.47 b	61.16 B	4.67	6.40	8.67	6.58 E
Treatment mean	**26.23 *C***	**51.26 *B***	**91.70 *A***			**5.72 *C***	**7.73 *B***	**10.02 *A***

* Means with the same lower-case or upper-case letter in rows or columns are not significantly different at $P \leq 0.01$ by the Tukeys's Honest Significant Difference (HSD) Test. HSD ($P \leq 0.01$) for plant height: comparison of date at each level of treatment=2.08, comparison of treatment at each level of date = 3.57; for plant girth: comparison of date at each level of treatment =0.26, comparison of treatment at each level of date =0.92T_0 = Control, T_1 = Farm Yard Manure (FYM) @ 12.5kg + N @ 160g + P_2O_5 @ 105 g + K_2O @ 105 g plant^{-1}, T_2 = FYM @ 12.5kg + N @ 200 g + P_2O_5 @ 130 g + K_2O @ 130g plant^{-1}, T_3 =FYM @ 12.5kg + N @ 240 g + P_2O_5 @ 160 g + K_2O @ 160 g plant^{-1}, T_4 = FYM @ 25 kg + N @ 160 g + P_2O_5 @ 105 g + K_2O @ 105 g plant^{-1}, T_5 = FYM @ 25 kg + N @ 200 g + P_2O_5 @ 130 g + K_2O @ 130 g plant^{-1}, T_6 = FYM @ 25 kg + N @ 240 g + P_2O_5 @ 160 g + K_2O @ 160 g plant^{-1}, T_7 = Vermicompost (VC) @ 7.5kg + N @ 160g + P_2O_5 @ 105g + K_2O @ 105g plant^{-1}, T_8 =VC @ 7.5 kg + N @ 200 g + P_2O_5 @ 130 g + K_2O @ 130 g plant^{-1}, T_9 = VC @ 7.5 kg + N @ 240 g + P_2O_5 @ 160 g + K_2O @ 160 g plant^{-1}, T_{10} =VC @ 15 kg + N @ 160 g + P_2O_5 @ 105 g + K_2O @ 105 g plant^{-1}, T_{11} =VC @ 15 kg + N @ 200 g + P_2O_5 @ 130 g + K_2O @ 130 g plant^{-1}, T_{12} =VC @ 15 kg + N @ 240 g + P_2O_5 @ 160 g + K_2O @ 160 g plant^{-1}, DAT = Days after transplanting

the plants applied with higher doses of FYM and NPK fertilizers in T_6 (25kg FYM+ 240 g N+160g P_2O_5 + 160g K_2O plant^{-1}) followed by T_5 (90.33 DAT) and T_{12} (higher doses of VC and NPK) (90.33 DAT) (Table 6).

Number of fruiting shoot (3.00) and fruit number (31.67) was also highest in the plants treated with T_7 which was about three times more than control (T_0) (1.33 and 11.67 respectively). Second highest fruiting shoot (2.00) was found in T_1, T_4 andT_8 and they are statistically analogous to T_0(1.33), T_2(1.33), T_9 (1.67) and T_{10} (1.67). Lowest number of fruit per plant was produced by T_3 (9.33) (Table 6).

Organic manure improves soil physical properties by increasing soil porosity and decreasing soil bulk density (Shah *et al*., 2023) which ultimately improves root environment, nutrient uptake and vigorous growth resulted highest number of fruits per plant (Rabindra *et al*., 2021). Shamsuddin *et al*. (2020) also reported higher yield with higher rate of organic manure.

Largest fruits were produced in T_7 in respect of fruit length (61.33 mm), fruit diameter (55.00 mm) and fruit girth (168.90 mm) followed by T_{10} (length 56.67mm, diameter 51.33 mm and girth 148.03 mm). The lowest reading was in the fruits harvested from T_6 (36.67mm, 32.67 mm and 94.43 mm, respectively). The treatment T_7 showed superiority for fruit weight (57.00 g) over all other fertilizer compositions followed by T_{10} (51.67 g) and T_9 (45.33 g). Average fruit weight from T_7 was recorded as 57.00 g i.e. 5.33g to 27.00g more than fruit weight produced by other treatments. Total harvested fruit per plant (1.81 kg plant^{-1}) and yield per hectare (4.53 ton ha^{-1}) were also highest in T_7 followed by T_{10} (1.18 kg plant^{-1} and 2.94 ton ha^{-1}) (Table 6).

Higher leaf length and width formed from organic fertilizer (vermicompost or FYM) in

Table 6: Response of the yield parameters of *Ficus carica* to different combination of manure and fertilizer

Treatment	Days to fruit set	Number of fruiting shoot	Number of fruit	Fruit length (mm)	Fruit diameter (mm)	Fruit girth (mm)	Days to fruit maturity	Average fruit weight (g)	Total fruit per plant (kg)	yield per hectare (ton)	Cost benefit ratio
T_0	75.00 g	1.33 bc	11.67 e	42.00 e	39 .00 de	122.37 cd	58.00 de	33.00 de	0.38 d	0.96 d	1.23 cd
T_1	72.33 h	2.00 b	21.33 bc	47.00 d	50.67 ab	155.33 b	60.00 cd	41.00 c	0.88 c	2.19 c	2.16 b
T_2	79.00 ef	1.33 bc	12.00 e	41.00 ef	38.00 ef	116.50 de	65.33 b	30.00 e	0.36 d	0.89 d	0.87 de
T_3	83.00 d	1.00 c	9.33 e	39.33 ef	34.33 fg	106.77 ef	68.33 a	31.00 e	0.29 d	0.72 d	0.69 e
T_4	76.33 g	2.00 b	24.00 b	38.00 ef	34.00 fg	104.53 ef	59.33 cde	32.67 de	0.79 c	1.97 c	1.53 c
T_5	90.33 b	1.00 c	11.67 e	38.33 ef	34.33 fg	103.30 f	67.00 ab	33.67 de	0.39 d	0.98 d	0.75 e
T_6	93.33 a	1.00 c	10.00 e	36.67 f	32.67 g	94.43 f	68.00 a	31.67 de	0.32 d	0.79 d	0.61 e
T_7	66.00 i	3.00 a	31.67 a	61.33 a	55.00 a	168.90 a	54.00 f	57.00 a	1.81 a	4.53 a	3.95 a
T_8	79.67 e	2.00 b	16.33 d	49.67 d	43.00 cd	130.63 c	57.67 e	44.33 c	0.71 c	1.79 c	1.50 c
T_9	82.33 d	1.67 bc	10.33 e	48.67 d	42.00 cde	131.30 c	61.00 c	45.33 bc	0.47 d	1.67 d	1.01 de
T_{10}	77.00 fg	1.67 bc	23.00 b	56.67 b	51.33 ab	148.03 b	59.33 cde	51.67 ab	1.18 b	2.94 b	1.99 b
T_{11}	87.67 c	1.00 c	19.00 cd	54.33 bc	49.33 b	132.17 c	65.33 b	44.33 c	0.85 c	2.12 c	1.43 c
T_{12}	90.33 b	1.00 c	11.67 e	50.33 cd	44.67 c	131.00 c	67.33 ab	38.33 cd	0.45 d	1.13 d	0.75 e
HSD	2.48	0.97	3.53	4.37	4.34	12.45	2.32	7.03	0.21	0.52	0.42

* Means with the same lower-case or upper-case letter in rows or columns are not significantly different at $P \leq 0.01$ by the Tukeys's Honest Significant Difference (HSD) Test.T_0 = Control, T_1 = Farm Yard Manure (FYM) @ 12.5kg + N @ 160g + P_2O_5 @ 105 g + K_2O @ 105 g plant^{-1}, T_2 = FYM) @ 12.5kg + N @ 200 g + P_2O_5 @ 130 g + K_2O @ 130g plant^{-1}, T_3=FYM @ 12.5kg + N @ 240 g + P_2O_5 @ 160 g + K_2O @ 160 g plant^{-1}, T_4= FYM @ 25 kg + N @ 160 g + P_2O_5 @ 105 g + K_2O @ 105 g plant^{-1}, T_5= FYM @ 25 kg + N @ 200 g + P_2O_5 @ 130 g + K_2O @ 130 g plant^{-1}, T_6 = FYM @ 25 kg + N @ 240 g + P_2O_5 @ 160 g + K_2O @ 160 g plant^{-1}, T_7= Vermicompost (VC) @ 7.5kg + N @ 160g + P_2O_5 @ 105g + K_2O @ 105g plant^{-1}, T_8 =VC @ 7.5 kg + N @ 200 g + P_2O_5 @ 130 g + K_2O @ 130 g plant^{-1}, T_9 = VC @ 7.5 kg + N @ 240 g + P_2O_5 @ 160 g + K_2O @ 160 g plant^{-1}, T_{10} =VC @ 15 kg + N @ 160 g + P_2O_5 @ 105 g + K_2O @ 105 g plant^{-1}, T_{11} =VC @ 15 kg + N @ 200 g + P_2O_5 @ 130 g + K_2O @ 130 g plant^{-1}, T_{12} =VC @ 15 kg + N @ 240 g + P_2O_5 @ 160 g + K_2O @ 160 g plant^{-1}

combination with lower rates of NPK fertilizers which enlarged the photosynthetic area and photosynthete accumulation that yielded larger fruit with higher weight and ultimately triggered the yield hectare^{-1}. Kurubar *et al*. (2017) also inferred that superior fruit characters were found in fig when both organic and inorganic fertilizers were applied together. Choudhury *et al*. (2020) also reported significant positive effect of organic manure (FYM, vermicompost and mustard cake) on yield and quality of sapota.

It is pivotal for any commodity to ensure that the particular product is marketable at the right time. Plants that received 7.5 kg vermicompost with 160g N, 105g P and 105g K (T_7) showed 3.67 to 14.33 days early maturity of fruit than rest of the treatments followed by T_8 (57.67 DAT) without significant difference with T_0 (58 DAT), T_4 (59.33 DAT) and T_{10} (59.33 DAT).Most delayed maturity was observed in T_3 (68.33 DAS) followed by T_6 (68 DAT),where maximum chemical fertilizer was used along with FYM. Statistically similar result was exhibited by T_{12} (67.33 DAT) where highest dose of vermicompost (15 kg) along with highest amount of NPK were used (Table 6). Naik (2005) also recorded delayed flowering and maturity in the high nitrogen treated capsicum plants.

Benefit cost ratio (BCR)

Maximum benefit cost ratio (3.95) was also observed in T_7 followed by T_1 (2.16) and T_{10} (1.99). The ratio derived from T_7 was about three times more than that of control. Lowest ratio was resulted from T_3 (Table 6). Due to increased rate of fertilizer, plant growth and yield might be inhibited. Moreover, increased fertilizer incurred higher production cost with lower harvest. Lokappa *et al*. (2018) reported that the investment in fig orchard will be economically viable with a benefit cost ratio of > 3. The difference is that, they measured the payback for three years and in the current study output from the orchard was measured for only 8 months from planting the saplings.

CONCLUSION

Common fig (*Ficus carica*) is not very fertilizer loving plant. It can produce fruits even in control in Bangladesh but a combination of vermicompost with a low level of NPK can yield best. Among all treatments, 7.5 kg vermicompost with N @ 160 g + P_2O_5 @ 105 g + K_2O @ 105 g plant^{-1}performed as the best combination in respect of better yield and economic return.

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Response of natural rooting substances on leaf cuttings of two cultivars of ZZ plant (*Zamioculcas zamiifolia*)

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ABSTRACT

The present investigation on "Multiplication of ZZ (Zamioculcas zamiifolia) cultivar leaf cutting using different natural rooting substances" was carried out during the year 2021-2022 at Horticultural Farm, School of Agricultural Sciences and Rural Development, Medziphema, Nagaland. The experiment was laid out in completely randomized design with 12 treatments C_1- ZZ Super Nova and C_2- ZZ Raven, and different natural rooting substances (T) viz., Control (T_0), Honey (T_1), Cinnamon powder (T_2), Aloe vera gel (T_3), Apple cider (T_4), Coconut water (T_5)and replicated thrice. The leaf cutting of two cultivars (Super Nova and Black Raven) were dipped in different prepared natural rooting substances, Honey, Cinnamon, Aloe vera, Apple cider, Coconut water and control, and planted in coarse sand. Early rhizome initiation (45.54 days), root initiation (52.09 days), number of roots at 75 and150 DAP (4.46 and 8.04), length of primary root at 75 and 150 DAP (1.88cm and 3.65cm), rate of root growth at 75 and 150 DAP (71.76% and 86.55%), diameter of rhizome at 90, 120 and 150 DAP (0.93cm, 2.10cm and 2.49cm) days to shoot bud emergence (182.76) and shoot length at 45 days after emergence (2.31cm) were recorded in C_1(Super Nova). While T_5 (Coconut water) exhibited the highest values in all the growth characters viz. rhizome initiation (46.50 days), root initiation (50.73 days), number of roots at 75 and 150 days after planting (5.33 and 8.73), length of primary root at 75 and 150 DAP (2.45cm and 4.93cm), rate of root growth at 75 and 150 DAP (78.59% and 89.44%), diameter of rhizome at 90, 120 and 150 DAP (1.05cm, 2.10cm and 2.46cm) days to shoot bud emergence (182.20) shoot length at 45 days after emergence (3.09cm). The leaf cutting of ZZ Super Nova grows much faster than ZZ Black Raven when given with same soil and climate condition.

***Keywords*:** *Aloe vera, apple cider, cinnamon, coconut water, honey, leaf cutting, multiplication, Zamioculcas zamiifolia*

INTRODUCTION

Zamioculcas zamiifolia, known by several common names such as African cootie, aroid palm, arum fern, cardboard palm, emerald frond, and ZZ plant, is a stemless tropical herbaceous perennial native to submountain and lowlands forests of eastern Africa (Chen *et al.*, 2002). It was discovered in the year 1905 (Mayo *et al.,* 1998) and the only species belonging to the genus *Zamioculcas* (Chen *et al.,* 2005). *Zamioculcas* has thick, fleshy petioles supporting attractive dark green and glossy alternate pinnate leaflets. Petioles arise from succulent rhizomes that can range from 0.4 to 10 cm in diameter. Mature plants can form a short yellow-brown flowering spadix at the base of the plant, but inflorescences do not have ornamental value. The roots of ZZ are rich in steroid, triterpenoid, flavonoid, and polyphenolic. The extract is also a source of antioxidants and the juice is used to treat ear-ache by Malawian people meanwhile the roots are used for treating gastric problems by Sukuma people in Tanzania. The locals also use the entire plant to treat the inflammatory condition–*Mshipa*. The leaves of the plants are also used by the shamans in the jungles of Ghana to cure stomach ailments (Rini *et al.,* 2018).The potential for *Zamioculcas* to become a popular foliage plant exists because it has naturally dark green glossy foliage, limited disease, insect pests, and it performs well in low light and dry conditions also improves air quality which helps enhance cognitive function (Lopez *et al.,* 2009). However, the ZZ plant grows

very slowly. Starting with leaflet cutting, it takes a year or more to reach a saleable size.

ZZ plants can be propagated asexually by rhizome division, leaflet cutting, and petiole cutting. To propagate from a leaflet cutting, an individual leaflet is directly inserted into rooting medium or large leaflets may be cut horizontally in half to two pieces to increase the number of propagules and propagated by sticking the basal cut end into the rooting media. The main objectives of study were to see the effect of natural rooting substances on rooting, rhizome, and shoot growth of *Zamioculcus zamiifolia* cultivars. Natural root promoting substances are cheap and safe to use as an alternative for rooting of horticultural crops. They are environment friendly and can be used as a substitute for synthetic plant growth hormones like IBA (Sherif *et al.,* 2017).

Natural rooting substances can be utilised for the propagation of hardwood and softwood cuttings of horticultural crops. They are sustainable, cost effective, and environment-friendly and improve crop growth and quality. They increase sustainability of the soil and make it more productive. Synthetic growth regulators have restrictions in some countries including European Union. In this regard, the use of natural rooting hormones is highly significant in propagation of horticultural crops (Pacholczak *et al.,* 2016). Hence there is a need of utilizing alternative hormone for the propagation of cuttings.

MATERIALS AND METHODS

The field experiment was conducted at the Horticulture Farm, SASRD, Medziphema, Nagaland. The institution is located at 25°45'43" N latitude and 93°53'04" E longitude at an elevation of 305 m above mean sea level. Medziphema falls under sub-tropical region with humid and moderate temperature having medium to high rainfall. The temperature varies from 31-7°C during winter to 34-13°C during summer and average maximum RH of 79-92% and minimum of 38-64%. The annual rainfall of experimental site was ranges from 200 cm to 300 cm. The experiment was laid out in completely randomized design (factorial) with three replications and two factors. Treatment consists of (i) Two cultivars (C) of zz plant; C_1- ZZ Super Nova and C_2- ZZ Raven, and different natural rooting substances *viz*., Control (T_0), Honey (T_1), Cinnamon powder (T_2), *Aloe vera* gel (T_3), Apple cider (T_4) and Coconut water (T_5). Healthy mother plant of ZZ cultivars, Super Nova and Black Raven were procured from a reliable commercial nursery named as 'Zapzeer Nursery' in Assam. The Super Nova ZZ plant is a cultivar that is a very well-known house plant, not only for its attractive, exotic looking foliage but for being borderline indestructible. The ZZ (Black Raven) first appeared in a South Korean nursery in 2006. Hyuk Jin Lee, a nursery owner, and ZZ plant grower noticed when one of his plants sent out a single dark-colored branch this mutation appeared naturally and when propagated, the same characteristics carried over to next generations. The Black Raven plants are slow-growing plant, which can grow up to 2-3 feet tall. ZZ leaf cuttings were taken from a healthy mother plant having matured shoot, dense leaf growth and glossy look. Healthy and matured leaves were cut with the petiole from the mother plant with the help of sharp Blade. Transparent plastic disposal cup of 60 ml was used for planting/ propagation of leaf cuttings. Coarse river bed sand, being sterile was used as rooting medium. The rooting media were moistened with water and leaf cuttings were inserted into the media in upright position, leaving most of the leaf exposed and pressed firmly to the media. After that, the planted cuttings were kept under a poly house for further growth of rhizome and root. The potting mixture was prepared by mixing top soil, sand and Farm Yard Manure in the ratio of 1:1:1, after removing all the grasses and foreign materials. Thereafter, the rooted cuttings were transplanted to the polybag from disposal cup with the potting mixture filled in a poly bag of 4 ×6 inch in size. Then the rooted leaves with rhizome were transplanted in the field after 75 days of planting in polybags. Proper care was taken during transplanting to avoid injury to the root.

The length of the root growth was calculated by the following formula and represented in percentage:

Growth rate = $(L_t - L_0)/L_t \times 100$

L_t= length at the end of time, L_0= initial root length

The survivability percentage of the leaf cutting for both the cultivars (Black Raven and Super Nova) was calculated by the following formulae and expressed in percentage.

Plant survival rate (%) = Number of remaining plant/numbers of plant originally×100

Mean data obtained during the period of investigation were statically analyzed by the analysis of variance method (Gomez and Gomez, 1984). The significance of the different source of variance were tested by error mean square, using Fisher Snedecor 'F' test of probability at 0.5% level of significance.

RESULTS AND DISCUSSION

Days to rhizome initiation

The results pertaining days to rhizome initiation of ZZ (*Zamioculcas zamiifolia)* leaf cutting are presented in (Table 1, Fig. 1A.). It was observed that the days to rhizome initiation varied significantly in both the cultivars, where C_1 (Super Nova) took minimum days (45.54) to rhizome initiation, while rhizome initiation in C_2 (Black Raven) was observed after 56.28 days of planting the leaf cuttings. The difference in days to rhizome initiation may be attributed to several factors including genetic factor as well as the interaction of the varieties with the climatic condition. The present findings are in conformity with the results of (Benedetto *et al.,* 2020) who stated that the difference among the cultivars is a varietal trait and is probably governed by genetic makeup. Further, analysis of the data showed that there was a significant variation among the natural rooting substances with respect to days to rhizome initiation as shown in (Table 1). It was found that ZZ leaf cuttings treated with T_5 (coconut water) initiates rhizome formation earlier (46.50 days), which was at par (47.97 days) with T_1 (honey). The maximum number of days (55.80) to rhizome initiation was noted in control. The present results are in agreement with the findings of Davies, (2004) who noted that coconut water contains auxins and gibberellins and stimulate the cambial activity, thereby causes formation of large xylem and phloem. Similarly, Agele *et al.* (2010) stated that coconut water promotes root formation and shoot emergence in stem cuttings of various species. The interactions between cultivars and natural rooting substances failed to evoke any significant effect on days to rhizome formation.

Days to root initiation on rhizome

The analysis of the data on days to root initiation varied significantly in the studied cultivars, represented in (Table 1, Fig. 1B.). The minimum number of days (52.09) to root initiation was recorded in C_1 (Super Nova), while C_2 (Black Raven) took the maximum days (60.47) to root initiation. The variation may be due to genetic makeup of the individual cultivars. The spongy mesophyll cell of black leaves receives lesser light than those of green leaves of *O. planiscapus* 'Nigrescens' which might have attributed to slow down the growth rate. Further critical examination of the results presented in (Table 1) showed a significant difference among the natural rooting substances. ZZ leaf cutting dipped in T_5 (coconut water) took minimum days (50.73) to root initiation followed by T_1 (honey) and T_4 (apple cider) which took 53.17 days and 51.77 days respectively to initiate root. While the maximum number of days (61.17) to root initiation was noted in T_0 (control). The present result corroborates with the finding (Hartmann and Kester, 2007) who opined that coconut water enhances the hydrolysis and translocation of carbohydrates resulting in root initiation and a greater number of roots per cuttings. Similarly, (Ibironke, 2016) also stated that bougainvillea, cuttings treated with coconut water showed significant effect on root emergence and root growth. Honey is a natural source of vitamins such as vitamin A and vitamin B_1, which might have attributed to the initiation of rooting in cuttings. The interactions between cultivars and natural rooting substances were found to cause an appreciable impact on days to root initiation. The minimum days to root initiation (46.20) was recorded in C_1T_5 while C_2T_0 took the maximum days (65.07) to root initiation.

Fig. 1:
A. Rhizome initiation **B. Root initiation** **C. Number of roots**

Table 1: Effect of natural rooting substances on days to rhizome initiation, root initiation onrhizome, emergence of shoot bud, shoot length at 45 days afteremergence and survivability of *ZZ* cultivar leaf cuttings

Treatments	Days to Rhizome initiation	Days to Root initiation on rhizome	Emergence of shoot bud (Days)	Shoot length at 45 days after emergence	Survivability (%)
C_1	45.54	52.09	182.76	2.26	100%
C_2	56.28	60.47	194.13	2.31	100%
SEm(±)	**0.23**	**0.18**	**1.10**	**0.02**	
CD	**0.67**	**0.53**	**3.20**	**0.06**	
T_0	55.80	61.17	196.00	1.54	100%
T_1	47.97	53.17	180.27	2.96	100%
T_2	53.97	59.30	192.60	1.60	100%
T_3	50.53	57.53	188.90	1.91	100%
T_4	50.70	55.77	190.70	2.63	100%
T_5	46.50	50.73	182.20	3.09	100%
SEm(±)	**0.33**	**0.26**	**1.55**	**0.03**	
CD	**0.95**	**0.75**	**4.53**	**0.08**	
Interaction (CxT)					
C_1T_0	50.13	65.07	189.87	1.55	100%
C_1T_1	42.67	57.60	174.13	2.90	100%
C_1T_2	49.27	63.53	187.27	1.68	100%
C_1T_3	45.20	62.07	183.33	1.75	100%
C_1T_4	45.00	59.27	184.67	2.70	100%
C_1T_5	41.00	55.27	177.27	3.01	100%
C_2T_0	61.47	57.27	202.13	1.54	100%
C_2T_1	53.27	48.73	186.40	3.01	100%
C_2T_2	58.67	55.07	197.93	1.51	100%
C_2T_3	55.87	53.00	194.47	2.07	100%
C_2T_4	55.50	52.27	196.73	2.55	100%
C_2T_5	52.00	46.20	187.13	3.18	100%
SEm(±)	**0.56**	**0.44**	**2.69**	**0.05**	
CD	**NS**	**1.29**	**NS**	**0.14**	

C= Cultivars, T= Natural rooting substances

Table 2: Effect of natural rooting substances on rhizome size (cm), root length, number of roots and rate of root growth of *ZZ* cultivar leaf cuttings

Treatments	Diameter of rhizome (cm)			Length of primary root (cm)		No. of root sper cutting		Rate of root growth (%)	
	90DAC	120 DAC	150 DAC	75DAP	150 DAP	75DAP	150DAP	75DAP	150DAP
C_1	0.93	2.10	2.49	1.88	3.65	4.46	8.04	71.76	86.55
C_2	0.82	1.52	1.76	1.83	2.82	4.36	7.15	70.43	82.55
SEm(±)	**0.02**	**0.07**	**0.06**	**0.10**	**0.17**	**0.18**	**0.23**	**1.50**	**0.72**
CD	**NS**	**0.19**	**0.16**	**0.29**	**0.51**	**0.52**	**0.67**	**4.39**	**2.10**
T_0	0.76	1.42	1.80	1.23	2.56	3.17	6.33	59.22	80.24
T_1	0.91	2.03	2.34	2.11	3.99	5.10	8.53	76.16	86.92
T_2	0.84	1.65	1.93	1.62	2.94	3.91	6.82	67.73	82.33
T_3	0.81	1.76	2.00	1.82	3.18	4.43	7.00	71.40	83.94
T_4	0.89	1.91	2.22	1.91	3.48	4.50	8.17	73.49	84.42
T_5	1.05	2.10	2.46	2.45	4.93	5.33	8.73	78.59	89.44
SEm(±)	**0.32**	**0.25**	**0.25**	**0.14**	**0.25**	**0.25**	**0.32**	**2.13**	**1.02**
CD	**0.94**	**0.73**	**0.72**	**0.41**	**0.72**	**0.73**	**0.94**	**6.21**	**2.97**
Interaction (CxT)									
C_1T_0	0.83	1.61	2.19	1.21	2.81	2.87	6.60	58.67	82.18
C_1T_1	0.97	2.35	2.69	2.15	4.45	5.40	9.27	76.57	87.96
C_1T_2	0.91	1.98	2.32	1.82	3.34	3.96	7.00	71.63	84.79
C_1T_3	0.87	2.09	2.33	1.88	3.59	4.47	7.00	73.92	85.91
C_1T_4	0.97	2.11	2.51	1.82	4.05	4.47	8.93	72.17	87.14
C_1T_5	1.02	2.46	2.89	2.41	5.80	5.60	9.47	77.60	91.29
C_2T_0	0.69	1.23	1.41	1.24	2.31	3.47	6.07	59.77	78.30
C_2T_1	0.85	1.70	1.98	2.07	3.54	4.80	7.80	75.75	85.87
C_2T_2	0.77	1.98	1.53	1.45	2.55	3.87	6.63	63.83	79.87
C_2T_3	0.75	1.42	1.66	1.72	2.78	4.40	7.00	68.88	81.96
C_2T_4	0.81	2.11	1.94	1.99	2.92	4.53	7.40	74.80	81.70
C_2T_5	1.07	1.73	2.04	2.49	4.05	5.07	8.00	79.57	87.60
SEm (±)	**0.06**	**0.16**	**0.14**	**0.24**	**0.42**	**0.43**	**0.56**	**3.68**	**1.76**
CD	**NS**	**NS**	**NS**	**NS**	**NS**	**NS**	**NS**	**NS**	**NS**

DAC= days after cutting, DAP= days after planting

Days to emergence of shoot bud

The data on days to emergence of shoot bud are depicted in Table 1. The result showed a significant difference between cultivar. The minimum days (182.76) to shoot bud emergence was noted in C_1 (Super Nova), while C_2 (Black Raven) took maximum days of 194.13 for shoot bud emergence. The variation in shoot bud emergence might be due to the genetic makeup of the cultivars. Apparently, the variation in vegetative characters was attributed to genetics factor whose performance will be varied over a wide range of environmental conditions (Srilatha *et al.,* 2015). Further, perusal of the data showed a significant influence on shoot bud emergence among the different natural rooting substances, perented in Tables 1. The earliest shoot bud initiation (182.20 days) was noticed in T_5 (coconut water), which was at par (180.27 days) with T_1 (honey), while the slowest shoot bud initiation (196 days) was recorded from T_0 (control) treatment. Coconut water contains urea diphenyl which has activities of cytokinins thereby increases growth and differentiation of cells affecting cell division leading to better shoot growth. The increase in rate of shoot initiation may also be due to auxin effect on the cell wall, turgor and osmotic pressure and water permeability which causes cell enlargement resulting in enhanced vegetative

growth (Taiz and Zeiger, 2006). Prathibha *et al.* (2018) reported that in *Zamioculcas zamiifolia,* enhanced shoot initiation and shoot growth was observed in cuttings treated with cytokinin and NAA and even multiple shoot formation was observed. The interaction effect between cultivars and natural rooting substances on shoot bud emergence was found to be non-significant.

Shoot length at 45 days after emergence

The data regarding shoot length at 45 days after emergence are shown in (Table 1). It is evident from the result that the cultivar had significant impact on the shoot length. It was observed that C_1 (Super Nova) recorded the maximum shoot length (2.31 cm), while C_2 (Black Raven) recorded the minimum shoot length (2.26 cm) at 45 days after emergence. Differences in growth may be due to nature of cultivars, changes in any phenotypic character like color, shape or size of flower and chlorophyll variegation in leaves which can impact the character of plant (Datta *et al.,* 2006). Further, examination the data showed that different natural rooting substances evoked a significant difference on shoot length at 45 days of emergence, presented in (Table 1). The maximum shoot length (3.09 cm) was recorded in cuttings treated with T_5 (coconut water), which was at par (2.96 cm) with T_1 (honey), while the least shoot length (1.54 cm) was observed in T_0 (control) at 45 days after emergence. The best result was observed in cuttings treated with coconut water, which may be due to increased number of roots which probably drive more nutrients and water from the soil and enhanced photosynthesis which ultimately resulted in more growth (Singh and Singh, 2005). Moreover, coconut water contains urea diphenyl which has activities of cytokinins thereby increases growth and differentiation of cells affecting cell division leading to longer shoot. The increase in shoot length may also be due to auxin effect on the cell wall, turgor and osmotic pressure and water permeability which causes cell enlargement resulting in enhanced vegetative growth (Taiz and Zeiger, 2006). Comparable finding was observed by (Ogunsiji *et al.,* 2022) in *Ceiba petandra* L., cuttings. The interactions between the cultivars and different natural rooting substances were found to cause an appreciable impact on the shoot length after 45 days of emergence. The highest shoot length (3.18 cm) was observed in C_1T_5 and least shoot length (1.51cm) was recorded in C_2T_2.

Survivability percentage

All the cuttings displayed 100 % survivability, represented in (Table 1). The growth of adventitious roots and rhizome of ZZ plant leaflet cutting during propagation is largely determine by the carbohydrates reserve in the source and the photosynthesic capacity of the cuttings after root emergence.

Diameter of rhizome

Thorough scanning of the data pertaining to the effect of cultivars on the diameter of rhizome at 90, 120 and 150 days after planting presented in Table 2, it was apparent that the cultivars did not have any significant response on growth of rhizome at 90 days after planting. However, a significant effect was noted at 120 and 150 days after cutting. Out of the two cultivar, C_1 (Super Nova) recorded the maximum diameter (2.10 and 2.49 cm) of rhizome while C_2 (Black Raven) exhibited the least (1.52 and 1.76 cm) diameter at 120 and 150 DAP, respectively. Benedetto *et al.* (2020) opined that significant difference in growth rate among clones is known to occur and the difference tends to increase during plant development. Further analysis of data showed that the natural rooting substance had significant impact on the rhizome growth, presented in Table 2. However, it was observed that the cutting dipped in T_5 (coconut water) induced larger rhizome diameter size (1.05, 2.10 and 2.46 cm) at 90, 120 and 150 DAP, which was at par with T_1 (honey), which recorded 0.91, 2.03 and 2.34 cm at 90, 120 and 150 DAP, respectively. The maximum rhizome size on all dates of observation was observed with coconut water treated leaf cutting which might be due to maximum number of roots which probably drive more nutrients and water from the medium which ultimately resulted in more growth (Singh and Singh, 2005) of rhizome. Moreover, coconut water contains urea diphenyl which has activities of cytokinins thereby increases growth and differentiation of cells affecting cell division (Taiz and Zeiger, 2006).The

interaction effect between cultivars and natural rooting substances on the rhizome size was found to be non-significant.

Length of primary root

It was evident from the data given in Table 2 that there was a significant variation in the length of primary root between the cultivars. The root length was maximum (1.88 and 3.65 cm) in C_1 (Super Nova), while C_2 (Black Raven) recorded the minimum root length (1.83 and 2.82 cm) at 75 and 150 days after planting respectively. Cultivars difference may be due to nature of cultivar, changes in any phenotypic character like color, shape or size of flower and chlorophyll variegation in leaves can impact the character of plant (Datta *et al.,* 2006). Further examination of the data revealed that the maximum root length was observed in leaf cuttings treated with T_5 (coconut water) with (2.45 and 4.93 cm), which was at par (2.11 and 3.99 cm) with T_1 (honey) at 75 and 150 DAP respectively whereas the minimum primary root length (1.23 and 2.56 cm) was recorded in T_0 (control) at 75 and 150 DAP, respectively, presented in Table 2. The increase length of root treated with coconut water may be due to the presence of auxins and cytokinins in coconut water which stimulates the formation of masses of undifferentiated cells (callus) and also increased the carbohydrate metabolism and metabolite translocation (Karunarathna and Kumuthini, 2016). The present findings are in line with the result of Richard (2015) who reported that in *Rhizospora stylosa,* highest root length was observed in samples treated with coconut water. The interactions between cultivars and natural rooting substances failed to reach the level of significant effect on the length of primary root at 75 and 150 days after planting, respectively.

Number of roots per cutting

The data pertaining to number of roots per leaf cutting at 75 and 150 DAP as influenced by cultivars are presented in Table 2, Fig. 1 C. The number of roots per cutting was found to be maximum (4.46 and 8.04) in C_1 (Super Nova), while the least number of roots (4.36 and 7.15) was observed in C_2 (Black Raven) at 75 and 150 days after planting, respectively. The variation in number of roots might be due to the genetic makeup of the cultivars. Apparently, the variation in growth characters was attributed to genetic factor whose performance will be varied over a wide range of environmental conditions (Srilatha *et al.,* 2015). Further, scanning of the data showed a significant influence on number of roots among the natural rooting substances. The maximum number of roots (5.33 and 8.73) per cuttings was recorded in T_5 (coconut water), which was found to be at par (5.10 and 8.53) with T_1 (honey) at 75 and 150 DAP respectively whereas the minimum number of roots (3.17 and 6.33) per cuttings was recorded in T_0 (control) at 75 and 150 DAP respectively, depicted in (Table 2). Coconut water contains auxins and gibberellins which stimulates the cambial activity and leads to formation of larger xylem and phloem which influence formation and growth of roots, (Davies, 2004). Comparable results were observed by Bamigboye *et al.* (2016) who reported that in *Dioscoreophyllum cumminssi* and Serendipity Berry plant sample treated with coconut water showed highest number of roots. Abo *et a.,* (2018) reported that besides growth regulators and hormones, another main criterion affecting rooting phenomenon is sucrose which is in high concentration in honey. The interactions between cultivars and natural rooting substances did not show any significant effect with respect to number of roots per cutting on both days of observation.

Rate of root growth

It was evident from the data Table 2, that the cultivars had a profound effect on the rate of root growth. The rate of root growth was maximum (71.76% and 86.55%) in C_1 (Super Nova) while C_2 (Black Raven) recorded minimum root growth (70.43% and 82.55%) at 75, 150 days after planting (DAP) respectively. Cultivars difference may be due to nature of cultivar, changes in any phenotypic character like color, shape or size of flower and chlorophyll variegation in leaves can impact the character of plant (Datta *et al.,* 2006). Further examination of the data showed a significant effect on the rate of root growth among the different rooting substances. The highest rate of root growth (78.59% and 89.44%) was observed in cuttings treated with T_5 (coconut water), followed by T_1

(honey), exhibiting a root growth percentage of 76.16 and 86.92% respectively at 75 and 150 DAP. The minimum rate of root growth (59.22% and 80.24%) was recorded in T_0 (control) treatment at 75 and 150 DAP, presented in (Table 2). Coconut water contains auxins and gibberellins which together stimulates the cambial activity, causing formation of larger xylem and phloem, (Davies, 2004). Comparable results were reported by Richard (2015) in *Rhizospora stylosa,* where the highest root length was observed in samples treated with coconut. Karunarathna and Kumuthini (2016) also reported that cuttings of Ixora treated with coconut water recorded highest root length. The interaction effect between cultivars and different natural rooting substances on the rate of root growth rhizome size was found to be non-significant on both days of observation.

CONCLUSION

Out of both cultivars, it can be inferred from the present research work that the leaf cutting of ZZ cv. Super Nova grows much faster than ZZ cv. Black Raven for all parameters from rhizome initiation, root initiation, root length, rhizome size, shoot emergence, shoot length, root growth and root number even when given with same soil and climatic condition. In case of natural rooting substances used in the experiment, coconut water was considered to be the best in respect of rhizome initiation and root and shoot growth.

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Effect of Salicylic acid, 2,4-dichlorophenoxyacetic acid and gibberellic acid on growth, flowering, and fruit quality of Cape gooseberry (*Physalis peruviana* L.)

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ABSTRACT

Cape gooseberry (Physalis peruviana L.) has received significant attention recently due to its high value and medicinal qualities. The experiment was carried out in Randomized Block Design (RBD), with 10 treatments, replicated 03 times. The goal of the current study was to examine the effects of three plant growth regulators on the growth, blooming, and fruit quality of cape gooseberries, namely salicylic acid (SA), 2,4-D, and gibberellic acid (GA_3) at various doses. Salicylic acid (20, 40, and 60 ppm), 2,4-D (4, 8 and 12 ppm), GA_3 (20, 40, and 60 ppm), and a control were used as treatments. The best treatment combination in terms of plant height (90 cm), number of leaves (106.11), leaf area (36.11 cm^2), and number of branches (12.11) was T9-GA_3 @ 60 ppm. The higher concentration of GA_3 @ 60 ppm led to earlier flowering, earlier fruiting, and more flowers overall. GA_3 @ 60 ppm significantly improved the total soluble solid (12.93 0B), ascorbic acid content (53.30 mg/100g), and TSS:acid ratio (15.34). Acidity decreased with increasing concentration of GA_3.

***Keywords*:** Cape gooseberry, flowering, fruit quality**,** gibberellic acid, salicylic acid

INTRODUCTION

The Cape gooseberry *Physalis peruviana* L.), belongs to family Solanaceae, is one of the world's lesser-known and underutilized fruit crops. Many distinct names, including Poha, Tepari, Golden Berry, and Husk Berry, are used to refer to Cape gooseberries around the world. It belongs to the *Physalis* genus, which is part of the Solanaceae family and contains 80 species. Of these, only three, *P. peruviana* L., *P. pubescens* L., and *P. ixocarpa* Brot., have been recognized as species that produce edible fruit. When maturing, the Cape gooseberry's berries are a yellow-orange color, 1 to 3.5 cm in diameter, very juicy, aromatic, and have a distinct bitter-sweet flavor. The bigger, papery crescent epicalyx encloses the fruits. Its fruit contains a variety of bioflavonoids, including vitamin P, which reduce inflammation and function as natural blood thinners. It effectively lowers cholesterol levels and has anti-ulcer properties (Mayorga *et al*., 2001). The fruit's pulp contains 1.6 mg/100g carotene, 0.1-0.18 mg/100g thiamine, 0.03-0.18 mg/100g riboflavin, 0.8-1.7 mg/100g niacin, 20-43 mg/100g vitamin C, 210-467mg/100g K, 7-19 mg/100g Mg, 8-28 mg/100g Ca, 27-55.3 mg/100g P, 0.3-1.2 mg/100 (Puente *et al*., 2011; Ramadan and Morsel, 2009). The plants are naturally fast-growing and are easily propagated through seeds and cuttings. Being a warm season crop, it needs a lengthy growing season to provide a profit. Farmers continue to occasionally cultivate cape gooseberries on a modest scale, producing low-quality fruits that prevent them from realizing the full value of the crop. Plant growth regulators are chemicals that, when used sparingly, alter plant development, typically by activating a portion of the plant's inherent growth regulation system. In this regard, plant growth regulators (PGR) may be crucial to the crop's ability to produce high-quality fruits. Yet, there is a dearth of literature on PGRs' impact on fruit quality. Plant growth regulators (PGR) are widely used in horticulture crops to promote output

by boosting flowering time, fruit set, and fruit size. By interacting with important metabolic processes including nucleic acid metabolism and protein synthesis, plant growth regulators like promoters, inhibitors, and retardants play a crucial role in directing internal mechanisms of plant growth. It aids in changing the canopy structure and other yield factors. A useful approach to boost crop yield may be the application of plant growth regulators (PGR). The significance of PGRs in boosting crop productivity has just come to light on a global scale.SA plays a crucial part in the growth and development of the plant by improving the plant's response to tolerance and resistance to numerous plant-affecting diseases. It increases the representation of CO_2 gas, gas exchange, protein synthesis, ion absorption, and nutrition transfer. It also stimulates blooming.2, 4- D enhances fruit set, fruit number, TSS, number of secondary roots, and yield; however, it significantly decreases plant height, internode length, days to flowering, acidity, and number of seeds per fruit. Gibberellic acid is essential for the growth of the morphological traits that give plants and their fruits their distinctive appearance. GA_3 has the ability to speed up some sub apical meristem cell processes like cell division and mitosis. Internodes length, leaf number, branch count, fruit and berry size all increase as a result.With the aim of determining the appropriate concentrations of Salicylic acid, 2,4-D, and GA_3 plant growth regulators on the vegetative development, flowering, and quality of Cape gooseberries, the current study was conducted.

MATERIALS AND METHODS

The experiment was conducted during October 2020 to March 2021 at Horticulture Research Farm, Department of Horticulture, Naini Agricultural Institute, Sam Higginbottom University of Agriculture, Technology and Sciences, Prayagraj. The region of experimental site (Naini, Prayagraj) has a sub-tropical climate, situated in the South-East part of U.P. at 25.45^0 North latitude and 81.84^0 East longitudes and an elevation of 98 meters above sea mean level (MSL). In hot summer temperatures rise up to 46 ^{0}C to 48 ^{0}C and minimum 4 ^{0}C – 5 ^{0}C in winter and average rainfall is 1013.4 mm. With three replications and ten treatments, the experiment was set up using a randomized block design. As a foliar spray, three plant growth regulators in each of three concentrations were used. The treatment consisted T_0- Control, T_1- SA @ 20 ppm, T_2 - SA @ 40 ppm , T_3 - SA @ 60 ppm , T_4 - 2-4,D @ 4 ppm , T_5 - 2-4,D @ 8 ppm, T_6 - 2-4,D @ 12 ppm, T_7 - GA_3 @ 20 ppm, T_8 - GA_3 @ 40 ppm, T_9 - GA_3 @ 60 ppm. Application of PGRs was given at 20 days after transplanting, At flowering, at fruit set and maturity stage. Data on numerous parameters from chosen plants were collected at various phases of plant growth and statistically examined using the Analysis of Variance (ANNOVA) method.

RESULTS AND DISCUSSION

Growth parameters

The treatment T_3 - SA @ 60 ppm, showed maximum plant height which is 26.22 cm followed by T_9 - GA_3 @ 60 ppm after 25 days after transplanting. T_9 - GA_3 @ 60 ppm registered the maximum height in the plant after 50 days and 75 days after transplanting. Control T_0 had the shortest plant height. The outcomes demonstrate a beneficial impact on plant height from the usage of GA_3 at higher doses. Because GA_3 encourages cell division and cell elongation, which results in cell enlargement, it is conceivable that this is the cause of the increased plant height. Additionally, Wanyama *et al.* (2006) and Udden *et al.* (2009) reported a similar conclusion. Final reading of Table 1 shows that treatment T_9 (GA_3 @ 60 ppm), which has significantly more leaves (106.11), leaf area (79.89 cm^2), and branches (12.11) than control. A higher quantity of GA_3 may have caused an increase in leaf counts, which is one explanation for the phenomenon. There are more leaves on each plant as a result of gibberellin's stimulation of auxin action. In comparison to other treatments, treatment T_9 (GA_3 @ 60 ppm) was shown to be the best and recorded the largest leaf area and branches, followed by treatment T8 (GA_3 @ 40 ppm). In term of Salicylic acid treatment T_3 - SA @ 60 ppm showed increased number of leaves (10.11), leaf area (70.56 cm^2) and number of branches (9.67) as compare to its lower concentration and have no significance difference to other salicylic acid treatment but at par with control. GA_3 encourages cell expansion and boosts photosynthetic rate, increasing leaf area. Similar findings were provided by Kaur and Kaur (2016). Gibberellic acid's anti-

Table 1: Effect of Salicylic acid, 2,4- dichlorophenoxyacetic acid and Gibberellic acid on growth of Cape gooseberry (*Physalis peruviana* L.).

Treatment symbols	Treatments	Plant height (cm)			No. of leaves			Leaf area (cm^2)			No. of branches	
		25 days	50 days	75 days	25 days	50 days	75 days	25 days	50 days	75 days	50 days	75 days
T_0	Control (Water spray)	17.78	32.22	57.67	7.32	31.89	66.22	13.67	26.67	60.56	2.67	5.44
T_1	SA @ 20 ppm	22.44	38.55	72.67	9.44	42.11	85.11	14.78	32.56	66.22	4.22	8.11
T_2	SA @ 40 ppm	19.55	42.67	76.44	8.56	48.66	88.33	15.89	31.78	65.44	4.55	8.89
T_3	SA @ 60 ppm	26.22	45.33	86.78	10.11	47.78	95.78	19.89	34.45	70.56	4.44	9.67
T_4	2,4-D @ 4 ppm	19.55	37.11	65.55	8.33	39.78	78.44	15.88	29.56	61.89	3.89	7.78
T_5	2,4-D @ 8 ppm	18.66	36.11	64.22	8.44	41.99	81	14	29.89	62.33	3.55	7.33
T_6	2,4-D @ 12 ppm	16.11	35.67	62.78	7.89	36.89	75.22	14.22	27.89	61.33	3.33	7
T_7	GA_3 @ 20 ppm	23.78	39	82.11	9.44	46.67	98.56	18.44	34.89	72.89	4.56	10.44
T_8	GA_3 @ 40 ppm	20.33	40.89	79.89	9.55	45.33	91.89	17.22	35.78	76.44	**5.11**	11
T_9	GA_3 @ 60 ppm	24.67	49.44	90	9.22	53.55	106.11	19.67	36.11	79.89	4.78	12.11
	CD at 5%	6.26	7.14	6.46	2.11	10.56	13.47	4.67	7.81	6.94	1.34	3.32
	F – test	NS	S	S	NS	S	S	NS	NS	S	S	S

Table 2: Effect of Salicylic acid, 2,4- dichlorophenoxyacetic acid and Gibberellic acid on flowering, fruiting, yield and quality of Cape gooseberry.

Treatment symbols	Treatments	Days taken to first flowering	Days taken to first fruiting	No. of flower per plant	Fruits/ plant	Yield (g/plant)	TSS (^{0}B)	Acidity (%)	TSS : Acid ratio	Ascorbic acid (mg/100g)
T_0	Control (Water spray)	56.11	67.89	93.78	61.03	384.67	7.7	1.12	6.85	35.53
T_1	SA @ 20 ppm	50.33	59	106.11	84.30	699.98	10.47	0.97	10.76	37
T_2	SA @ 40 ppm	51.67	60.78	108.22	87.63	768.45	10.03	0.95	10.61	44.53
T_3	SA @ 60 ppm	49.56	57.89	109.89	89.30	848.53	11.7	0.93	12.6	45.97
T_4	2,4-D @ 4 ppm	53	63	101.11	73.03	545.28	8.6	1.01	8.51	38.7
T_5	2,4-D @ 8 ppm	53.89	64.33	98	68.60	534.96	8.97	1.05	8.51	40.53
T_6	2,4-D @ 12 ppm	54.55	65.11	96	66.94	471.26	9.43	1.08	8.74	42.67
T_7	GA_3 @ 20 ppm	48	56.67	112.44	95.02	871.41	10.93	0.89	12.22	49.05
T_8	GA_3 @ 40 ppm	48.89	55.56	115.33	97.38	970.75	12.26	0.87	14.11	50.89
T_9	GA_3 @ 60 ppm	47.33	54.89	117.11	102.40	1096.18	12.93	0.84	15.34	53.30
	CD at 5%	1.31	2.31	1.88	2.24	40.19	0.79	0.06	1.20	1.23
	F – test	S	S	S	S	S	S	S	S	S

mitotic action, which has an inhibitory influence on apical growth, is what causes the increase in branch number. Gocher *et al.* (2017), Tiwari and Singh (2014), and El-Tohamy *et al*. (2012) reported findings that were similar to present findings.

Flowering and yield parameter

From the Table 2 it is clear that T_9 - GA_3 @ 60ppm (47.33 days) and T_7 - GA_3 @ 20ppm (48 days) produced the best results in terms of early blooming when compared to other treatments. With the untreated plant T_0, blooming took longer than usual (56.11 days). T_9 (GA_3 @ 60 ppm) recorded the shortest number of days needed to reach first fruiting (54.89 days), followed by T_8 (GA_3 @ 60 ppm), which is (55.56 days). T_0 (control) took the longest number of days to first fruiting (67.89). Early blossoming was evident in all GA_3 treatments, and this was noteworthy. A higher GA_3 concentration causes early flowering because it produces more leaves and encourages vegetative growth. With the higher concentration of GA_3, the first fruiting start time was greatly accelerated. The cape gooseberry's early flowering results in early fruit setting. This outcome is in agreement with Verma *et al. (*2014). Treatment T_3 - SA @ 60 ppm record minimum days to flowering (49.56 days) followed by T_1 - SA @ 20 ppm T_2 - SA @ 40 ppm and (50.33 and 51.67 days respectively) and also results early fruiting treated with salicylic acid as compared to non treated plant. About 2,4-D, blooming time rises as concentration increases. Compared to its previous treatments (2,4-D @ 8 and 12 ppm), 2,4-D @ 4 ppm concentration significantly shortens the flowering period, resulting in early fruiting. This finding is supported by the tomato experiment conducted by Tiwari and Singh (2014) and Pundhir and Yadav (2001). The number of flowers was revealed to be 112.44, 115.33, and 117.11 at GA_3 concentrations of 20, 40, and 60 ppm, respectively. T_9 (GA_3 @ 60 ppm) produced better results, whereas the control had the fewest flowers. Plants produced more flower primordia when gibberelic acid was present. The GA_3 treated plants were able to accumulate and store enough carbohydrates, which resulted in increased flower production. Both Udden *et al.* (2009) and Verma *et al.* (2014) validated these findings. In salicylic acid treatment, highest no of flower per plant (109 flower) was counted in T_3 (SA @ 60 ppm) followed by T_2 (SA @ 40 ppm) and T_1 (SA @ 20 ppm) which is 108.22 and 106.11 flower per plant respectively. All salicylic acid treatment give superior result over all 2,4-D treatments and control. The greatest number of flowers were evident at the lowest 2,4-D concentration of 4 ppm compared to its higher values. When 2,4-D is present in higher concentrations, it has a herbicidal effect on plants and causes flower bud abscission, which results in flower drop. Tiwari and Singh (2014) and Pundhir and Yadav (2001) also came to the same conclusion.The results showed that T_9 (GA_3 @ 60 ppm), followed by T_8 (GA_3 @ 40 ppm) and T_7 (GA_3 @ 20 ppm), had recorded the highest number of fruits per plant (102.4) and fruit output (1096.18 g/plant) when compared to other treatments. The control group, however, also recorded minimum yield and fruit production. T4 (2,4-D @ 4 ppm) recorded 73.03 fruits, although 2,4-D concentrations above this result in less fruits because of their herbicidal effects, which produce bud abscission. When the cape gooseberry has an adequate supply of carbohydrates and an optimal concentration of growth regulators, fruiting is sustained. As food reserves develop, the fruiting process becomes more active and produces more fruit. This conclusion was very similar to that of Kaur *et al.* (2013). Higher concentration of salicylic acid (SA @ 60 ppm) record 848.53 g/plant which is at par with its lower concentration and showed significant difference over control. 646.99 g/plant fruit yield recorded under T_4(2,4-D @ 4 ppm) which was maximum over its higher concentration. Yield reduced with increasing the concentration of 2,4-D. The similar findings were reported by Kavyashree *et al.*(2018) in sapota, Singh *et al*. (2018), Kaur *et al.,* (2013) and EL-Tohamy *et al.* (2012) on cape gooseberry.

Quality parameter

The results show that T_9 (GA_3 @ 60 ppm) significantly reported the highest total soluble solids value (12.93 ^{0}B), followed by T_8 (12.26 ^{0}B), as opposed to 7.7 ^{0}B under control. Fruit TSS values increased in proportion to GA_3 content. Higher concentration of salicylic acid (SA @ 60 ppm) revealed increase T.S.S. over lower concentration

of GA_3, 2,4-D (4, 8 and 12 ppm) and control. Another explanation for the rise in TSS is the rapid metabolic conversion of starch and pectin into soluble compounds and the translocation of sugar from the leaves to the washbasin (fruits). El-Thohamy *et al.* (2012) and Kaur *et al.* (2013) also opined that the TSS of cape gooseberry fruit rises with the administration of gibberellic acid, The results showed that T_9 considerably reported a lower acidity percentage (0.84%), followed by T_8 (0.87%), whereas T_0 (the control) recorded an acidity percentage (1.12%). Every treatment has a considerable impact on the acidity % compared to the control. Salicylic acid have lower acidity percentage than 2,4-D treatments and control. Increase in T.S.S. is result of decreasing in percentage of acidity. Salicylic acid @ 60 ppm showed 0.93 % acidity over control. The best treatment was discovered to be T_9 (GA_3 @ 60 ppm), followed by T_8 (GA_3 @ 40 ppm), and T_7 (GA_3 @ 20 ppm).That might be because the fruit TSS value increases when GA_3 is applied, and the cape gooseberry's acidity percentage drops (Tohamy *et al.,* 2012 Kaur *et al.,* 2013 and Gelmesa *et al*., 2010).T_9 significantly had the highest TSS: Acid ratio (15.34), followed by T_8 (14.11). T0 (the control) reported the lowest value (6.85). When gibberellic acid is used, total soluble solids rise and acidity falls as concentration rises. Best salicylic acid treatment in respect to T.S.S: Acid ratio was SA @ 60 ppm (12.6) and lowest was SA @ 40 ppm (10.61) and showed significant difference with each salicylic acid treatments. Increased in TSS caused by higher GA_3 content, and higher TSS: Acid ratio in fruits is caused by lower acidity values. Singh and Lal (2001) supported the aforementioned conclusion. The maximum ascorbic acid content observed significantly under T_9 (53.3 mg) followed by T_8 (50.89 mg). T_9 - GA_3 @ 60 ppm (53.3 mg) was found to be best treatment over control. SA @ 60 ppm showed best result for ascorbic acid content (45.97 mg/100g) over control. 2,4-D 8 ppm and @ 12 ppm showed best result over SA @ 20 ppm. In accordance to the result lower concentration of 2,4-D showed superior result in flowering and fruiting while higher concentration of 2,4-D found suitable for quality content in cape gooseberry. Ascorbic acid content in comparison to other PGRs was significantly impacted by GA_3concentration. On the other hand, Cape gooseberry plants that had not been treated were given a minimum value. Due to enhanced ascorbic acid synthesis-related enzyme activity and a reduced rate of oxygen oxidation during respiration, GA_3 raises ascorbic acid concentration. By rising gibberellic acid concentration, ascorbic acid concentration rose. Kaur *et al.* (2013) also reported similar findings.

CONCLUSION

According to the results of the experiment, treatment T_9 (GA_3 @ 60 ppm) was determined to be the best treatment in terms of vegetative growth, including plant height, number of leaves, leaf area, and number of branches. TreatmentT_9 (GA_3 @ 60ppm) exhibited early blooming and fruiting, number of fruit per plant and yield. While the application of GA_3 at a concentration of 60 ppm increased TSS, the TSS: acid ratio, and the amount of ascorbic acid, it lowered the acidity percentage. When compared to higher concentration of 2,4-D, lowest concentration of 2,4-D produced effective results, and the minimum values for all the qualities were disclosed by the control. Under the agro-climatic conditions of Prayagraj, the treatment T_9 (GA_3 @ 60 ppm) demonstrated the best results and was determined to be the optimum course of action.

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Morphological characterization and principal component analysis of cowa (*Garcinia cowa* Roxb.) germplasm

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ABSTRACT

Garciania cowa Roxb. is a neglected underutilized minor fruit in Bangladesh. There are several types of Cowa found in different locations of the country. Therefore, it is necessary to investigate the variations among different types of Cowa. To do so, five Cowa accessions were collected from Barishal and Patualkhali districts of Bangladesh to analyze the morphological variations among the germplasm and also conducted the principal component analysis (PCA) to determine the ideal amount of morphological characteristics that account for a large portion of variability. Morphological traits viz., leaf shape, base, apex, length, width, petiole length, diameter, flower characters, weight of fruit, length of fruit, diameter of fruit, seed number per fruit, length of seed, diameter and weight of seed were recorded. Wide variations were observed in leaf, fruits and seed characters of different Cowa germplasm. The longest leaves (15.70 cm), maximum fruit weight (30.56 g), highest fruit length (4.43 cm), diameter (3.90 cm) and highest seed weight (7.42 g) were found in ACS GC4. The number of seeds per fruit was minimum in cased of ACS GC3. PCA for 17 morphological characters of Cowa elucidated that four principle components with eigen values more than one accounted for 100 per cent of the total variability and the maximum variation was exhibited by the leaf length which accounted for 91.146%. From genotypic scatter plot, it was noticed that ACS GC3 has the highest variability as compared to other four germplasm. Based on the PCA and genotypic scatter plot, germplasm ACS GC3 can be a potential material for future varietal improvement program.

***Keywords*:** Cowa germplasm, morphological traits, principal component analysis

INTRODUCTION

Cowa (*Garcinia cowa* Roxb.) is a tropical and subtropical evergreen wild edible fruit that is a member of the Clusiaceae family (or Guttiferae). More than 250 species of evergreen, lactiferous, dioecious trees and shrubs are included in the genus *Garcinia*, which is found in moist, lowland tropical forests (Sweeney, 2008). The tree of Cowa can be 5-10 meters tall with several branches and green leaves. In the context of Bangladesh, this fruit is frequently referred to as Kawphal or Kao-gola, among other local names. The fruit in question, sometimes referred to as a "cowboy fruit," is considered to be of low significance or underutilized within the agricultural context of Bangladesh (Ashrafuzzaman *et al*., 2021).

The utilization of various components of Cowa, including its roots, leaves, and dried fruits, as traditional remedies is prevalent in several regions of India. The Cowa fruit possesses a distinct acidic taste and exhibits astringent properties that have been traditionally employed in the management of dysentery. Cowa fruit is recognized as a significant and valuable reservoir of bioactive chemicals, which have been traditionally employed for the therapeutic management of wounds and ulcers. It has been found that cowa fruit possesses various additional bioactive components with anti-inflammatory and antioxidant properties (Panthong *et al.,* 2009).

According to the report of Tripathi (2021), the fruit of *Garcinia cowa* possesses significant medicinal properties. The researchers successfully

identified and described a collection of compounds, including an uncommon class of polyprenylated acylphloroglucinol derivatives, as well as three further novel garcicowins and nine previously identified analogues. These compounds were extracted from the twigs of *Garcinia cowa*. They tested the cytotoxicity of the derived compound and their toxicity toward the cancer cellswere demonstrated selectively. Cow fruit is a vital source of minerals particularly Na and Fe and it contains 29.41mg/100g DW Na and 4.56 mg/100g DW Fe (Hossain *et al.,* 2021).

The plant species Cowa is found in its native habitat along the roadside and inside the forests of various districts in Bangladesh, including Chottogram Hill Tracts, Cox's Bazar, Barisal, Sylhet, Patuakhali, Bagerhat and Moulavibazar. Cowa germplasm's morphological traits somewhat different to some extent. Therefore, this study was conducted to collect different Cowa germplasm from two locations, conserved them at the BAU-GPC (Bangladesh Agricultural University Germplasm Center) and investigate the morphological traits of leaves, flowers, fruits and seeds. Principal Component Analysis of all studied traits were accomplished to find out a germplasm with higher variability.

MATERIALS AND METHODS

The morphological characters of *Garcinia cowa* Roxb. were recorded from mature and bearing trees. A total of five accessions of Cowa were collected from three locations of Bangladesh namely BAU-GPC, Mymensingh, Barishal and Patuakhali district. Leaves, flowers, fruits were collected from the experimental site to the Department of Horticulture, Bangladesh Agricultural University for investigation. Length, diameter of leaves, fruits, seeds were measured by using measuring tape and slide calipers, respectively. Weight of the sample was recorded by digital balance. Data were statistically analyzed using Statistix 10.0. The Duncan Multiple Range Test (DMRT) was employed to determine the statistically significant differences between two mean values at significance levels of 5% and 1%. The use of Principal Component Analysis (PCA) was carried out using Ward's approach.

RESULTS AND DISCUSSION

Morphological traits of leaves

Leaf traits of different germplasm slightly varied among each other. Leaf shape of two germplasm were elliptically lanceolate, rest two germplasm were lanceolate and one germplasm was acuminate. Leaf apex was obtuse in ACS GC1 while all other germplasm showed acuminate leaf apex. The base of leaves was cuneate, rounded and acute (Table 1, Plate 1 and 2).

Significant variations were observed in the measurements of leaf length, leaf breadth, petiole length, petiole diameter, and leaf area among different germplasm samples of Cowa. The leaf length measurements revealed that ACS GC4 exhibited the greatest value of 15.70 cm, followed by ACS GC2 with a length of 14.80 cm, ACS GC3 with a length of 14.43 cm, and ACS GC5 with the lowest length of 12.53 cm, as presented in Table 2. The maximum leaf width observed in ACS GC1 was 4.83 cm and the minimum width recorded in ACS GC3 (4.10cm). The petiole length of leaves varied from 0.87 cm to 1.07 cm while petiole diameter varied from 0.18 cm to 0.20 cm (Table 2). In case of leaf area, a wide variation was observed among the Cowa germplasm and the range was 36.74cm^2 to 56.66cm^2 (Table 2).

Morphological traits of flowers

The flowers exhibited dioecy, with a tiny size and a yellow coloration. Notably, the male flowers were comparatively smaller in size when compared to the female flowers. Additionally, the flowers were found to be either axillary or terminal in their arrangement. The male flowers exhibited yellow petals, with a base characterized by four bracts. Additionally, the flowers possessed 40-50 anther filaments that formed a cohesive mass. Female flowers were light-yellow color, usually solitary, axillary, larger than male, flower buds were round in shape.The length of buds exhibited variation between the different accessions of Cowa. The maximum bud length was seen in ACS GC1 and ACS GC4, measuring 1.23 cm, while the minimum length was recorded in ACS GC3 and ACS GC5, measuring 1.03 cm (Table 3).The highest sepal length was observed in ACS GC4 (0.63 cm), while the lowest sepal length was observed in ACS GC5

Table 1: The shape, apex, and base of cowa leaf according to the germplasm

Germplasm	Leaf shape	Leaf apex	Leaf base
ACS GC1	Elliptically lanceolate	Obtuse	Cuneate
ACS GC2	Elliptically lanceolate	Acuminate	Rounded
ACS GC3	Lanceolate	Acuminate	Cuneate
ACS GC4	Lanceolate	Acuminate	Cuneate
ACS GC5	Acuminate	Acuminate	Acute

Table 2: The measurements of length and breadth of leaf as well as petiole and leaf area for the Cowa germplasm.

Germplasm	Leaf length (cm)	Leaf diameter (cm)	Petiole length (cm)	Petiole diameter (cm)	Leaf area(cm^2)
ACS GC1	13.87 c	4.83 a	0.87 c	0.19 b	50.07c
ACS GC2	14.80 b	4.70 a	1.03 ab	0.19 b	52.51b
ACS GC3	14.43 b	4.10 d	0.93 bc	0.18 c	47.16d
ACS GC4	15.70 a	4.53 b	1.07 a	0.20 a	56.66a
ACS GC5	12.53 d	4.27 c	0.93 bc	0.18 c	36.74e
LSD (0.05)	0.46	0.16	0.11	0.09	0.99
Level of Sig.	**	**	**	**	**
CV(%)	1.71	1.84	5.31	2.77	6.12

** indicates significant at 1% levels of probability

Table 3:The flower features of Cowa germplasm.

Germplasm	Bud length (cm)	Sepal length (cm)	Sepal width (cm)	Petal length (cm)	Petal width (cm)
ACS GC1	1.23 a	0.54 b	0.24 b	1.17 a	0.60 b
ACS GC2	1.07 b	0.60 a	0.24 b	1.03 b	0.70 a
ACS GC3	1.03 b	0.42 c	0.30 a	1.23 a	0.62 b
ACS GC4	1.23 a	0.63 a	0.31 a	1.27 a	0.73 a
ACS GC5	1.03 b	0.42 c	0.22 b	0.87 c	0.52 c
LSD (0.05)	0.11	0.05	0.03	0.11	0.05
Level of Sig.	*	**	**	**	**
CV(%)	5.57	5.31	6.14	3.78	6.14

*&** indicates significant at 5% and 1% levels of probability

Table 4: Fruit and seed traits of Cowa germplasm

Germplasm	Fruit fresh weight (g)	Fruit length (cm)	Fruit width (cm)	No. of seeds/ fruit	Seed weight (gm)	Seed length (cm)	Seed diameter (cm)
ACS GC1	24.09c	3.54c	3.75b	6.00b	6.12c	2.16c	1.14bc
ACS GC2	24.82c	3.52c	3.68b	7.33a	7.11b	2.06c	1.08c
ACS GC3	28.66b	4.18b	3.81ab	4.00d	6.96b	2.55a	1.40a
ACS GC4	31.29a	4.43a	3.90a	7.33a	7.42a	2.38b	1.20b
ACS GC5	18.26d	3.11d	3.37c	5.00c	5.21d	1.76d	0.79d
LSD 0.05	1.16	0.18	0.15	0.73	0.26	0.15	0.08
Level of sig	**	**	**	**	**	**	**
CV(%)	6.53	2.51	2.42	3.72	3.92	2.07	3.92

** Significant at 1% level of significance

Table 5: Eigen value and percentage of variance for corresponding 17 component characters of 5 Cowa germplasm

Components	Initial Eigenvalues Total	% of Variance	Cumulative %
Leaf length (cm)	78.216	91.146	91.146
Leaf width (cm)	7.056	8.222	99.369
Leaf petiole length (cm)	0.421	0.491	99.860
Leaf petiole width (cm)	0.120	0.140	100.000
Leaf area (cm^2)	0.000	0.000	100.000
Bud length (cm)	0.000	0.000	100.000
Length of sepal (cm)	0.000	0.000	100.000
Width of sepal (cm)	0.000	0.000	100.000
Length of petals (cm)	0.000	0.000	100.000
Width of petals (cm)	0.000	0.000	100.000
Fruit weight (gm)	0.000	0.000	100.000
Fruit length (cm)	0.000	0.000	100.000
Fruit width (cm)	0.000	0.000	100.000
No. of seed/fruit	0.000	0.000	100.000
Seed weight (gm)	0.000	0.000	100.000
Seed length (cm)	0.000	0.000	100.000
Seed width (cm)	0.000	0.000	100.000

Table 6: Score of Cowa germplasm

Germplasm	PC 1	PC 2	PC 3	PC 4
ACS GC1	-0.28474	0.82323	2.1076	-0.16935
ACS GC2	0.59293	1.7345	-1.0169	-0.99438
ACS GC3	0.59333	-3.4471	-0.12362	-0.3319
ACS GC4	4.1596	0.62389	-0.3924	0.901
ACS GC5	-5.0612	0.26553	-0.57471	0.59463

(0.42 cm) (Table 3).The cow accessions exhibited variation in sepal breadth as well. Table 3 displays the observation that ACS GC4 had the highest sepal width measurement at 0.31 cm, while ACS GC5 exhibited the lowest measurement at 0.22 cm.

The petal length of Cowa accessions was observed to exhibit variation among the accessions. The findings of the study indicate that the petal length of ACS GC4 was the largest, measuring 1.27 cm. This measurement was statistically comparable to the petal lengths of ACS GC3 (1.23 cm) and ACS GC1 (1.17 cm). Conversely, the lowest petal length was observed in ACS GC5, measuring 0.87 cm (Table 3).Variation in petal breadth was seen among the different accessions. The observation revealed that ACS GC4 exhibited the greatest petal width, measuring 0.73 cm, which was comparable to ACS GC2 with a petal width of 0.70 cm. Following these, ACS GC3 displayed a petal width of 0.62 cm, ACS GC1 had a petal width of 0.60 cm, and ACS GC5 exhibited the lowest petal width at 0.52 cm (Table 3).

Morphological traits of fruits

Variation in fruit shape was observed among the Cowa germplasm. Among the five germplasm, fruit shape of three germplasm (ACS GC1, ACS GC2 and ACS GC5) was rounded and the rest two germplasm (ACS GC 3 and ACS GC 4) found globose shape. Fruit surface color also varied from brownish to yellow (Plate 3). Fruit flesh color varies from light orange to orange (Plate 4).

The fresh weight of the fruit was assessed subsequent to harvesting, revealing variations in fruit weight among different accessions. The fruit weight analysis revealed that ACS GC4 had the

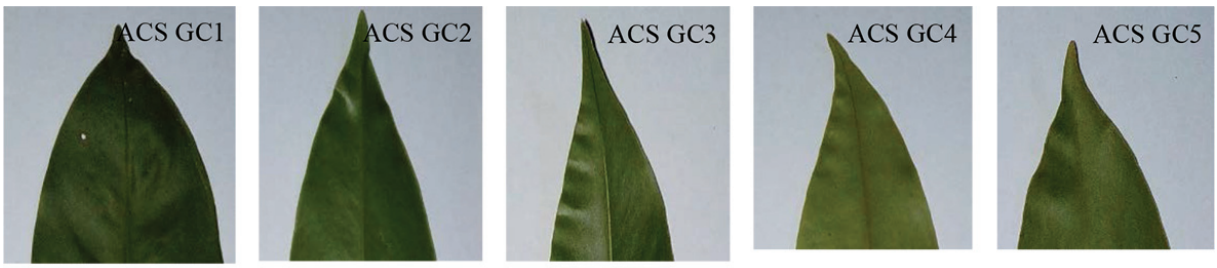

Plate 1. Leaf apex status of the Cowa germplasm

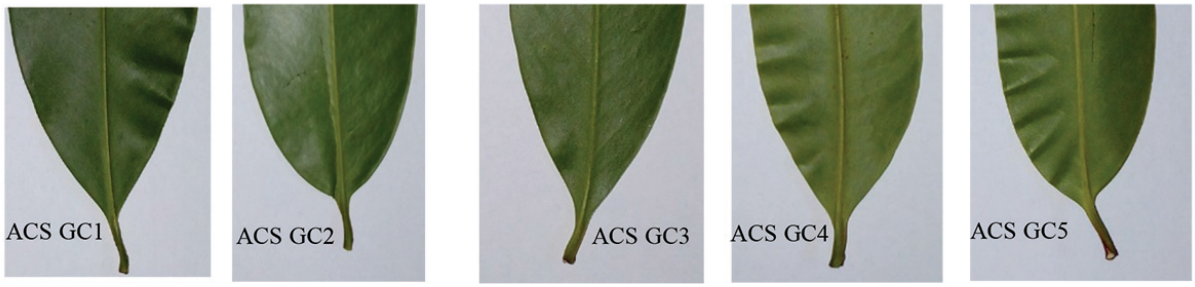

Plate 2. Leaf base status of the Cowa germplasm

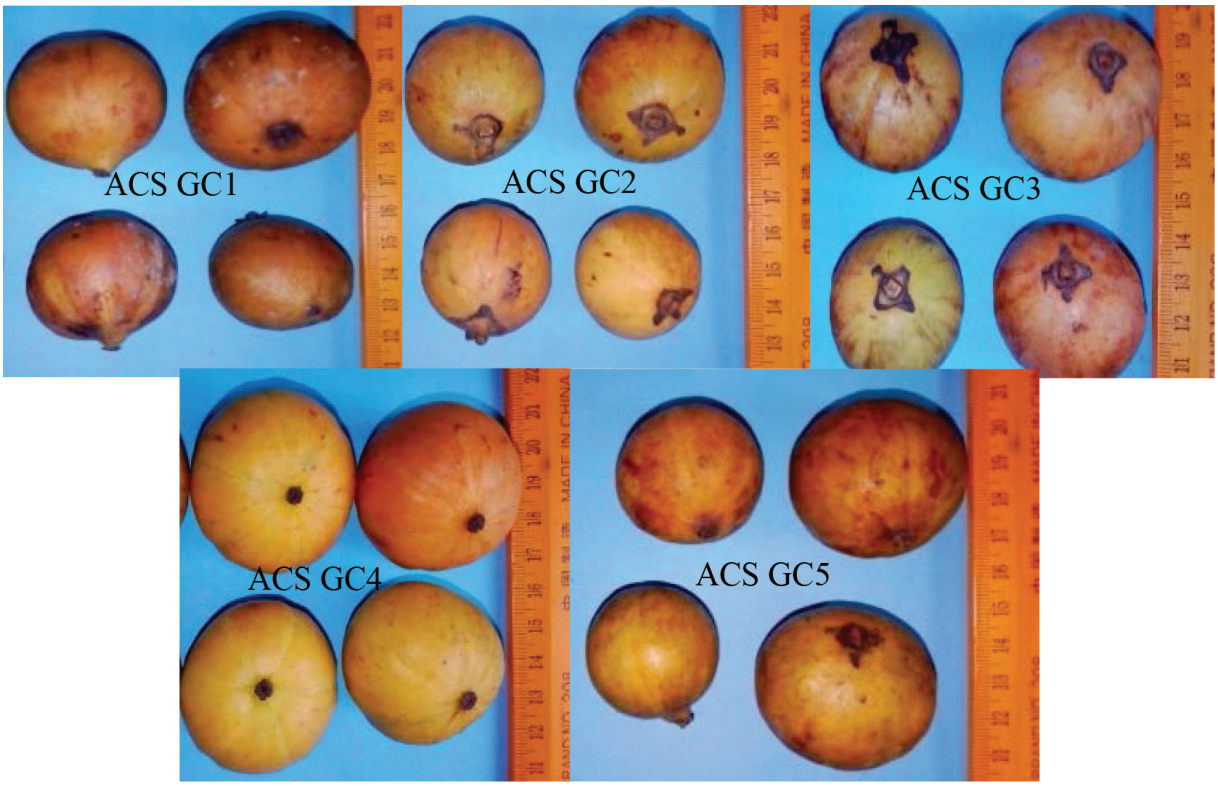

Plate 3. Fruit shape and surface color of different Cowa germplasm

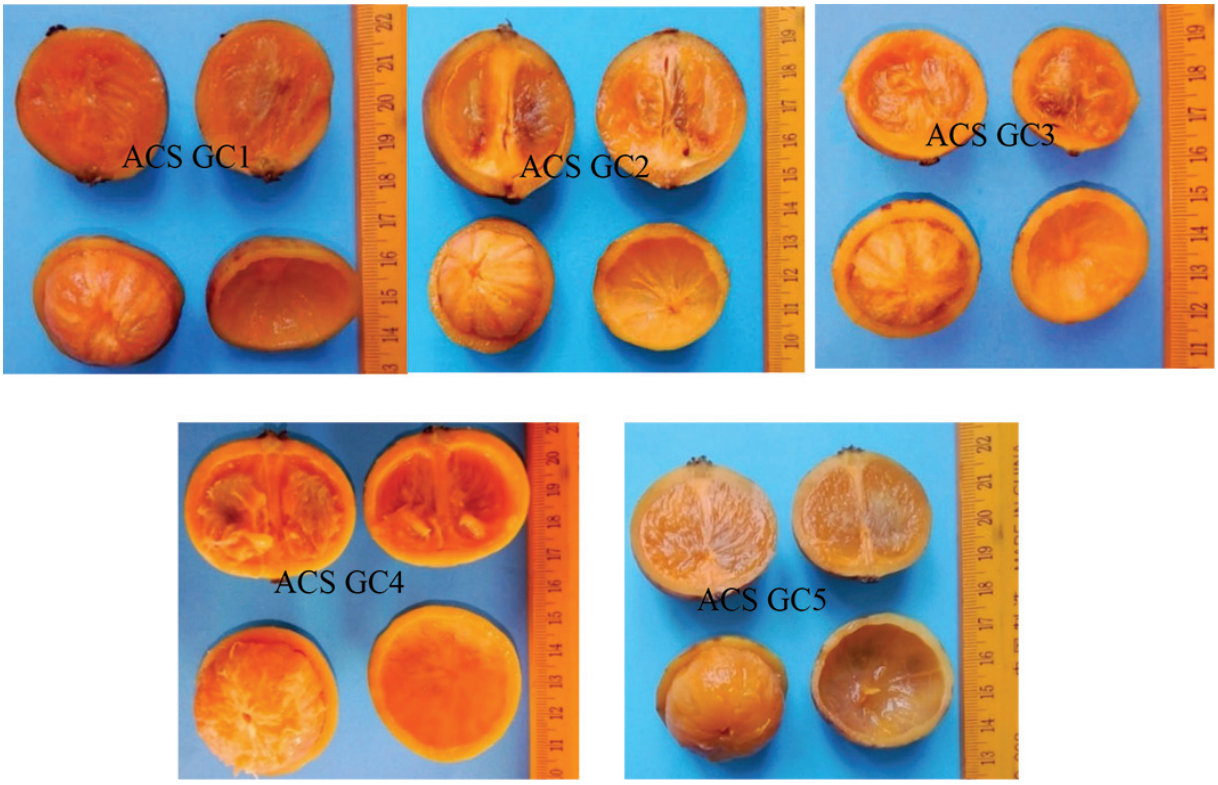

Plate 4. Flesh color of different Cowa germplasm

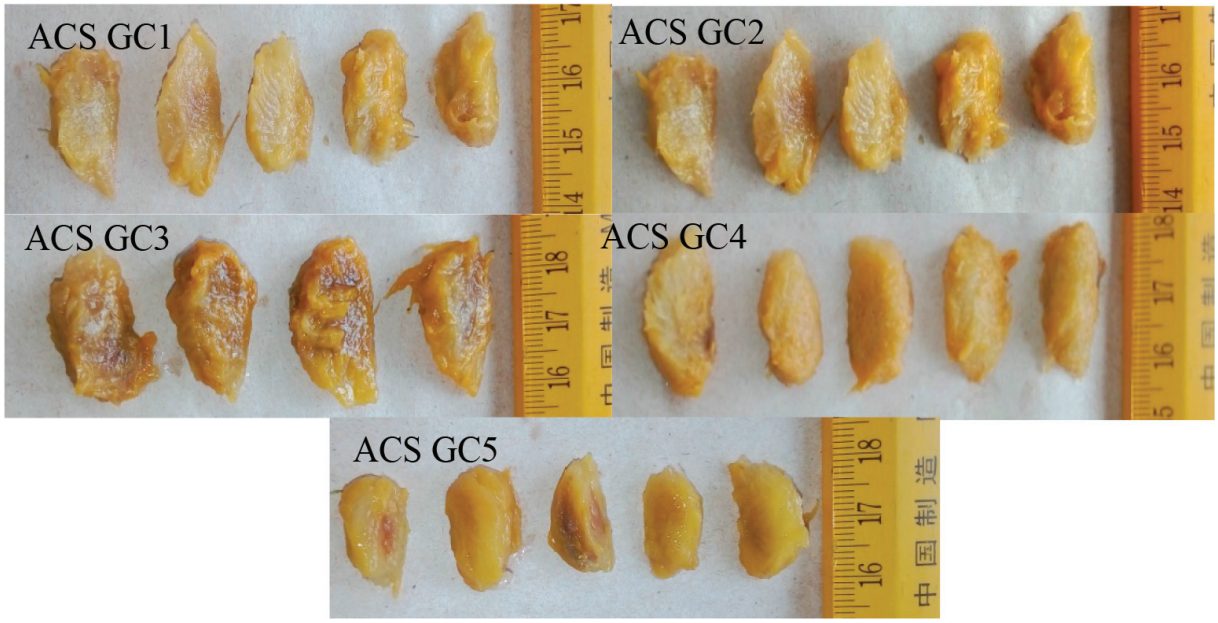

Plate 5. Seeds of different Cowa germplasm

Principal component Analysis of Cowa germplasm

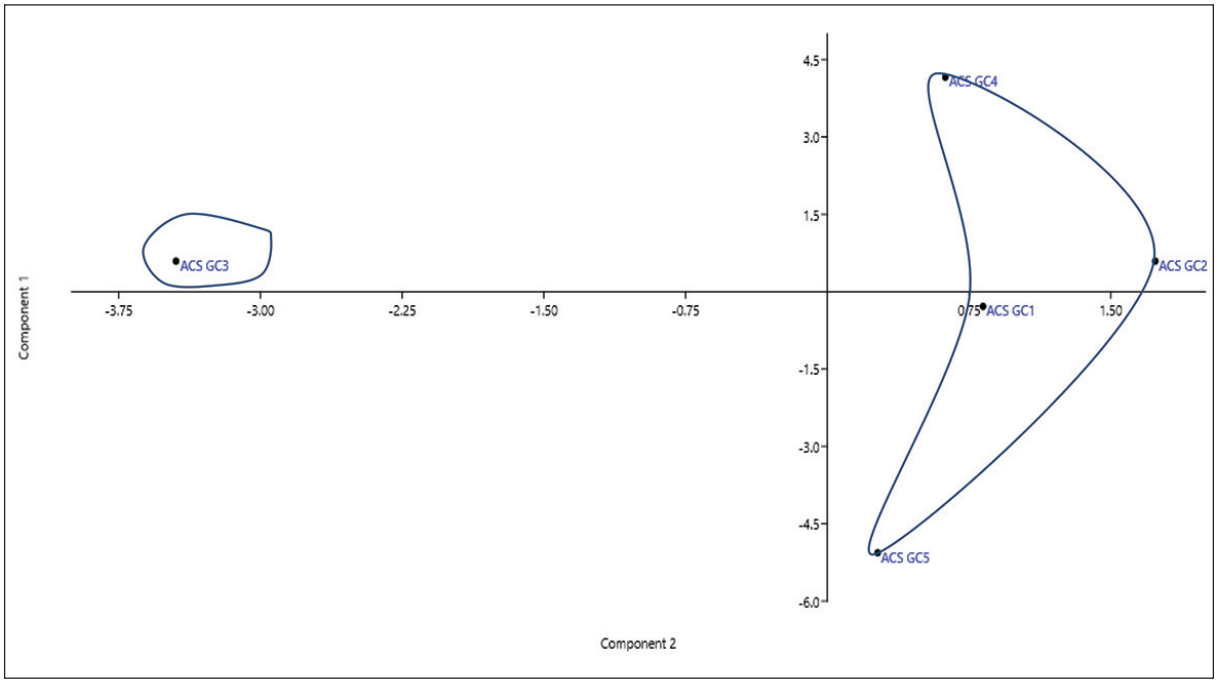

Fig. 1: Genotypic scatter plot illustrating the relationship between PC1 and PC2 germplasm

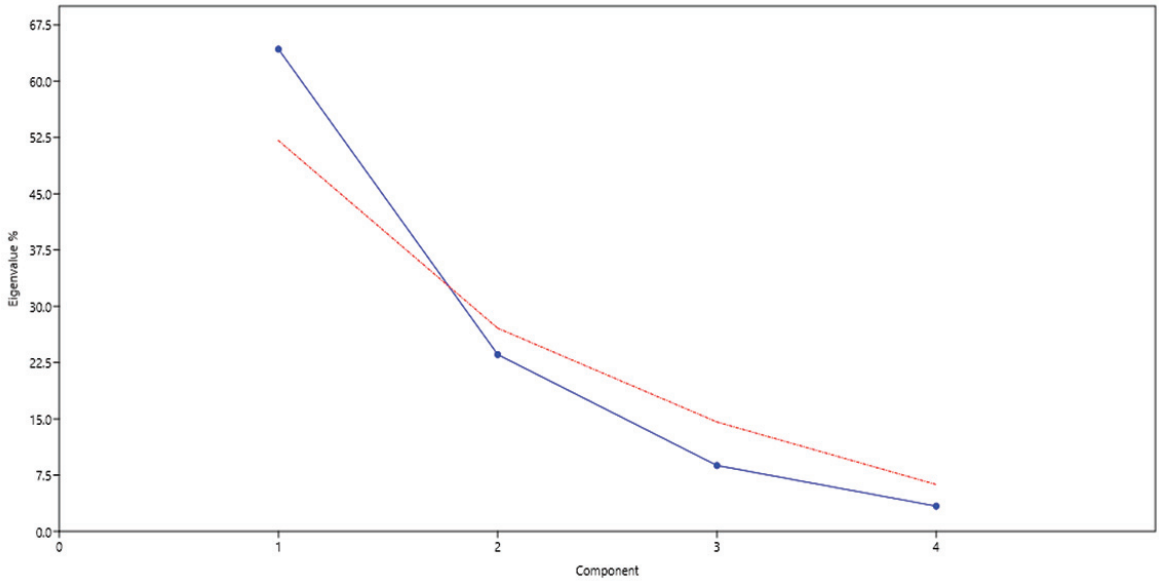

Fig. 2: Scree plot of Cowa germplasm

highest weight at 31.29 gm, followed by ACS GC3 at 28.66 gm, ACS GC2 at 24.82 gm, and ACS GC5 had the lowest weight at 18.26 gm (Table 4).There was considerable variation in fruit length among the accessions. The fruit length measurements revealed that ACS GC4 had the highest value at 4.43 cm, followed by ACS GC3 at 4.18 cm, ACS GC1 at 3.54 cm, and ACS GC5 had the lowest value at 3.11 cm (Table 4).The fruit width of various Cowa accessions exhibited significant variation among each other. The observation revealed that ACS GC4 exhibited the greatest fruit width, measuring 3.90 cm. This was followed by ACS GC3 with a fruit width of 3.81 cm, ACS GC1 with a fruit width of 3.75 cm, and ACS GC5 with the smallest fruit width of 3.37 cm, as indicated in Table 4.

Morphological traits of seeds

The hue of Cowa seeds was predominantly yellowish, as depicted in Plate 5. The seed count per fruit exhibited variability throughout the germplasm. The highest number of seeds was observed in ACS GC2 and ACS GC4, with a recorded value of 7.33. This was followed by ACS GC1 with 6 seeds, ACS GC5 with 5 seeds, and the lowest number of seeds was found in ACS GC3 with 4 seeds (Table 4).The seed weight, length, and diameter exhibited variation within the germplasm. The recorded seed weights for ACS GC4, ACS GC2, ACS GC3, and ACS GC5 were 7.42 gm, 7.11 gm, 6.96 gm, and 5.21 gm, respectively, as shown in Table 4. In a similar vein, the dimensions of the seeds were observed to be greater in ACS GC3, with a length of 2.55 cm and a diameter of 1.40 cm. This was followed by ACS GC4, which exhibited a length of 2.38 cm and a diameter of 1.20 cm. ACS GC1 displayed slightly smaller seed dimensions, with a length of 2.16 cm and a diameter of 1.14 cm. Conversely, the smallest seed dimensions were observed in ACS GC5, with a length of 1.76 cm and a diameter of 0.79 cm. These findings are presented in Table 4 and visually depicted in Plate 5.

Four lower and higher inter-accessions distances between pairs of 5 different Cowa germplasm accessions. The greatest inter genotypic distance observed from principal coordinate analysis was 78.22, which was observed between accession ACS GC3 and ACS GC1, followed by distances of 7.06, 0.42 and 0.12 among the accessions ACS GC2, ACS GC4 and ACS GC5. The shortest observed distance of 0.12 was found between ACS GC2 and ACS GC4 germplasms, while a distance of 0.42 was observed between ACS GC2 and ACS GC1 germplasm (Fig.1).

The findings of the principal component analysis unveiled that the first principal axis; leaf length largely accounted for the variation among the accessions, which alone contributed 78.216% of the total variations (Table 5). The cumulative contribution of the initial four characters along the major component axis, which possess eigen values exceeding unity, encompassed the entirety of the overall variance among 17 characters describing 5 Cowa germplasm. The rest eight characters contributed remaining 0.0% of total variation.

Principal Component Analysis (PCA) can be employed to examine the morphological features of Cowa germplasm in order to infer insights into the underlying structure of variables or components as follows:

- **Component 1** correlates well with the variables leaf petiole length, leaf area, bud length, length of sepal, width of sepal, length of petals, width of petals, fruit weight, fruit length, fruit width, seed weight and seed length showing as major factor to affect overall yield of the selected germplasm.

- **Components 2, 3, 4 and 5** were strongly correlated with variables No. of seeds/fruit, leaf width (cm), leaf length (cm) and seed weight (gm) and as express them as stand-alone factor to affect yield as a yield contributing factors.

-Variables close to an axis correlate with that principal component; one may consider that axis is a combination of its neighboring variables.

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Effect of different mulching materials on yield and quality of litchi grown in Nagaland

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ABSTRACT

To know the effect of mulching on litchi in Nagaland, an experiment was conducted with different organic and inorganic mulch materials viz., T_1 (Black polythene), T_2 (White polythene), T_3 (dry grass), T_4 (Paddy straw), T_5 (Dry Banana leaves), T_6 (Banana pseudo stem mat), T_7 (Leguminous cover crop- Soyabean) and T_8 (No mulch). The mulch materials were applied to soil surrounding the plant stem, in the month of September 2020-21 to 2021-22 in the research experimental block of Horticulture department, School of Agriculture Sciences and Rural Development, Nagaland University, Medziphema campus, Nagaland. Among different mulches, black polythene showed 27.56 & 30.07 N, 4.19 & 5.70 P and 14.87 & 11.67 K kg/ha followed by white polythene mulch with 22.39 & 24.25 N, 4.18 & 6.24 P and 18.78 & 16.68 K kg/ha availability in 2021 & 2022 respectively while, black polythene mulch showed high percent of soil moisture in 2021(14.80%) and 2022 (15.50%) retention surrounding the tree. Flowering (74.44%), fruit set (36.78%), fruit retention/panicle (13.53), average fruit weight (18.23g) and yield/ tree (18.00 kg/tree) was recorded highest in trees under black polythene mulch, which is on par with banana pseudo stem mat (72.34%, 36.06%, 13.10, 18.28g & 17.10 kg/tree) followed by soyabean cover crop (72.11%, 35.17%, 12.86, 15.47g & 12.46 kg/tree) mulching. Highest fruit cracking (16.70%) was recorded under control compared to other treatments.

***Keywords*:** Flowering, fruiting, litchi, mulching, N-P-K, soil moisture, yield

INTRODUCTION

Litchi is considered as the queen of sub-tropical fruits due to its excellent quality *viz,* juicy aril having excellent sugar and acid blend, characteristic pleasant flavour and attractive colour and also nutritional value (Pande *et al.,* 2005). It is a sub-tropical evergreen fruit tree, needs highly specific climatic requirements for improving the fruit yield and quality. Due to this reason, its cultivation is restricted to few subtropical countries in the world, where it is grown commercially (Sharma and Kathiravan, 2009). The main litchi growing countries are China, Israel, Australia, Thailand, Taiwan, India, Vietnam, parts of Africa and at higher elevations in Mexico and Central and South America. India ranks second in the world next to China in litchi production (Sahni *et al.,* 2020). In Nagaland, cultivars like China, Shahi, and Tejpur litchi are the varieties are grown and cultivar 'Shahi' being predominant in the state. Nagaland has a good potentiality of producing litchi especially in the foothills where temperature of 4-12°C is exist for a month or more. The foothills and midhills of Dimapur, Mokokchung, Wokha, Peren, Kohima and Zunhebeto districts are also congenial for litchi cultivation. Fruit maturity in this state is quite late which comes in the market up to the last week of June.

Cronje and Mosturt (2010) stated that soil moisture acts an important role in litchi cultivation with high yield and quality. Moisture deficiency at the time of flowering severely disturbs the fruit set and retention (Carr and Menzel, 2014). Soil moisture fluctuations during fruit growth cause serious reductions in individual fruit weight and in severe cases may lead to fruit cracking. This reduces the fruit quality, ultimately crop productivity and marketing. Conservation of soil moisture reserves the key interventions for bearing behaviour and quality production in litchi (Kaur and Kaundal, 2009). Physiological disorders such as poor fruit set, fruit drop, fruit cracking and sunburn can be

minimized with proper water management. Moisture conservation through mulching using dried leaves, plant parts or polythene sheet mulches has been found useful. Frequency of irrigation is reduced by adopting mulching (Shirgure *et al.,* 2003). Thus, the present experiment was conducted to observe the effect of different mulch materials on nutritional content of litchi soil, yield and quality of fruits.

MATERIALS AND METHODS

The present investigation has been conducted during 2020-21 to 2021-22 in the research experimental block of Horticulture department, School of Agriculture Sciences and Rural Development, Nagaland University, Medziphema campus, Nagaland. Twenty two years old plants of *China* variety of litchi of uniform size and vigour were selected for the study. The trail was laid out with 8-mulching treatments, *viz.,* black polythene, white polythene, dry grass, paddy straw, dry banana leaves, banana pseudo stem mat, leguminous cover cop with soyabean and no mulch following Randomized Block Design (RBD) with three replications and three plants in each. The mulch materials were applied around the plant covering 2m radius. The thickness of black and white polythene was 40 micron. Thickness of organic mulch materials was 4 cm. Different mulching treatments were applied on 15th September in each year. Weather parameter during the study has been presented in the **Annexure 1**. The prevailing climatic condition of Medziphema Campus is humid and falls under sub-tropical region with an average annual rainfall ranging from 2000-2500 mm, with predominantly high humidity of 70-90%. The mean temperature ranges from 21^{0}C to 32^0 C during summer and during winter from 10^{0}C to 15^0 C, rarely goes below 8^0 C in winter. The soil of the experimental site was sandy loam, acidic in nature with mean pH of 4.4.

Observation taken on soil parameters like available moisture per cent, available N,P & K, yield attributing parameters like and qualitative parameters like TSS, total sugar and titratable acidity were observed.

Analysis of soil NPK

Soil samples were collected before application of mulch material and after harvest of the crop in each treatment and a composite sample was prepared, thoroughly mixed analysed to determine the nutrient status of the soil. The soil samples were spread evenly and big soil clods were crushed. After drying, pounding was done with wooden pestle and mortar to break the soil aggregates. The crushed samples were passed through 2 mm (8 mesh) sieve. Available nitrogen was estimated by Kjeldahl's method as described by Jackson (1973). Available phosphorus was determined by Olsen's method, using spectrophotometer as described by Jackson (1973). Available potash was extracted and estimated by neutral normal ammonium acetate method using flame photometer (Jackson, 1973).

Observation on flowering per cent, fruit set, fruit drop, fruit weight, fruit cracking and yield were made following the method described by Rangkham (2015).

Flowering per cent: Five randomly selected flower panicles were collected from each replication and the average value was worked out and expressed in percentage.

Fruit set: The total number of flowers at full bloom and the initial number of fruits at the end of blooming stage on the labelled panicles in all treatments were counted and recorded then the percentage of fruit set was calculated as the following equation.

$$\text{Fruit set}(\%) = \frac{\text{No. of fruit lets set}}{\text{Total number of female flowers}} \times 100$$

Fruit drop: Number of fruits present on the randomly selected branches of each replication of each treatment trees at the time of fruit set were recorded and number of fruits retained on these branches till maturity was recorded. The recorded data was expressed as per cent fruit drop.

$$\text{Fruit drop}(\%) = \frac{\text{Final fruit retention}}{\text{Initial fruit set}} \times 100$$

Fruit weight: Weight of ten fruits from each treatment per replication were randomly selected and recorded by weight on top pan balance and average weight of fruit was expressed in grams (g).

Fruit cracking : Observations on fruit cracking were recorded from first May, at an interval of 7 days. For recording the data on fruit cracking one panicle was tagged in each of the four directions (east, west, north and south) of tree. Percentage fruit cracking was calculated on the basis of observations

recorded on four panicles. The percentage fruit cracking in a particular treatment was worked out by using the following formula

$$\text{Fruit cracking}(\%)=\frac{\text{No. of fruits cracked per panicle at harvesting stage}}{\text{No. of fruits retained per panicle at harvesting stage}}\times 100$$

Yield : The fruits were harvested from each replication and all the fruits from the individual trees were picked manually and collected under the trees. The total weight of the marketable fruits per tree was recorded using a pan balance of 5kg capacity and the data were expressed in kg per tree.

Fruit quality measurement

TSS: Total Soluble Solids, in the juice of representative sample were determined by using Digital refractometer (range of 0-32^0 Brix) and expressed in degree brix (^{0}B). The fruit juice was extracted from the mature fruits and the total soluble solids (TSS) were measured using a handheld refractometer, after prior calibration using distilled water. After each test, the prism plate was cleaned with distilled water and wiped with a soft tissue. The value was recorded and TSS was expressed in ^{o}Brix.

Total sugar: Total sugar content of fruit juice was determined as per Lane and Eynon method (Ranganna, 1986). 50ml filtered juice was mixed with 100ml distilled water and neutralized with 0.1N NaOH solution using phenolphthalein as indicator and the solution was allowed to stand for ten minutes. Then 8ml of potassium oxalate solution was added and total volume was made up to 250 ml by adding distilled water. 5ml of the extract was taken in burette and titrated again 10ml mixed Fehling's (5ml Fehling's solution A+5ml Fehling's solution B) solution using methyl blue as indicator. The end point is indicated by appearance of deep brick red colour precipitation. Calculation of total sugar is done with the fallowing formula :

$$\text{Total sugar}(\%)=\frac{\text{Factor of Fehling's solution}\times\text{Dilution factor}}{\text{Titre value}\times\text{wt of sample taken}}\times 100$$

Where, factor for Fehling's solution denotes the gram of invert sugar

Factor = (Titre value × 2.5)/100

Titratable acidity: Pulp (20 g) from 15 fruit without symptoms of disease was homogenized in a grinder and the supernatant phase was collected to analyze TA. Five ml aliquot was mixed with one to two drops of phenolphthalein and was titrated against 0.1N NaOH. The appearance of light pink colour marked as end point as per method described in the manual of analysing of fruits and vegetables product by Ranganna (1991). The acidity was expressed in percentage by following formula:

$$\text{Titratable acidity}(\%)=\frac{\text{Titre value}\times\text{Normality of alkali}\times\text{Equivalent weight of acid}}{\text{Volume of sample taken}}\times 1000$$

Method of statistical analysis

The mean values of different treatments were analyzed with the statistical software –OPSTAT (Sheoran *et al.,* 1998) along with corresponding standard error of mean (S.E.m±).

RESULTS AND DISCUSSION

Soil moisture content (%)

The data on soil moisture content has been presented in the Table 1 and revealed that during 2020-21, the increased soil moisture retention percentage range from -1.36 to 14.80 percent after mulching with different materials. It was recorded maximum (14.80 %) in T_1 (Black polythene mulch) whereas it was minimum (-1.36 %) in trees under T_8 (no-mulch). A similar trend was recorded during 2021-22, maximum (15.50 %) under the treatment of T_1 (Black polythene mulch) whereas it was minimum (-2.70 %) in T_8 (no-mulch). It may be due to higher percentage of moisture retaining ability under plastic mulches, due to less loss from soil. The water vapours that loss from the soil surface gets cached in the plastic film and dropped back to the soil surface which improves the soil moistness content in the near root zone (Khan *et al*. 2016).

Table 1: Effect of various mulching material on available soil moisture & nutrients

Treatments	Soil moisture (%)						Available soil Nitrogen (kg/ha)					
	2021			2022			2021			2022		
	Before mulch	After mulch	Change	Before mulch	After mulch	Change	Before mulch	After mulch	Change	Before mulch	After mulch	Change
T_1: (Black polythene mulch)	11.25	26.05	14.80	11.90	27.40	15.50	386.50	414.06	27.56	350.17	380.24	30.07
T_2: (White polythene mulch)	11.88	23.94	12.06	12.50	25.00	12.50	342.17	364.56	22.39	333.21	357.46	24.25
T_3: (Dry grass mulch)	12.29	20.48	08.19	12.36	20.41	08.05	344.50	351.94	07.44	317.97	325.78	07.81
T_4: (Paddy straw mulch)	11.87	21.33	09.46	12.97	22.80	09.83	357.20	372.47	15.27	340.92	358.66	17.74
T_5: (Dry banana leaves mulch)	10.21	15.83	05.62	10.40	17.86	07.46	340.56	345.71	05.15	322.83	329.27	06.44
T_6: (Banana pseudo stem mat mulch)	12.46	26.41	13.95	12.20	26.10	13.90	366.50	376.50	10.00	360.18	368.17	07.99
T_7: (Leguminous cover crop-Soyabean mulch)	10.18	14.25	04.07	10.67	16.71	06.04	319.70	339.00	19.30	308.56	325.58	17.02
T_8: (No- mulch)	12.92	11.56	-01.36	12.40	9.70	-02.70	331.42	336.22	04.80	328.61	331.96	03.35

Table 2: Effect of various mulching material on available soil nutrients

Treatments	Available soil P_2O_5 (kg/ha)						Available soil K_2O (kg/ha)					
	2021			2022			2021			2022		
	Before mulch	After mulch	Change	Before mulch	After mulch	Change	Before mulch	After mulch	Change	Before mulch	After mulch	Change
T_1: (Black polythene mulch)	48.54	52.73	4.19	47.28	52.98	5.70	146.94	161.81	14.87	141.83	153.50	11.67
T_2: (White polythene mulch)	43.83	48.01	4.18	41.40	47.64	6.24	155.62	174.40	18.78	152.64	169.32	16.68
T_3: (Dry grass mulch)	46.13	46.83	0.70	44.01	45.21	1.20	143.22	146.11	2.89	141.58	142.96	1.38
T_4: (Paddy straw mulch)	49.11	52.21	3.10	44.56	46.17	1.61	151.28	164.30	13.02	149.54	161.27	11.73
T_5: (Dry banana leaves mulch)	43.18	43.58	0.40	40.72	41.72	1.00	144.46	146.97	2.51	142.50	145.17	2.67
T_6: (Banana pseudo stem mat mulch)	41.97	43.82	1.85	38.23	40.94	2.71	135.78	145.23	9.45	132.28	140.81	8.53
T_7: (Leguminous cover crop-Soyabean mulch)	50.17	52.74	2.57	46.28	49.30	3.02	139.50	147.50	8.00	136.82	141.50	4.68
T_8: (No- mulch)	41.61	41.91	0.30	37.94	38.50	0.56	137.64	140.24	2.60	136.91	138.24	1.33

Table 3: Effect of various mulching material on flowering and fruit parameters

Treatments	Flowering percentage (%)			Fruit set percentage/ panicle (%)			Fruit drop percentage (%)			Fruit retention/ panicle(number)		
	2021	2022	Pooled	2021	2022	Pooled	2021	2022	Pooled	2021	2022	Pooled
T_1: (Black polythene mulch)	73.15	75.73	74.44	35.12	38.45	36.78	69.00	67.19	68.09	12.99	14.08	13.53
T_2: (White polythene mulch)	71.45	72.61	72.03	36.00	37.22	36.61	72.90	72.60	72.75	12.50	13.70	13.10
T_3: (Dry grass mulch)	64.12	65.66	64.89	32.50	34.15	33.32	73.33	74.66	73.99	11.30	12.70	12.00
T_4: (Paddy straw mulch)	66.33	69.74	68.03	34.99	35.45	35.22	75.41	73.50	74.45	12.00	12.99	12.49
T_5: (Dry banana leaves mulch)	65.00	65.87	65.43	32.33	34.12	33.22	78.08	75.93	77.00	11.56	12.41	11.98
T_6: (Banana pseudo stem mat mulch)	70.20	74.49	72.34	36.00	36.12	36.06	72.44	70.63	71.53	12.88	13.33	13.10
T_7: (Leguminous cover crop-Soyabean mulch)	72.00	72.22	72.11	34.93	35.42	35.17	76.70	73.00	74.85	12.43	13.30	12.86
T_8: (No- mulch)	40.30	41.58	40.94	30.63	32.20	31.41	86.52	81.12	83.82	9.23	11.90	10.56
SEm±	**0.28**	**0.58**	**0.69**	**0.70**	**0.01**	**0.51**	**0.91**	**0.90**	**1.01**	**0.39**	**0.04**	**0.33**
CD@5%	**0.95**	**1.98**	**2.35**	**2.40**	**0.04**	**1.73**	**3.10**	**2.75**	**3.43**	**1.35**	**0.16**	**1.12**

Table 4: Effect of various mulching material on fruit quality attributes

Treatments	Fruit weight (g)			Fruit cracking percentage (%)			Yield (kg/tree)		
	2021	2022	Pooled	2021	2022	Pooled	2021	2022	Pooled
T_1: (Black polythene mulch)	17.52	18.95	18.23	13.26	13.04	13.15	17.34	18.67	18.00
T_2: (White polythene mulch)	16.33	16.75	16.54	13.51	13.20	13.35	16.59	17.57	17.08
T_3: (Dry grass mulch)	12.89	13.14	13.01	13.00	13.90	13.45	12.46	13.19	12.82
T_4: (Paddy straw mulch)	16.21	16.00	16.10	13.94	13.75	13.84	14.28	14.34	14.31
T_5: (Dry banana leaves mulch)	12.24	12.60	12.42	15.16	14.18	14.67	12.02	12.25	12.13
T_6: (Banana pseudo stem mat mulch) 18.00	18.56	18.28	13.68	13.70	13.69	17.00	17.21	17.10	
T_7: (Leguminous cover crop- Soyabean mulch)	15.43	15.52	15.47	13.70	13.79	13.74	12.42	12.50	12.46
T_8: (No- mulch)	10.42	9.33	9.87	16.23	17.18	16.70	10.53	11.49	11.01
SEm±	**0.19**	**1.01**	**0.35**	**0.38**	**0.004**	**0.31**	**0.36**	**0.22**	**0.24**
CD@5%	**0.67**	**3.43**	**1.21**	**1.29**	**0.015**	**1.08**	**1.25**	**0.77**	**0.83**

Table 5: Effect of various mulching material on bio chemical attributes of litchi

Treatments	TSS (°B)			Total sugar (%)			TitraTable acidity (%)		
	2021	2022	Pooled	2021	2022	Pooled	2021	2022	Pooled
T_1: (Black polythene mulch)	17.82	17.50	17.66	16.89	16.14	16.51	0.55	0.45	0.50
T_2: (White polythene mulch)	17.19	16.55	16.87	15.50	14.64	15.07	0.50	0.50	0.50
T_3: (Dry grass mulch)	14.62	15.20	14.91	13.84	13.76	13.80	0.40	0.55	0.47
T_4: (Paddy straw mulch)	15.00	15.75	15.37	14.05	14.07	14.06	0.55	0.62	0.58
T_5: (Dry banana leaves mulch)	14.26	15.30	14.78	12.38	12.69	12.53	0.67	0.70	0.68
T_6:(Banana pseudo stem mat mulch)	15.61	14.75	15.18	14.97	14.34	14.65	0.44	0.50	0.47
T_7: (Leguminous cover crop- Soyabean mulch)	14.87	14.00	14.43	12.56	11.86	12.21	0.50	0.60	0.55
T_8: (No- mulch)	14.52	13.65	14.08	12.27	12.59	12.43	0.70	0.75	0.72
SEm±	**0.61**	**0.44**	**0.40**	**0.12**	**0.49**	**0.24**	**0.03**	**0.02**	**0.03**
CD@5%	**2.10**	**1.51**	**1.37**	**0.43**	**1.67**	**0.83**	**0.11**	**0.08**	**0.12**

Available nitrogen content in the soil (kg/ha)

During 2020-21, soil nitrogen content was increased from 4.80 to 27.56 kg/ha after mulching with different organic and in organic materials, it was recorded maximum under the treatment of T_1 (black polythene mulch) *i.e.*, 27.56 kg/ha followed by T_2 and T_7 (white polythene mulch and leguminous cover crop- soyabean mulch) *i.e.*, 22.39 and 19.30 kg/ha respectively, whereas minimum (4.80 kg/ha) with T_8 (no-mulch) treatment. A similar trend was recorded during 2021-22, where nitrogen content increased from 3.35 to 30.07 kg/ha after mulching (Table 1).

Available phosphorus and potassium content in the soil (kg/ha)

During 2020-21, soil phosphorus content was increased from 0.30 to 4.19 kg/ha after mulching with different organic and in organic materials, it was recorded maximum under the treatment of T_1 (black polythene mulch) *i.e.*, 4.19 kg/ha followed by T_2 and T_4 (white polythene mulch and paddy straw mulch) *i.e.*, 4.18 and 3.10 kg/ha, whereas minimum (0.30 kg/ha) with T_8 (no-mulch) treatment. A similar trend was recorded during 2021-22, where phosphorus content increased from 0.56 to 6.24 kg/ha after mulching. Among the treatments maximum (6.24 kg/ha) soil available phosphorus recorded under T_2 (white polythene mulch) followed by T_1 (black polythene mulch) *i.e.*, 5.70 kg/ha, whereas it was minimum (0.56 kg/ha) with T_8 (no-mulch) treatment (Table 2).

During 2020-21, soil potassium content was increased from 2.51 to 18.78 kg/ha after mulching with different organic and in organic materials, it was recorded maximum under the treatment of T_2 (white polythene mulch) *i.e.*, 18.78 kg/ha followed by T_1 (black polythene mulch) *i.e.*, 14.87 kg/ha, whereas minimum (2.51 kg/ha) with T_5 (dry banana leaves mulch) treatment. A similar trend was recorded during 2021-22, where potassium content increased from 1.33 to 16.68 kg/ha after mulching. Among the treatments maximum (16.68 kg/ha) soil available potassium recorded under T_2 (white polythene mulch) whereas it was minimum (1.33 kg/ha) with T_8 (no-mulch) treatment (Table 2).

High availability of nutrients on soil surface under polythene film mulch was the effect of mineralization of organic content (Das and Dutta,

Annexure 1: Weather parameter during the period of study.

Month	Temperature (ºC)				Relative Humidity (%)				Rainfall (mm)	
	Max.		Min.		Max.		Min.			
	2020- 21	2021- 22	2020- 21	2021-22	2020- 21	2021-22	2020- 21	2021- 22	2020-21	2021- 22
September	34.00	33.10	23.50	23.80	93.00	94.00	68.00	68.00	98.70	116.20
October	33.80	32.10	23.00	22.10	95.00	95.00	67.00	68.00	114.30	130.00
November	30.00	28.50	15.00	14.80	95.00	96.00	51.00	51.00	00.00	00.00
December	26.50	25.10	12.50	11.30	94.00	95.00	50.50	51.00	02.50	16.40
January	24.00	22.70	08.90	10.10	96.00	96.00	50.00	56.00	03.40	34.60
February	27.10	23.20	09.70	09.60	95.00	95.00	40.00	48.00	02.30	56.30
March	31.10	32.20	14.90	15.50	93.00	90.00	41.00	40.00	43.50	02.30
April	33.10	30.90	17.90	19.90	87.00	90.00	34.00	68.00	59.60	175.70
May	32.80	30.50	21.90	21.90	90.00	92.00	58.00	71.00	85.40	224.70
June	33.10	32.00	24.30	23.90	93.00	95.00	69.00	72.00	117.40	160.80

2018). Parallel results was also got by Duta and Majmder (2009) in *Psidium gujava*.

Flowering and fruiting

The effect of different mulching treatments was found to be significantly induced the flowering in the litchi during both the years as compared to control(Table 3). Pooled data of two consecutive years, showed highest percentage (74.44 %) of flowering was found in T_1 (black polythene mulch) which was at par with T_6 (banana pseudo stem mat mulch) and T_7 (leguminous cover crop- soyabean mulch) and the value were 72.34 and 72.11 percent respectively. The lowest flowering (40.94 %) was observed in T_8 (no-mulch), The results are in line with the findings of Mal *et al*. (2006) who reported that a greater number of flowers recorded in plants under black polythene mulch in pomegranate cv. Ganesh. The pooled data of 2021 & 2022 showed that maximum fruit set percentage content (36.78 %) was observed in treatment T_1 (black polythene mulch) while the minimum fruit set percentage content (31.41 %) was recorded in T_8 (no-mulch). which was at par with T_2 (white polythene mulch), T_6 (banana pseudo stem mat mulch), T_4 (paddy straw mulch) and T_7 (leguminous cover crop- soyabean mulch) and the value were 36.61, 36.06, 35.22 and 35.17 percent respectively.

Bakshi *et al.* (2014) also stated maximum number of fruits per plant recorded under black polythene mulch in strawberry cv. Chandler. It might be due to good weed control was found under the effect of mulch and drip irrigation which reduced the competition for nutrients and soil moisture, it leads to better flowering and fruiting percentage.

The lowest (10.56) number of fruits retention at harvest was recorded in T_8 (no-mulch), whereas treatments T_1 (black polythene mulch) recorded maximum (13.53) number of fruits per panicle, followed by T_2 (white polythene mulch), T6 (banana pseudo stem mat mulch), T_7 (leguminous cover crop- soyabean mulch) and T_4 (paddy straw mulch) *i.e*., 13.10, 13.10, 12.86 and 12.49 respectively (Table 3). Singh *et al*. (2015) recorded maximum number of fruits in guava cv. Allahabad safeda under plastic mulch with drip irrigation.

The data presented in Table 4 reveals that weight of fruit varied from 9.87 to 18.28 g with significant

differences among the treatments. However highest fruit weight (18.28 g) was observed in T_6 (banana pseudo stem mat mulch) followed by T_1 (black polythene mulch) i.e., 18.23 g. Whereas lowest (9.87 g) was reported in T_8 (no-mulch). On the other hand, all the treatments had a significant effect on fruit weight as compared to control. Kumari and Khare (2019) reported similar findings in litchi fruit that the fruit weight (23.5g) was found maximum in plastic mulched trees followed by dry leaves mulched trees having fruit weight (21.8g). Similarly, Das and Dutta (2018) in mango recorded that, maximum fruit weight (263.42 g) under black film mulch. It may be due to improved soil moisture maintenance and good soil temperature maintained under the mulched treatments.

Fruit cracking

Data given in Table 4 indicates that there was significant effect of different mulching material on fruit cracking. Minimum fruit cracking (13.15 %) was observed in T_1 (black polythene mulch) followed by T_2 (white polythene mulch) *i.e*., 13.35 % which was significant over all treatments and maximum cracking was observed in T_8 (un-mulched) fruits of 16.70 percent. Joshi *et al*. (2011) observed significant reduction in fruit cracking in litchi with the application of mulch and drip irrigation. The organic and inorganic mulching materials improved available soil moisture and nutrients in plant basin due to which the treatments plants showed less cracking percentage.

Fruit yield

The data in Table 4 depicted that effect of different mulching materials in respect to litchi yield was found to be significant due to different treatments. The average yield during both the years ranged from 11.01 to 18.00 kg/tree in various treatments. Highest yield (18.00 kg/tree) was recorded in T_1 (black polythene mulch) fallowed by T_6 (banana pseudo stem mat mulch) *i.e*., 17.10 kg/tree and the lowest (11.01 kg/tree) in trees under T_8 (un-mulch).

Similarly, Bakshi *et al*. (2014) evaluated the effect of mulching material on yield of strawberry and reported that maximum yield per plant was under black polythene because of larger fruit owing to better hydrothermal regime of soil and complete weed-free environment. Das and Dutta (2018) also recorded yield (243.72 fruits/tree) in polythene mulch, while un-mulched (control) gave the minimum values (192.72 fruits/tree) in mango.

Fruit quality parameters

The data presented in Table 5 shows that all the treatments significantly increased the Total soluble solids content in the litchi. The pooled data of 2021 & 2022 showed that maximum TSS content (17.66 ºB) was observed in treatment T_1 (black polythene mulch) followed by T_2 (white polythene mulch) (16.87 ºB) while the minimum TSS content (14.08 ºB) was recorded in T_8 (un-mulched).

Das and Dutta (2018) reported maximum (19.20 ºB) TSS in black polythene mulch than un-mulched treatment (table 5). Improvement in fruit qualitative attributes with various mulching treatments may be due to the result of leaf potassium and an enhanced rate of photosynthesis which cumulatively enhanced the fruit quality. Iqbal *et al*. (2015) also reported similar findings that the total soluble solids were recorded highest in black polythene (10.73 ºB) followed by paddy straw mulch (10.20 ºB) while, the treatment un-mulched control produced the fruits of minimum TSS (9.70 ºB) in aonla.

The data presented in Table 5 shows that all the treatments significantly affected total sugar content. The pooled analysis of total sugar content indicated that maximum total sugar content (16.51 %) was found in treatment T_1 (black polythene mulch) followed by T_2 (white polythene mulch) (15.02 %). On the other hand, minimum total sugar content (12.21 % and 12.43 %) was found in treatment T_7 (leguminous cover crop- soyabean mulch) and T_8 (no-mulch). Enhanced sugars may be due to slow hydrolysis of starch to sugars and the gradual build-up of sugars during ripening of fruits (Kulkarni and Yewale 2012).

The data presented in Table 5 shows that all the treatments significantly reduced the titratable acidity per cent. A critical examination of pooled data indicated that treatments T_8 (no-mulch) resulted in maximum acidity per cent (0.72) whereas, the minimum acidity (0.47 % and 0.47 %) was recorded with T_3 (dry grass mulch) and T_6 (banana pseudo stem mulch). Iqbal *et al*. (2015) also reported that maximum titratable acidity (1.92%) was recorded in fruits under un-mulched

plants while the least titratable acidity (1.64%) was recorded under black polythene mulching in aonla. Maximum acidity was obtained in control may be due to reduced cell size and cell division due to less turgor pressure and internal auxin content. Highest percentage of acidity was also recorded by El-Tawell and Farag, 2015 in un-mulched plants of pomegranate.

CONCLUSION

In the case of above study, findings revealed that different mulch materials significantly affected on the soil moisture content (%), soil available nutrients (kg/ha), fruit retention, number of fruits/ panicle and bio-chemical quality parameters of fruits. Polythene mulch, paddy straw mulch and banana pseudo stem mat mulching were found to give best results in retaining moisture, available nutrients content and fruit yield.

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Performance of pomegranate cultivars under semi-arid climatic conditions

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ABSTRACT

Pomegranate (Punica granatum L.) is a popular fruit crop of arid and semiarid regions of the world with immense therapeutic and nutritional value. An experiment was conducted at the Rani Lakshmi Bai Central Agricultural University, Jhansi to evaluate the performance of eight pomegranate cultivars under semi-arid climate during 2020–22. The cultivars were Bhagwa, Super Bhagwa, Ganesh, G-137, Ruby, Mridula, Arkata and Jalore Seedless. The results showed that Jalore Seedless had the tallest plant, G-137 had the widest canopy (east-west) and Ruby had the widest canopy (north-south). The flowering period varied from last week of February to 3rd week of April among different cultivars. The fruit maturity period ranged from 145 days to 170 days. Bhagwa had the highest percentage of hermaphrodite flowers and Jalore Seedless and Ganesh had the highest percentage of male flowers. Ganesh had the highest pollen viability percentage and Jalore Seedless had the lowest. Super Bhagwa had the highest yield (15.7 kg), number of fruits per plant (61.0) and fruit weight (258.4g). Ganesh, G-137, Ruby, Mridula and Arkata had round fruits, Bhagwa, Super Bhagwa and Jalore Seedless had ovate fruits. Bhagwa, Super Bhagwa, Ruby, Mridula and Arkata had red fruit skin. Bhagwa and Super Bhagwa had red arils, Mridula and Arkata had dark red arils. Bhagwa, Super bhagwa G-137, Ruby and Mridula had low fruit drop. Bhagwa, Super Bhagwa, Ruby, Mridula, Arkata, Jalore Seedless had low fruit cracking. Among the cultivars tested, Bhagwa, Ruby, Mridula and Arkata showed low fruit borer attack. Super Bhagwa and Ganesh had the highest TSS content (14.3^{0}Brix), while G-137 had the lowest acidity content (0.43%). Ganesh also had the highest TSS/acid ratio and total sugar content, but Bhagwa had the highest vitamin C content. Super Bhagwa was the best cultivar for marketable yield, fruit quality and pest tolerance under semi-arid conditions of Bundelkhand region.

***Keywords*:** Cultivars, flowering, pomegranate, yield and quality

INTRODUCTION

Pomegranate (*Punica granatum* L.) is an ancient and valuable fruit crop that belongs to the Punicaceae family. It can grow in various agro-climatic regions, especially in arid and semi-arid areas, where it produces high-quality fruits with hardy and adaptable characteristics (Teixeira da Silva *et al*., 2013; Marathe *et al*., 2017). The fruit is popular for its juicy, sweet arils that have a refreshing taste. The fruit has a high demand for fresh consumption and processing into various products, such as juice, syrup, squash, wine and anardana, a souring agent. Pomegranate is also a functional food that has many health benefits, as it contains several phytochemicals that can prevent diseases (Kanoun *et al.,* 2020; Parashar, 2010; Melgarejo and Salazar, 2003).

India is one of the world's largest pomegranate producers and has a year-round production cycle with a peak season from February to May. The main pomegranate growing states in India are Maharashtra, Gujarat, Andhra Pradesh, Karnataka, Tamil Nadu and Rajasthan. Maharashtra alone accounts for more than 70% of the total area under pomegranate and produces 17.48 lakh MT of pomegranate annually (NHB, 2022). The country has seen a rise in the area and production of pomegranate due to the identification and development of suitable cultivars and standardization of their agro-techniques. Pomegranate can adapt to a wide range of climates from tropical to temperate regions, and it shows a lot of variability depending on genetical, soil type, environmental factors. In this study, we collected

promising pomegranate genotypes, grew them under semi-arid region of Jhansi and evaluated their performance to determine the best cultivars for local climatic conditions.

MATERIALS AND METHODS

Eight promising cultivars were collected from National Research Centre on Pomegranate, Solapur, Maharashtra in September, 2017. The hard wood rooted cutting planting materials of cvs 'Arkata', 'Bhagwa', 'Super Bhagwa', 'Ganesh', 'G-137', 'Jalore Seedless', 'Mridula' and 'Ruby' planted at 3 x 4 m spacing during 2017-18 at Fruit Research Station, Rani Lakshmi Bai Central Agricultural University, Jhansi. Geographically the farm is situated at latitude $25^0$30'' N, longitude $78^0$32''E and at an elevation of 258 meters above main sea level. It has semi-arid climate with average annual rainfall of 800 to 900 mm and temperature ranging between 3.0°C to 47.8°C. These climatic conditions are considered ideal for the cultivation of high-quality pomegranates.

The experiment was carried out for two consecutive years during ambe bahar i.e., flowering period of March-April in the years of 2021 and 2022 at fruit research station, RLBCAU, Jhansi. The experiment was conducted in randomized block design with five replications. The plants were given uniform cultural operations.

The observations recorded were plant height, plant spread, tree growth habit, foliage density, floral biology, fruit set to maturity period, yield and quality attributes characters. The plant height was measured from ground the ground level to the apex of the crown, using a marking stick and average height of the replication was expressed in cm. Spread of the plant from two sides, i.e., east-west and north-south was observed with the help of a linear scale and expressed in cm. The growth habit was observed and recorded as per its visual appearance and expressed as spreading, semi-spreading and erect type based on plant height to spread ratio. Foliage density was observed in running per meter and interpreted as sparse, medium and dense. The plant growth parameters were recorded, as recommended in the descriptor of NBPGR (Mahajan *et al.*, 2002) and guidelines for DUS testing of PPV and FRA. Simultaneously, differences for appearance of flower bud to full bloom period and fruit set to maturity were noted and expressed as flowering and fruit set to maturity duration in terms of days. The percentage of hermaphrodite/male flowers was computed by dividing total number of hermaphrodite/male flowers blossomed within the tagged flowers by total numbers of flower. The sex ratio of flowers was computed by dividing the number of hermaphrodite flowers by the number of male flowers. The yield was recorded at the time of harvest and expressed in terms of kg per plant. The total soluble solids were determined with the help of a hand refractometer and reading corrected at room temperature. For biochemical analysis of the fruits, the methods were followed as described by A.O.A.C (1990). The data were analyzed statistically and test of significance were done by following the statistical method RBD as described by Panse and Sukhatme (1985)

RESULTS AND DISCUSSION

Plant growth parameters

Table 1 shows the plant growth parameters of eight pomegranate cultivars. These parameters include plant height, spread, growth habit and foliage density. The plant height among the selected cultivars exhibited significant variations and the maximum plant height was recorded in 'Jalore Seedless' (230.0 cm) followed by Ruby (222.6 cm), G-137 (221.8 cm), whereas 'Arkata' gave the minimum plant height (178.4 cm). The plant height varied from 2.13 m to 2.63 m among eight pomegranate cultivars grown in Jammu climate, which is consistent with the findings of Bhat *et al*. (2019). The genetic makeup of the plant may influence these variations in plant height. However, the variations in the plant height of same varieties in the different area may be due to adaptability and performance of varieties with the prevailing local soil and climate condition.

The highest plant spread on east-west direction was recorded for G-137 (201.8 cm), whereas, Arkata exhibited the lowest east-west spread (103.8 cm). Ruby gave the highest north-south spread (201.0 cm) and Arkata exhibited the lowest north-south spread (101.2 cm). Similar type of variation in plant height and plant spread has been reported in pomegranate (Sharma and Bist, 2005; Meena *et al*., 2011; Bhat *et al*., 2019). Out of eight genotypes,

Table 1: Plant growth parameters of eight cultivars of pomegranate

Treatments	Tree height (cm)	Tree spread (cm)		Tree growth habit	Tree foliage density
		E-W	N-S		
Bhagwa	213.0	172.4	198.2	Spreading	Medium
Super Bhagwa	212.4	170.0	155.8	Spreading	Medium
Ganesh	204.2	176.4	174.6	Spreading	Medium
G-137	221.8	201.8	181.4	Spreading	Medium
Ruby	222.6	191.6	201.0	Spreading	Dense
Mridula	201.0	162.0	170.0	Spreading	Dense
Arkata	178.4	103.8	101.2	Compact	Dense
Jalore Seedless	230.0	195.2	176.2	Upright	Medium
CD at 5%	15.17	15.58	14.65	-	-

Table 2: Duration of flower and fruit set to maturity of eight cultivars of Pomegranate.

Treatments	Date of start of flowering	Date of end of flowering	Duration from fruit set to maturity
Bhagwa	2nd week of March	3rd week of April	170.0
Super Bhagwa	2nd week of March	3rd week of April	169.0
Ganesh	3rd week of March	1st week of April	162.0
G-137	3rd week of March	1st week of April	153.0
Ruby	1st week of March	3rd week of April	164.0
Mridula	Last week of February	2nd week of April	145.0
Arkata	Last week of February	2nd week of April	154.0
Jalore Seedless	1st week of March	2nd week of April	164.0
CD at 5%	-	-	8.94

Table 3: Yield attributes characters of eight pomegranate cultivars

Treatments	Fruit yield per plant (kg)	No. of the fruits per plant	Fruit weight (g)	Fruit shape	Fruit skin colour	Aril colour
Bhagwa	10.6	43.0	247.0	Ovate	Red	Red
Super Bhagwa	15.7	61.0	258.4	Ovate	Red	Red
Ganesh	10.6	45.0	235.8	Round	Yellowish	Pink
G-137	12.2	51.8	236.8	Round	Yellowish	Pink
Ruby	12.7	55.2	231.2	Round	Red	Pink
Mridula	7.9	39.8	200.4	Round	Red	Dark Red
Arkata	8.9	45.2	197.0	Round	Red	Dark Red
Jalore Seedless	6.4	25.6	252.8	Ovate	Yellowish	White
CD at 5%	3.7	16.32	21.52	-	-	-

Bhagwa, Super Bhagwa, Ganesh, G-137, Ruby, Mridula showed spreading tree growth habit. Most pomegranate varieties exhibited a spreading growth habit, except for Arkata and Jalore Seedless, which had a compact and upright habit. These results are consistent with those of Sharma and Bist (2005), who reported that the cultivars Ganesh, Mridula and G-137 showed a spreading growth habit. The tree foliage density i.e., dense foliage density was showed in Ruby, Mridula and Arkata whereas,

Table 4: Physiological disorder, insect and pest incidence of eight cultivars of pomegranate

Treatments	Fruit Drop (kg/plant)	Fruit cracking (kg/plant)	Sun scald	Fruit borer	Fruit sucking moths	Bacterial blight	Wilt
Bhagwa	1.45	1.32	Low	Low	Nil	Nil	Nil
Super Bhagwa	1.49	1.17	Low	Medium	Nil	Nil	Nil
Ganesh	2.24	2.27	Low	Medium	Nil	Nil	Nil
G-137	1.66	2.29	Low	Medium	Nil	Nil	Nil
Ruby	1.62	1.70	Low	Low	Nil	Nil	Nil
Mridula	1.40	1.45	Low	Low	Nil	Nil	Nil
Arkata	1.53	1.20	Low	Low	Nil	Nil	Nil
Jalore Seedless	2.86	1.74	No	Medium	Nil	Nil	Nil
CD at 5%	0.54	0.63	-	-	-	-	-

Table 5: Fruit quality parameters of eight cultivars of pomegranate

Treatments	TSS (^{0}Brix)	Acidity (%)	TSS/Acid ratio	Total sugar (%)	Vitamin-C (mg/100g of aril)
Bhagwa	12.4	0.53	23.3	8.92	11.64
Super Bhagwa	14.3	0.48	29.7	9.00	10.82
Ganesh	14.3	0.47	30.4	9.03	10.57
G-137	12.1	0.43	28.1	8.70	11.02
Ruby	12.7	0.52	24.4	8.45	10.00
Mridula	12.4	0.44	28.1	8.64	10.34
Arkata	11.6	0.56	20.7	8.33	10.56
Jalore Seedless	13.2	0.60	22.0	8.54	10.11
CD at 5%	0.75	0.05	3.24	0.26	0.20

cultivar Bhagwa, Super Bhagwa, Ganesh, G-137 and Jalore Seedless showed medium foliage density.

Time and duration of flowering, flower type, pollen viability, maturity of fruits

Under climatic conditions at Jhansi, the pomegranate was observed to flower throughout the year. However, with the objective of obtaining higher fruit productivity, minimizing the loss due to vagaries of climate and ensuring confident demand in the market for the produce (with dark aril colour, higher production with less incidence of insect and pest), its flowering is conventionally regulated in three distinct seasons viz., flowering during February-March (ambe bahar), flowering during July-August (mrigbahar) and flowering during September-October (hasth bahar). The data presented table 2 reveals that in Jhansi climatic condition, date of start of flowering varied among different cultivars and was recorded between last week of February to 3rd week of March. Earliest cultivar to start first flowering was Mridula and Arkata (last week of February) whereas last cultivar to open its first flowering was Ganesh and G-137 (3rd week of March). The earliest cultivar to open its last flower was Ganesh and G-137 (1st week of April), whereas Bhagwa, Super Bhagwa and Ruby took more days to open its last flower on 3rd week of April. Duration from fruit set to maturity ranged from shortest 145 days to longest 170 days. The minimum duration from fruit set to fruit maturity was taken by the cultivar Mridula, whereas, Bhagwa took maximum duration from fruit set to maturity. The result is more or less in close conformity with the findings of Babu *et al.* (2017). The variation in maturity period of different pomegranate cultivars might be due to the genetic background and environmental conditions.

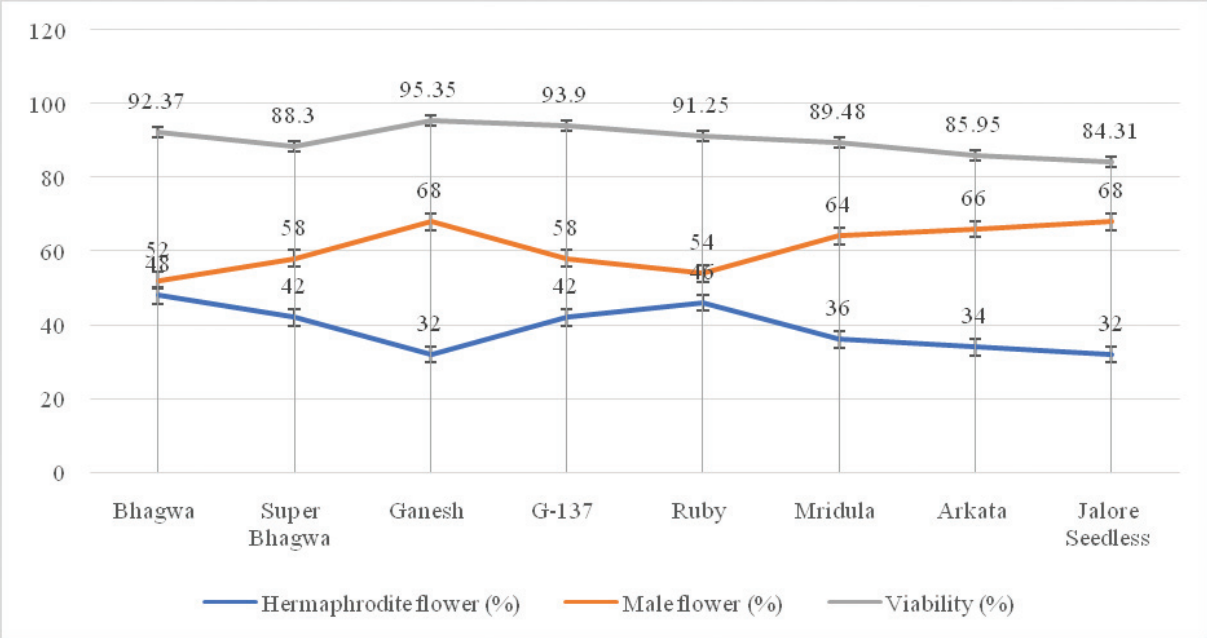

Fig. 1: Hermaphrodite and male flower, pollen viability percentage of eight cultivars of Pomegranate

Type of flowers and pollen viability of pomegranate flowers

The examination of data presented in Fig. 1 revels that the cultivar Bhagwa had the highest percentage of hermaphrodite flower (48 %), followed by Ruby (46 %), Super Bhagwa and G-137 (42.00 %) while in lowest Jalore Seedless and Ganesh (32 %). Whereas, cultivar Jalore Seedless and Ganesh (68 %) recorded highest percentage of male flower followed by Arkata (66 %), Mridula (64 %), while lowest in Bhagwa (52 %). The pollen viability of pomegranate flowers is an important phenomenon governing the fruit set to fruit harvesting. The pollen viability percentage ranged from 95.35 per cent being highest in cultivar Ganesh to lowest of 84.31 per cent in Jalore Seedless. Different dissimilarity was also reported by Babu *et al.* (2011). Pollen viability of most of the fruit crops is mostly genetic trait, as same variety under varied climatic conditions consistently recorded higher percentages of pollen viability. Different testing procedures and also time of bahar as well as time of collection of pollen done by different personnel may account for very minor discrepancies in pomegranate pollen viability.

Fruit yield and physical characteristics of fruit

Table 3 shows the fruit yield attributes, which depends on the number of fruits per plant, the fruit weight and other physical factors. Fruit yield showed significant variation among the eight cultivars evaluated. The highest yield was recorded in Super Bhagwa (15.7 kg/plant), followed by Ruby (12.7 kg/plant) and G-137 (12.2 kg/plant), whereas the lowest was recorded in Jalore Seedless (6.4 kg/plant). Similarly, the highest number of fruits per plant was recorded in Super Bhagwa (61.0) while, lowest number of fruits was found in Jalore Seedless (25.6). The highest fruit weight was recorded in Super Bhagwa (258.4 g), followed by Jalore Seedless (252.8 g), Bhagwa (247.0 g) and lowest fruit weight was found in Arkata (197.0 g). These finding are in close agreement with the results of Prasad *et al.* (2013). The variations in the yield attributes parameters in different cultivars might be due to genetic background, environmental condition and agronomical practices. The majority of pomegranate cultivars such as Ganesh, G-137, Ruby, Mridula and Arkata had round type of fruit shape, Bhagwa, Super Bhagwa and Jalore Seedless ovate type of fruit shape. The cultivar Bhagwa, Super Bhagwa, Ruby, Mridula and Arkata had red colour of fruit skin during time of harvesting,

Ganesh, G-137 and Jalore Seedless had yellowish fruit colour. The aril colour of Bhagwa and Super Bhagwa had red, Ganesh, G-137 and Ruby had pink, Mridula and Arkata had dark red, Jalore Seedless had white in colour. The peels and arils of pomegranate contain various types of flavonoids, anthocyanins, and tannins, which give them their distinctive colour and antioxidant properties (Zhao *et al.,* 2022). Ghosh *et al.* (2012) found that colour of aril was changed with time of maturity. It was pink when the fruits were harvested during May and became light red in June and red in July. Ranpise *et al*. (2014) found that in pomegranate arils anthocyanin content was generally lower in fruit harvested during June-July and higher in the fruit harvested during November-December. The decrease in anthocyanin content was possibly due to degradation of anthocyanins by high sunlight intensity observed during May-June.

Incidence of physiological disorder, insects and pests

Incidence of major physiological disorder, pest and disease determine the economic feasibility of a cultivar in the region. It was cleared from data presented in the table 4, showed that Jalore Seedless had highest incidence of fruit drop, medium incidence in Ganesh and Arkata, while lowest was in Bhagwa, Super Bhagwa, G-137, Ruby and Mridula. Majority of cultivars are also low with respect to incidence of fruit cracking observed during fruit growth and development period. The cultivars Bhagwa, Super Bhagwa, Ruby, Mridula, Arkata, Jalore Seedless recorded low incidence of fruit cracking, while Ganesh and G-137 recorded medium incidence of fruit cracking. The results obtained in this aspect was in conformity with the findings of Yuan *et al.* (2010), who also observed fruit cracking percentage to vary from 1.5 % to 45.9%. Different cultivars showed different susceptibility to fruit borer, the cultivar Bhagwa, Ruby, Mridula and Arkata recorded low incidence of fruit borer attack, while medium attack recorded in Super Bhagwa, Ganesh, G-137. All the cultivars were free from fruit sucking moths, bacterial blight and wilt during the period of investigation.

Quality characteristics of fruits

Based on the results on fruit quality parameters among the cultivars under investigation, it may be inferred from the study that the different cultivars showed a wide range of variability with respect to fruit quality attributes. Thus, an effective selection can be made based on quality characters for future improvement through a breeding programme. The data on fruit quality parameters (Table 5) showed that the highest total soluble solids content was found in cultivars Super Bhagwa and Ganesh (14.3 ^{0}Brix), followed by Jalore Seedless (13.2 ^{0}Brix), Ruby (12.7 ^{0}Brix), Bhagwa (12.4 ^{0}Brix) and Mridula (12.4 ^{0}Brix), while lowest in Arkata (11.6 ^{0}Brix). The studies are in confirmation with the early works made by Kumar *et al*. (2020) who had revealed that the Ganesh had maximum total soluble solids (14.71^{0}Brix) whereas, Patil *et al*. (2013) also found similar results and stated that the total soluble solids of Ganesh was higher than that of Arkata. The variations in total soluble solids content might be due to the occurrence of rainfall during fruit maturity which might have increased the moisture content of the fruit and dilution of carbohydrates. The acidity per cent was highest in Jalore Seedless (0.60 %), whereas, it was lower in G-137 (0.43 %) and Mridula (0.44 %). This type of variation has also been reported by Wani *et al.,* (2012). The variation of acidity might be due to different ripening stages and due to prevailing climatic conditions *i.e.* low temperature, lesser sunshine and high humidity. The TSS/acid ratio was highest in cultivar Ganesh (30.4), followed by Super Bhagwa (29.7), G-137 (28.1) and Mridula (28.1), while the lowest in Arkata (20.7). The total sugar content range between 8.33 in Arkata to 9.03 % in Ganesh. According to Bhat *et al*. (2019), the total sugar content of local Selection and G-137 varieties was 8.45% and 9.02%, respectively, under the sub-tropical conditions of Jammu. The highest vitamin-C content of 11.64 mg/100ml was recorded in cultivar Bhagwa which was statistically above all the cultivars. The minimum vitamin-C content was recorded as 10.0 mg/100ml in Ruby.

CONCLUSION

From the above findings, it is concluded that Super Bhagwa can be recommended for commercial cultivation under semi-arid climatic condition in Jhansi district of Utter Pradesh as it gave highest yield and also good quality fruit. Another cultivar that is suitable for cultivation is

Ruby, which has the second highest yield among the tested cultivars and produces fruits of good quality. From quality point of view, the cultivar Ganesh was found superior to others owing to its highest TSS: acid ratio.

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Assessment of pollen quality and quantity in white and black Turkish *Myrtus communis* L. accessions, through *in vitro* pollen germination under varied boric acid concentrations

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ABSTRACT

This study aimed to evaluate the pollen properties, pollen viability and germination with suitable germination medium and amount of pollen production in 6 Turkish myrtle genotypes. The genotypes were evaluated as their origin and fruit colours in terms of 3 different origins (Erdemli, Karaisali and Tarsus) and 2 fruit colours (black and white). Results showed that, pollen properties were affected from fruit colours and black genotypes generally showed higher pollen properties. The amount of pollen production of one flower was very high ranging from 2.5 to 4.5 million. Pollen viability and germination rates showed sufficient results for successful pollination in Turkish myrtle genotypes. Among pollen germination media, the best pollen germination rate was obtained from 50 ppm boric acid. Consequently, this study identified the pollen performance of white and black Turkish myrtle genotypes and suggested an optimal pollen germination medium for Myrtus communis which can be used in the future breeding programmes.

***Keywords*:** Anther, flower, Myrtaceae, pollen biology, pollination

INTRODUCTION

Myrtle (*Myrtus communis* L.) belongs to the family Myrtaceae and is one of the most significant species widely spread in the Mediterranean region, the Middle East and warmer regions of North America and Australia. In Turkey, it has been intensively grown naturally in the Mediterranean region especially near natural pine forests, particularly in the Taurus Mountains and riversides. The species are growing wild in warm and temperate regions of the Mediterranean basin, where it is well known for its medicinal and aromatic properties (Mulas *et al*., 1999; Yildirim *et al*., 2013). Essential oil from the leaves is used in the perfume and food industries (Lawrence, 1994; Boelens and Jimenez, 1992), while both leaves and berries are used to produce typical liqueurs (Mulas *et al*., 1999). Due to the success of myrtle liqueur and the number of processing industries, the demand for raw material has increased (Mulas and Fadda, 2004)

Myrtle has several genotypes with yellowish-white or bluish-black coloured fruits (San *et al.,* 2015; Simsek *et al*., 2020). Recent studies of myrtle have focused on the health functions of aromatic and medicinal plants, which have antioxidant, antimicrobial and mutagen properties due to the dietary intake of antioxidant compounds (Duh *et al.,* 1999; Yildirim *et al.,* 2013). Myrtle oil has a very extensive consumption as food, cosmetics, medicine, perfumery etc. (Jamoussi *et al.,* 2005, Mechchate *et al.,* 2022). In recent years as it has been understood as an important fruit, it gained importance and started to take place in markets and in herbalists (Montoro *et al.*, 2006; Alim, 2020). Due to high food value and wide usage of myrtle fruits and leaves, its production has been increased. Whether there is not a closed myrtle orchard in Türkiye, plants were started to plant as hedge plant in order to provide the demand of customers.

Myrtle flowers need pollination to set fruits and a successful pollination depends on quality of flowers especially in terms of pollen quality. Myrtle fruits has lots of seeds (Mulas and Fadda, 2004). Whether this seed existence disturbs customers, the studies for decreasing seed number caused excessive fruit drops (Gonzalez-Varo *et al.,* 2009, Alim, 2020). This shows that, seed formation after

successful pollination and fertilization is essential for adequate fruit set in myrtle.

Myrtle buds develop from the leaf axillae on young shoots as single flowers (Mulas and Fadda, 2004). It has a 1-2,5 cm green or red peduncle generally linked with fruit colour. Myrtle flowers are hermaphroditic with both stamens and pistil are at the same flower (Fig. 1a). Flowers comprise of 5 green sepals, 5 white or pinky petals, about 100-150 stamens with long filaments and a little anther. Pollens are triangular in shape and has tricolporate apertures with a diameter about 8-15 mm (Fig. 1b). The flower has a single pistil at the middle of the flower. It is embedded into the receptacle showing an inferior ovary type with 3 syncarpous carpels with axile plasentation of about 25-50 ovules (Mulas and Fadda, 2004). Flowers secrete lots of nectar at the bottom of the anthers and originally near from the apex of the anther in order to attract pollinator insects for a better pollination (Ciccarelli *et al*., 2008).

Recent studies have been showed that, myrtle pollen viability is very high immediately after anthesis. Along with the end of the first day, pollen viability declines and 36 hours after anthesis almost all grains become unviable. At the same time, while pollen viability remains high for several hours at high temperature and dry conditions, it rapidly decreases at high humidity (Aronne, 1999). So, myrtle pollen should be transferred to the stigma as soon as possible by pollinator insects which indicates the importance of pollen limitation and pollen viability one more time. Although myrtle is a self-compatible species and needs pollinators to increase fruit set, fruit size and quality increases with cross pollination showing that the pollen quality has a great importance for myrtle fruit formation (Aizen and Harder, 2007; Gonzalez-Varo *et al.,* 2009).

Considering the effects of pollen importance in myrtle flowers, this study aims to evaluate the pollen quality in terms of pollen viability and pollen germination rates, suitable *in vitro* pollen germination medium and *in vitro* pollen tube growth in 3 black and 3 white Turkish myrtle genotypes. At the same time, the amount of pollen production was also determined for relevant genotypes.

MATERIALS AND METHODS

This study was conducted in 2023 flowering season of 6 Turkish myrtle genotypes. The genotypes were selected for their superior qualities from Mediterranean region of Türkiye (Simsek *et al.,* 2020). The selected genotypes were from 3 different regions of Türkiye (Erdemli, Karaisali and Tarsus) due to their fruit colours (white and black) (Table 1). The selected genotypes were planted in research field of Cukurova University in 2020 at 3x2 m spacing. Plants were 3 years old, drip irrigated and all necessary cultural practices were followed uniformly in the research field.

In this study the amount of pollen production and pollen homogeneity, *in vitro* pollen viability and germination rates, suitable *in vitro* pollen germination medium and *in vitro* pollen tube growth rate parameters were evaluated. For this purpose, at least 100 unopened flowers were collected one day before anthesis from 5 trees of each genotype and immediately taken to the Cytology Laboratory in Cukurova University.

Amount of pollen production and pollen homogeneity

For determining amount of pollen production and homogeneity tests, 30 flowers were separated to 3 groups and anthers of each group (10 flowers for each) were taken to plastic containers. Prepared samples were left to dry for at least 15 days. Pollen production tests were made by hemacytometric method and counting slides were prepared according to Eti (1990) with 4 ml of sterile water (Fig. 2a). Then, the samples were analysed with Olympus BX 51 light microscope and micrographs were taken with DP72 camera with a specified area on the micrograph (Fig. 2b). Following preparation, the quantity of pollen within the specified area was tallied, and subsequent calculations were performed, incorporating a modification based on Eti (1990). While calculation, the counting place volume was found with multiplication of the created area and depth of hemacytometric slide. The amount of pollen at calculated volume was then rated to whole container volume to find "amount of pollen for 10 flowers". Then the "pollen number per flower" was found by dividing each data to 10. At least 12 areas were counted for each replication and the average data were used for calculation.

The "normally developed pollen ratio" was also determined in each micrograph by counting and rating normally developed pollen to total pollen in created volumes according to Anvari (1977).

In vitro pollen viability rate

In order to obtain pollen for viability tests, anthers from 6 genotypes of myrtus were collected randomly before anthesis. Anthers of the collected flowers were separated and placed in room temperature until dehiscence for about one night (Mulas and Fadda, 2004). After pollen dehiscence, pollen viability rates were determined using 1% TTC test (2,3,5 Triphenyl tetrazolium chlorid). TTC was prepared and evaluated according to Norton (1966). For each genotype 3 slide replication were prepared and at least 100 pollen grains were counted from each replication. Pollen counting were made by Olympus BX51 microscope. While counting, red pollens were considered as "viable", light pinkies as "semi viable" and uncoloured pollens as "non-viable" (Fig. 3a). Then the pollen viability rate was calculated according to Norton (1966) as the ratio of "sum of total viable and half of semi viable pollens" to "total pollen number".

Suitable *in vitro* pollen germination medium and *in vitro* pollen germination rate

In order to determine pollen germination data, same pollens were used that was prepared for pollen viability tests. The suitable *in vitro* pollen germination tests were all performed in a basic media, which consisted of 1% agar, 100 ml distilled water and 15% sucrose (Mulas and Fadda, 2004) with single factor experiment of boric acid (H_3BO_3) in concentration of 0, 50 and 100 ppm.

Media and germination petri dishes were prepared according to Karabiyik and Eti (2016). Pollen germination ratio for each medium was determined after 24 hours. The pollen grains were considered to be germinated when the pollen tube length was greater than the diameter of the pollen grain (Fig. 3b). At least 100 pollen grains were counted for each replication.

The *in vitro* pollen tube growth rate was also determined after 2, 4 and 10 hours of preparation in order to set out the difference at emergence and elongation of pollen tubes for each media and genotype. In order to measure pollen tube length in specified hours, little segments were taken from prepared media between slide and cover glass and immediately taken to -20^{0}C to stop growth and fix the media samples filled with pollen. Experiments were prepared as 3 replications for pollen tube measuring tests. The *in vitro* pollen tube growth was determined by measuring pollen tube length with Olympus BX 51 microscope equipped with a DP 72 digital camera (Fig. 3c). At least 30 pollen tubes were measured for each media, each hour and each replication. By this way, the fastest pollen germinating media could be determined for myrtle genotypes.

Statistical analysis

All data analysis was performed using JMP 13 statistical software. The effects of the treatments were analysed using one way anova analysis of variance. A P value of $< 0,05$ was considered to be significant. The statistical analysis was conducted in terms of the origins and fruit colour of genotypes. By this way, 2 factorial randomised design has been used for analysing pollen production, normally developed pollen and pollen viability data. In the other part of our experiment, the pollen germination rate was analysed by 3 factorial randomised design as genotype origin, colour and boron concentrations. Percentages were analysed after arc-sin transformation.

RESULTS AND DISCUSSION

Amount of pollen production and normally developed pollen rate

Amount of pollen production for 6 Turkish myrtle genotypes were given in Table 2 in terms of origins and fruit colours of the genotypes. The table shows that amount of pollen production in one flower significantly influenced by fruit colour and origin x colour interaction while differences between genotypes did not find to be important. The pollen production in each flower was very high ranging between 2 657 493 (Karaisali-White_KB) and 4 885 013 (Karaisali-Black_KS) pollens per flower and the average pollen production was definitely higher in black genotypes (3 843 840) than white genotypes (3 056 213). In terms of genotype origin averages, the highest pollen production was detected in Karaisali genotypes (3

Table 1: Origins and fruit colours of used genotypes

Genotype name	Genotype code	Origin	Fruit Colour
Erdemli beyazi	EB	Erdemli/Mersin	White fruited
Erdemli siyahi	ES	Erdemli/Mersin	Black fruited
Karaisali beyazi	KB	Karaisali/Adana	White fruited
Karaisali siyahi	KS	Karaisali/Adana	Black fruited
Tarsus beyazi	TB	Tarsus/Mersin	White fruited
Tarsus siyahi	TS	Tarsus/Mersin	Black fruited

Table 2: Amount of pollen production per flower in 6 Turkish myrtle genotypes in terms of their origins and fruit colours (pollen/anther)

Origins	Fruit Colours		Origins Average
	White	Black	
Erdemli	3 295 146 b[1]	3 412 266 b	3 353 706
Karaisali	2 657 493 b	4 885 013 a	3 771 253
Tarsus	3 216 000 b	3 234 240 b	3 225 120
Colours Average	3 056 213 B	3 843 840 A	

LSD_{origin}***: N.S.[2] LSD_{colour}**: 586 814,2 $LSD_{ori\ x\ col}$***:1 016 392,0

[1] Differences between averages showed by different letters are statistically significant

N.S. means not-significant; ** means p<0,01; *** means p<0,001.

Table 3: Normally developed pollen ratios in 6 Turkish myrtle genotypes in terms of their origins and fruit colours (%)[1]

Origins	Fruit Colours		Origins Average
	White	Black	
Erdemli	93.17	93.97	93.57 A[2]
Karaisali	89.35	88.02	88.68 B
Tarsus	93.95	93.75	93.85 A
Colours Average	92.16	91.91	

LSD_{origin}***: 1,985 LSD_{colour}: N.S. $LSD_{ori\ x\ col}$: N.S.

[1] Statistical analysis were made after arc-sin transformation.

[2] Differences between averages showed by different letters are statistically significant

N.S. means not-significant; *** means p<0,001.

Table 4: Pollen viability levels in 6 Turkish myrtle genotypes in terms of their origins and fruit colours (%)[1]

Origins	Fruit Colours		Origins Average
	White	Black	
Erdemli	42.84 cd[2]	85.46 a	64.15 A
Karaisali	51.99 b	35.11 d	43.55 B
Tarsus	50.30 bc	44.63 bc	47.47 C
Colours Average	48.38 B	55.07 A	

LSD_{origin}***: 3,605 LSD_{colour}**: 2,943 $LSD_{ori\ x\ col}$***: 5,098

[1] Statistical analysis were made after arc-sin transformation.

[2] Differences between averages showed by different letters are statistically significant

N.S. means not-significant; ** means p<0,01; *** means p<0,001.

Table 5: Pollen germination levels in 6 Turkish myrtle genotypes in terms of their origins, fruit colours and Boron concentrations (%)[1]

Origins	Boron Concent.	Fruit Colours		Origin x Boron	Origins Average
		White	Black		
Erdemli	0 ppm	26,20	46,34	36,27	41,58
	50 ppm	27,99	63,49	41,58	
	100 ppm	19,68	65,79	46,89	
Origin x Colour		24,62 b[2]	58,54 a		
Karaisali	0 ppm	31,00	68,07	49,53	42,53
	50 ppm	37,45	37,27	37,36	
	100 ppm	26,04	55,37	40,71	
Origin x Colour		31,50 b	53,57 a		
Tarsus	0 ppm	47,17	43,57	45,37	51,34
	50 ppm	51,15	67,14	59,15	
	100 ppm	50,54	48,49	49,52	
Origin x Colour		49,62 a	53,07 a		
Colours Average		35,25 B	55,06 A		

LSD_{origin}:N.S. LSD_{colour}***: 4,919 LSD_{boron}: N.S. $LSD_{ori\ x\ bor}$: N.S. $LSD_{ori\ x\ col}$***: 8,521 $LSD_{orixcolxbor}$: N.S.

[1] Statistical analysis were made after arc-sin transformation.

[2] Differences between averages showed by different letters are statistically significant

N.S. means not-significant; *** means p<0,001.

771 253), followed by Erdemli (3 535 706) and Tarsus (3 225 120) genotypes.

Pollen production is an important data for seeded cultivars as it guarantees the pollen meeting probability with the stigma. In this study, amount of pollen production in one myrtle flower was very high at about 2.5 millions and 4.5 millions. However, myrtle flowers have a lot of anthers between 120-150 (Mulas and Fadda, 2004) and this means each anther has at about 15 000 – 30 000 pollens. To the best of our knowledge, there is not any data for amount of pollen production in myrtle flowers and this report is the first for pollen production for *Myrtus communis*. However, the data was coherent with other species like citrus (Karabiyik and Eti, 2019), carob (Eti, 1990), strawberry (Karabiyik *et al*., 2016), pecan (Karabýyýk and Eti, 2018), watermelon (Adiguzel *et al.,* 2022), loquat (Karabiyik and Eti, 2015) etc. in terms of amount of pollen produced in one anther.

Nevertheless, pollen production for myrtle genotypes should be higher than most of the species. Because, Beardsell *et al.* (1989) have been stated that members of Myrtaceae has secretes under anthers of the stamens called anther glands that helps flowers to attract pollinator insects, especially the bees. The authors have been reported that this secrete mixes with pollen which acts as a food source for insects (Ciccarelli *et al*., 2008). This structure of flowers causes lots of pollen loses, so the high pollen production roles out these handicap.

Normally developed pollen ratios were given in Table 3. The data showed that normally developed pollen which shows pollen homogeneity level is generally high and only origin of the genotype affects normally developed pollen ratio. The highest ratio was obtained from Erdemli (93.57%) and Tarsus (93.85%) genotypes while Karaisali genotypes (88.68%) had the lowest homogeneity level. Fruit colour did not differ significantly and white genotypes had an average of 92.16% while black genotypes has an average of 91.91% pollen homogeneity level.

Besides the high pollen production of a genotype, it is important to have a genotype high normally developed pollen ratio. Anvari (1977) have been stated that the undeveloped or abnormal pollens, which shows diversions from normal shape and size, are unlikely to germinate. So, if the normal rate of pollen ratio approach to 100%, the

Fig. 1: Flower (a) and pollen (b) of myrtle

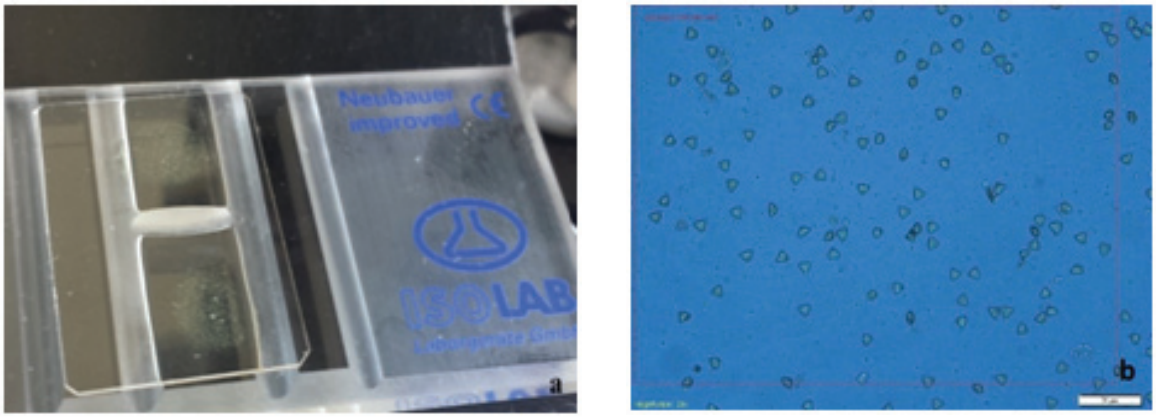

Fig. 2: The hemacytometric slide (a) and the pollen counting area (b)

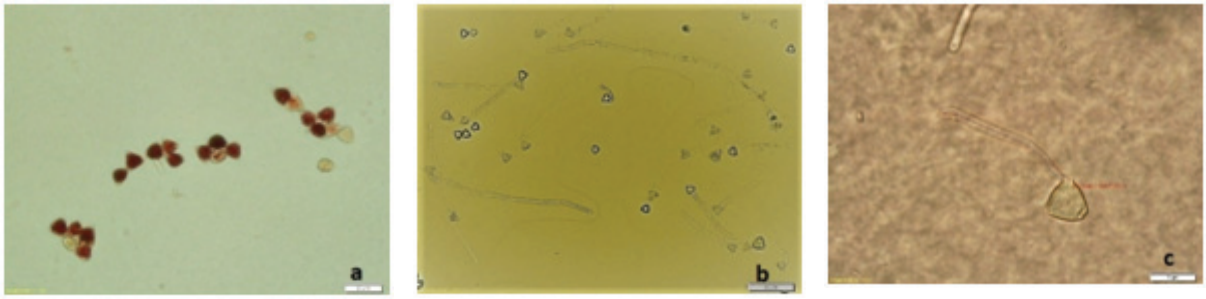

Fig. 3: a. pollen viability test, b. germinated pollens, c. Pollen tube elongation

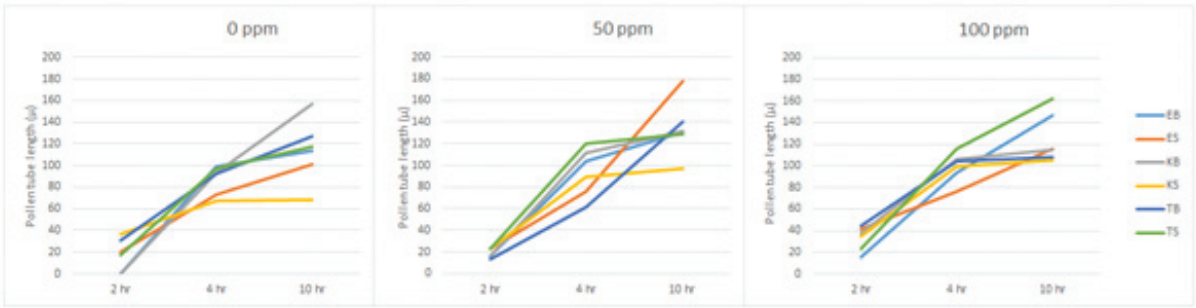

Fig. 4:Pollen tube growth rate of Turkish myrtle accessions in different *in vitro* pollen germination media.

pollination potential of genotype will increase in the same rate. The results showed that myrtle pollens have adequate pollen homogeneity and pollen production level for successful pollination.

In vitro pollen viability rate

In vitro pollen viability have been detected by TTC method and the results were shown in Table 4. The statistical analysis showed that origin, colour and origin x colour interaction significantly affected pollen viability rate of used myrtle genotypes. Pollen viability rate was highest in Erdemli originated genotypes with 64.15% while it was 47.47% in Tarsus and 43.55% in Karaisali. In terms of fruit colours, average of black cultivars showed better results (55.07%) than white genotypes (48.38%). The origin x colour interaction data had a wide range as much as 35.11% (Karaisali-Black_KS) and 85.46% (Erdemli-Black_ES).

As Mulas and Fadda (2004) have been reported, pollen viability rate was not differed between freshly opened flowers and dried flowers and the pollen viability rates of 10 myrtus genotypes were higher than our results that differed between 85% and 95%. It is thought that this difference is originated from differences in genotype and ecological conditions (Eti, 1991).

In vitro pollen germination rates and suitable pollen germination media

Table 4 shows *in vitro* pollen germination levels of 6 Turkish myrtle genotypes in terms of their origin and fruit colours as well as different germination media. In the study, different concentrations of boric acid (H_3BO_3) were tested for optimization of *in vitro* pollen germination medium. The results showed that boric acid concentration has no importance on pollen germination ratio while fruit colour and origin x colour interaction (genotype effect) showed statistically important values.

In terms of fruit colours, pollen germination levels were higher in black genotypes (55.06%) than white genotypes (35.25%), like pollen production and pollen viability levels. Origin x colour interaction that shows the germination levels of each genotype were highest in Erdemli-Black (ES) which was followed by Karaisali-black (KS) and Tarsus-black (TS) and the lowest from Erdemli-white (EB).

In general, pollen germination level gives the best results for the pollen quality of a genotype. Although genotypes should have a good pollen production and pollen viability percentage; pollen germination, pollen tube emergence and pollen tube growth has such a degree importance for pollination and especially for fertilization level. In a recent study, Mulas and Fadda (2004) tested 10 myrtle genotypes in terms of pollen germination levels with 10, 15 and 20% sucrose concentrations and found the rates of 6 genotypes as 70 to 85%; other 3 genotypes differing from 25 to 40% and one genotype with 0.7%. Authors reported the best media as 10 and 15% sucrose concentrations for most of the genotypes. The 15% sucrose concentration that was used in this study was suitable for myrtle pollen germination. Despite the statistical analysis has no significant importance, a 50 ppm Boric acid addition to germination media could increase pollen germination rate for most of the genotypes.

Pollen germination assays frequently require optimisation as well as being time consuming and difficult to reproduce (Rathod *et al*., 2018). The germination media should imitate stigma surface for the truest data. It has been reported that, boric acid content in the pollen is insufficient (O'Kelley, 1957) and Boron in the stigma and style is required to compensate for this; thus a certain amount of boric acid needs to be added to *in vitro* cultures (Johri and Vasil, 1961; Luo *et al*., 2020). There was not any test for additional substances for *Myrtus communis*. In general, 10 ppm (0.001%) or higher concentrations of Boric acid is toxic to plant growth while pollen grains can tolerate concentrations up to 1 200 ppm (0.12%) and optimum results were obtained between 10 and 150 ppm concentrations for pollen germination for most of the species (Johri and Vassil, 1961).

In this study, the results of viability and germination rates were in parallel except from Karaisali-black (KS). The chemical TTC (Tetrazolium) forms a red coloured compound formazan by H transfer reactions catalysed by the enzyme dehydrogenases (Norton, 1966). This results shows that, only TTC test might not give the real performance for some myrtle genotypes due to its possible insufficient reaction capacity with H^+ ions. So, different viability tests

accomplished with *in vitro* pollen germination could be used for better results in future studies.

Pollen tube growth rate was also evaluated in this study and results were given in Fig. 4. The pollen tube growth rate was faster in 50 ppm boric acid concentrations which means pollen tubes will emerge and grow faster in a boron fertilized genotype. In 0 ppm Boron (Control) especially KB and EB did not emerge any pollen tubes in first 2 hours while KS and ES could not show their real performance of pollen tube acceleration. At the same time, 100 ppm Boric acid also increases the first emergence and formed faster pollen tubes.

It is known that boron accelerates pollen tube by its promoting absorption of sugars and their metabolism by forming sugar-borate complexes, increased oxygen uptake and is involved in the synthesis of pectic materials required for the wall of the actively elongating pollen tube (Johri and Vasil, 1961). As pollen did not have sufficient boron content and stigma and style will compensate boron requirement, addition of boric acid to the germination medium simulates this situation and shows the real potential of performance of pollen tube elongation (Johri and Vasil, 1961; Jayaprakash *et al*., 2018; Luo *et al*., 2020). When Table 5 and Figure 4 were evaluated together boric acid was thought to be accelerated the first pollen tube emergence and pollen tube elongation ratio but not have any effect on pollen germination ratio.

CONCLUSION

Pollen quality and quantity is vital for understanding fertility and incompatibility, pollen-pistil interaction, breeding, crop improvement and seed industry. For successful pollination, the insight knowledge of pollen biology including pollen viability, germination and amount of pollen production is required for reasonable approaches to increase productivity. This study evaluates amount of pollen production, normally developed pollen ratio, pollen viability and germination levels with suitable pollen germination medium as well as pollen germination rates of 6 Turkish myrtle genotypes in terms of their origins and fruit colours. In *Myrtus communis*, amount of pollen production per flower was found to be very high as much as 2.5-4.5 million. The high pollen production is thought to be related with anther secretes that will cause pollen losses. Normally developed pollen ratio was also so high and sufficient for successful pollination. The pollen viability and germination rates were sufficient for most of the genotypes and pollen tube elongation was faster in the media containing 50 ppm boric acid with 15% sucrose and 1% agar solution which is thought to be the best media for *Myrtus communis* pollen germination tests. In general, all pollen properties were higher in black genotypes than the white genotypes.

In future studies, pollination capacity of white cultivars should be experienced for it's possible stimulatory effect because of their lower pollen characteristics. So, a pollination study should be planned for potential seed number decreasing effect of white genotypes.

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Studies on morpho-biochemical changes of longan [*Euphoria longana* (Lour) Steud.] fruit during fruit growth and development for determination of maturity

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ABSTRACT

Longan fruit is used as fresh as well as preserved as canned due to its sweet juicy and flavoured aril and becomes available when availability of litchi is over. It is a non-climacteric fruit and continues to ripen being attached to the plant. The fully ripened, freshly harvested fruit has a bark like shell, thin and firm, making the fruit easy to peel by squeezing the pulp to out. The seed is small, round, hard and of an enamel like, lacquered black. However, the proper maturity stage is yet to be known as scientific findings on physical and biochemical maturity indices of longan is scare. In the present experiment, the pattern of sequential development and changes in physio-chemical properties of longan fruit after fruit set were investigated in ten plants of 10 to 15 years age group. The experiment was laid under Randomized Block Design considering the stages of harvest (days after fruit set) as treatments and all ten plants as separate replication during the year 2022. Fruit growth based on fruit length, fruit diameter, seed diameter, aril thickness, weight of fruit, weight of seed, followed a smooth sigmoidal curve during the fruit growth and development. A similar trend of changes has also been observed with respect to total soluble solids (TSS), acidity, total sugar and reducing sugar. Both the fruit morphological characters as well as the quality parameters reached to the steady state in between 114 to 128 days after fruit set. Based on the attainment of fruit size, aril quantity and quality aspects, it can be concluded that the optimum stage for harvest of longan is 114-128 days after fruit set.

***Keywords*:** Fruit growth, longan, maturity, morphological changes, quality.

INTRODUCTION

Longan [*Euphoria longana* (Lour) Steud.] is an evergreen subtropical fruit tree species and originated from Southern China and came to India in the year 1978 (Morton *et al*., 1987). This fruit comes under the order Sapindales and family Sapindaceae (soapberry family). In India it is cultivated in very limited pockets of West Bengal and North Bihar. Thus it is considered as one of the minor as well as underutilized fruit in India like other crops (Hazarika and Lalruatsangi, 2016).

The fruits of longan are preferred and consumed as fresh due to its juicy refreshing taste and flavour. Fleshy aril is the edible portion of this fruit which is rich in sugar, vitamins (B-complex and C), minerals (iron, phosphorus, calcium) as well as various antioxidants and bioactive compounds which are reported to be antitumor, antidiabatic, antiallergic activities (Kumar *et al*., 2023, Nath *et al*., 2018). This fruit is considered as one of the important among the all minor and underutilized fruits like jamun, phalsa, Indian olive, latka etc. (Prasad *et al*., 2017; Halder *et al*., 2023). Along with the other underutilized fruit crop species, longan was also studied for its nutritional profiling by many scientists (Wang *et al*., 2011, Marisa, 2006, Jiang *et al*., 2002).

Longan trees usually produce shoot once in a year, although it can produce more than one shoot during summer and autumn that converts into 10-45 cm long panicle once during end of winter. The small flowers contain 5-6 yellow or brown petals in both unisexual and bisexual (hermaphroditic) flowers (Pham *et al*., 2016). The female flowers bear a carpellate ovary and the heart shaped, yellowish or light brown coloured, single seeded

fruits are small drupe of 20-37 mm in diameter and 5-20g in weight. Basically, the seed is orbicular with black or brown colour with a rounded white spot that has the appearance of a dragon's eye (Sun *et al*., 2010). Panicles of longan plants bear upto 350 fruits and take about 140-190 days as per the report of Crane *et al*. (2005). However, the proper maturity stage is not yet reported as scientific findings on the basis of fruit physical and biochemical maturity parameters. In this context, the present experiment has been conducted to study the pattern of sequential development and changes in physio-chemical properties of longan fruit to assess the proper maturity stage.

MATERIALS AND METHODS

The present experiment was conducted during the year 2021 and 2022 selecting ten longan plants under age group of fifteen to twenty years at various village locations (viz. Bahadurpur, Binuria, Raipur, Deuli, Supur, Ballavpur, Baganpara, Lohagar, Surul and Jambuni) of Bolpur Sriniketan Block, Birbhum, West Bengal. Twenty five branches of each plants bearing panicles have been randomly selected and tagged before the anthesis for recording various observations on fruit growth and development. Longan plants started bearing during mid-February to mid-March. After tagging of selected branchlets (panicles) recording of observations were started from the fifty one (51) days after fruit set at seven days interval upto thirteen different stages of fruit growth was taken for identifying the optimum maturity stage of longan. Thirteen different stages of fruit growth were considered as different treatments and ten different plants were considered as replications following Randomized Block Design with thirteen treatments and ten replications. Thirteen (13) different stages of fruit growth that considered were fifty one (51), fifty eight (58), sixty five (65), seventy two (72), seventy nine (79), eighty six (86), ninety three (93), hundred (100), hundred seven(107), hundred fourteen (114), hundred twenty one (121), hundred twenty eight (128) and hundred thirty five(135) days after fruit set. As the aril development of longan fruits was started only after 50 days of fruit set (Prasad *et al*., 2017), thus all the observations has been recorded only after 50 days of fruit set. Various observations on fruit physical parameters like average length of the fruit, diameter of the fruit, seed diameter, aril thickness, weight of the fruit (g) and seed weight (g) have been recorded as per following procedure:

Fruit physical parameters: The length of ten fruits from each replication was measured from the distal end to the proximal end of the fruit with the help of a digital vernier caliper (Mitutoyo, Japan) and the mean was worked out and expressed in millimetres. The fruit diameter of ten fruits per replication was recorded at the widest point of the fruit using a a digital vernier caliper and the mean value was expressed in millimetres. Data on seed diameter was recorded from three randomly selected seeds from each replication using a vernier caliper at the widest point and their mean value was expressed in millimetres. Aril thickness of ten fruits from each replication was measured with the help of a digital vernier caliper and the mean was calculated out and expressed in millimetres. Ten randomly selected fruits from each replication were used for measuring the fruit weight. The weight was measured on a pan balance and the average fruit weight was calculated and expressed in grams. Seed weight was measured on a simple pan balance and their mean weight was recorded and expressed in grams.

Fruit biochemical parameters: Total Soluble Solids of the juice of ten longan fruits from each treatment were recorded using a pocket digital refractometer (Atago, Model PAL-1, Japan) and expressed in °Brix. The titration method using the fruit juice against 0.1 N NaOH and phenolphthalein as indicator were used to determine the titrable acidity (Rangana, 1986). Total sugars was determined following the method as described by Rangana, (1986) by hydrolysis of juice with 1 or 2 drops of concentrated hydrochloric acid and neutralized by adding 1N sodium hydroxide solution followed by titration against standard Fehling's solution mixture A and B (1:1) using methylene blue indicator in a heated environment until brick red colour precipitated. Reducing sugars were determined by adopting the method given by Lane and Eynon (1923) taking prepared sample up to zero, and then it was titrated against combination of Fehling's solutions in a heated environment until brick red colour precipitated.

Statistical analysis of the data on different observations has been carried out using SPSS

(Version 18.0.2) considering thirteen different stages of fruit growth as treatments and ten different plants as replications. The analysis of observations of both the years (i.e. 2021 and 2022) is cited in the tables separately as well as pooled value for easy comprehension.

RESULTS AND DISCUSSION

The statistical analysis of data pertaining to the fruit physical parameters and biochemical parameters has been presented in Table 1 and 2 as separate analyses of both the years as well as pooled value for all the observations. However, after homogeneity test of data on all parameters, it was clear that both the years as well as pooled data has shown similar trend of variation. In the present experiment the observations on different physical and quality aspects of longan fruits has been recorded at thirteen different stages of fruit growth according to days after fruit set. The results on the basis of pooled value showed the significant variation in physical and biochemical parameters of longan fruits under different stages of fruit growth as follows:

Fruit length: Longan fruits have shown double sigmoid growth pattern in both the years as well as in pooled value, with respect to fruit length, as presented in Table 1 and Fig. 1. It has started to increase from 8.93mm at 51 days after fruit set and reached a steady state at 121 days after fruit set as 20.37mm as observed in pooled value. Although, at steady state the fruit attained a length of 20.82 mm at 135 days after fruit set after passing through double sigmoid pattern.

Fruit diameter: In the present study the changes in diameter of longan fruits in both the years as well as in pooled value have been presented in Table 1 and Fig. 1. The perusal of data of pooled of two years on fruit diameter shows a gradual increase starting from first day of observation (i.e. 51 days after fruit set) as 8.47mm and increased upto 19.93mm as on 121 days after fruit set after reaching at the steady state. Thus the growth pattern of the longan fruit with respect to fruit diameter was also double sigmoid.

Seed diameter: Critical review of the data on seed diameter of longan fruits in both the years as well as in pooled value (Table 1 and Fig. 1) revealed that it changed significantly with advancement of fruit growth and maturity. It is evident from the data of pooled value of both the years, at the initial stage of observation at 51 days after fruit set, the seed diameter was recorded as 2.01mm which ultimately attained the final plateau on 128 days after fruit set as 13.85mm. Although a little shrinkage of seed size was observed which was statistically non-significant.

Weight of fruit: With regard to the average fruit weight of longan (Table 1) with advancement of fruit growth, development and maturity in both the years as well as in pooled value, again double sigmoid curve have been observed (in pooled value) starting from 0.35g at 51 days after fruit set and attained the steady state of gaining weight at 114 days after fruit set as 5.08g which ultimately reached to 5.28g as on 135 days after fruit set, but such increment was within the steady state (i.e. the change was statistically non significant).

Seed weight: In the present experiment the statistical analysis of data on seed weight of longan fruits (Table 1) in both the years as well as in pooled value have shown increasing trend as starting from (in pooled value) 0.04g as on 51 days after fruit set to 1.63g as on 107 days after fruit set. The seed weight has been increased upto 1.80g which was non-significant. However, the seed weight of the longan fruit has shown double sigmoid growing habit.

Total Soluble Solids (TSS): The perusal of statistically analyzed data on changes in total soluble solids of longan fruit juice in the 2021 and 2022 as well as pooled value have directed the increasing trend in two phases (Table 2 and Fig. 2). It is clear from the pooled data of both the years that the TSS was started to increase from 3.51^0Brix at 51 days after fruit set in the first phase to 8.81^0Brix at 93 days after fruit set. Again in second phase the TSS of the longan fruits has increased from 10.22^0Brix at 100 days after fruit set to 16.29^0Brix at 135 days after fruit set. However, the highest TSS of longan fruits (16.46^0Brix) was observed at 128 days after fruit set after which it was gradually decreased.

Acidity: The observations on acidity of juice of longan fruits (Table 2) have increased significantly with the advancement of fruit growth and maturity as observed in both the years as well

Table 1: Changes in fruit physical parameters of longan with advancement of fruit growth towards maturity.

Days after fruit set	Fruit length (mm)			Fruit diameter (mm)			Seed diameter (mm)			Weight of fruit (g)			Seed weight (g)		
	2021	**2022**	**Pooled**	**2021**	**2022**	**Pooled**	**2021**	**2022**	**Pooled**	**2021**	**2022**	**Pooled**	**2021**	**2022**	**Pooled**
51 days	8.46	9.40	8.93	8.29	8.65	8.47	1.93	2.09	2.01	0.31	0.39	0.35	0.03	0.05	0.04
58 days	12.05	11.79	11.92	10.84	10.71	10.77	4.05	3.93	3.99	0.79	0.70	0.74	0.18	0.12	0.15
65 days	12.42	12.01	12.21	11.69	11.39	11.54	6.52	6.18	6.35	1.02	0.92	0.97	0.19	0.15	0.17
72 days	12.78	13.12	12.95	12.68	13.02	12.85	7.03	7.39	7.21	1.46	1.68	1.57	0.24	0.32	0.28
79 days	14.65	15.07	14.86	14.35	14.89	14.62	9.20	9.70	9.45	1.98	2.20	2.09	0.55	0.65	0.60
86 days	15.94	15.36	15.65	15.14	14.71	14.92	10.83	10.47	10.65	2.33	2.15	2.24	0.85	0.77	0.81
93 days	16.72	16.06	16.39	15.52	15.20	15.36	11.10	10.84	10.97	3.08	2.86	2.97	1.28	1.23	1.26
100 days	17.63	18.19	17.91	17.02	17.34	17.18	11.13	11.39	11.26	3.35	3.51	3.43	1.28	1.33	1.30
107 days	18.35	18.07	18.21	17.90	17.52	17.71	11.69	11.47	11.58	4.26	4.08	4.17	1.66	1.60	1.63
114 days	19.24	19.78	19.51	18.85	19.26	19.05	11.72	12.14	11.93	5.01	5.15	5.08	1.66	1.69	1.67
121 days	20.03	20.71	20.37	19.72	20.14	19.93	11.76	12.02	11.89	5.02	5.26	5.14	1.70	1.76	1.73
128 days	20.47	20.99	20.73	20.03	20.41	20.22	13.69	14.01	13.85	5.17	5.39	5.28	1.74	1.86	1.80
135 days	21.02	20.62	20.82	20.41	20.09	20.25	13.92	13.76	13.84	5.34	5.22	5.28	1.84	1.76	1.80
SE±m	0.21	0.23	**0.24**	0.21	0.18	**0.17**	0.23	0.19	**0.17**	0.13	0.14	**0.11**	0.07	0.08	**0.06**
CD 5%	0.66	0.71	**0.75**	0.62	0.55	**0.54**	0.69	0.58	**0.52**	0.39	0.41	**0.32**	0.22	0.24	**0.20**

Table 2: Changes in biochemical parameters of longan with advancement of fruit growth towards maturity.

Days after fruit set	TSS (^{0}Brix)			Acidity (%)			TSS: Acidity (%)			Total Sugar			Reducing Sugar (%)		
	2021	**2022**	**Pooled**	**2021**	**2022**	**Pooled**	**2021**	**2022**	**Pooled**	**2021**	**2022**	**Pooled**	**2021**	**2022**	**Pooled**
51 days	3.43	3.59	3.51	0.015	0.017	0.016	228.66	211.17	219.37	5.01	5.47	5.24	3.29	3.67	3.48
58 days	4.06	3.98	4.02	0.019	0.018	0.017	213.68	259.25	236.47	7.43	7.31	7.37	4.58	3.94	4.26
65 days	4.71	4.81	4.76	0.015	0.019	0.017	314.02	246.22	280.12	8.12	8.41	8.26	4.06	4.28	4.17
72 days	6.60	6.44	6.52	0.019	0.017	0.018	388.23	336.25	362.24	9.94	9.11	9.52	5.41	5.17	5.29
79 days	7.92	8.14	8.03	0.019	0.021	0.020	416.84	356.31	386.52	11.46	12.48	11.97	6.22	6.52	6.37
86 days	8.35	8.21	8.28	0.022	0.020	0.021	379.54	409.06	394.28	13.64	13.54	13.59	7.05	6.83	6.94
93 days	8.70	8.92	8.81	0.022	0.024	0.023	395.45	408.63	402.04	13.78	1.92	13.85	7.05	7.51	7.28
100 days	10.11	10.33	10.22	0.022	0.024	0.023	459.54	429.16	444.34	14.36	15.02	14.69	8.17	8.45	8.31
107 days	11.37	11.19	11.28	0.025	0.024	0.024	454.82	485.26	470.04	15.03	14.43	14.73	9.42	9.04	9.23
114 days	12.16	12.34	12.25	0.025	0.024	0.025	486.43	493.85	490.14	15.20	15.36	15.28	10.59	11.09	10.84
121 days	15.64	16.08	15.86	0.025	0.027	0.026	579.25	641.30	610.27	16.28	16.59	16.43	12.48	12.76	12.62
128 days	16.72	16.98	16.85	0.026	0.027	0.026	619.25	676.84	648.07	16.77	16.15	16.46	12.53	13.05	12.79
135 days	16.55	16.41	16.48	0.025	0.024	0.024	662.51	710.84	686.68	16.57	16.02	16.29	12.76	12.42	12.59
SE±m	0.15	0.13	**0.14**	0.001	0.01	**0.001**	9.08	8.60	**7.01**	0.31	0.32	**0.29**	0.12	0.12	**0.23**
CD 5%	0.46	0.49	**0.42**	0.003	0.04	**0.003**	27.21	25.73	**20.98**	0.94	0.96	**0.86**	0.35	0.36	**0.31**

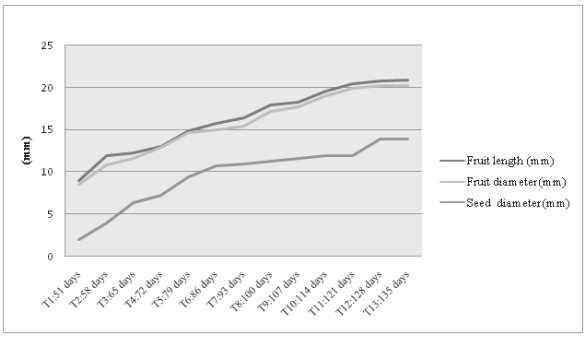

Fig. 1 : Changes in fruit size of longan with advancement of fruit growth

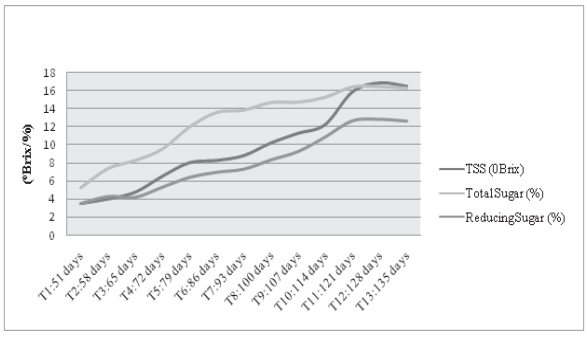

Fig. 2 : Changes in quality parameters of longan fruits with advancement of fruit growth

as in pooled value. The initial pooled value of acidity of longan fruit was only 0.016% at 51 days after fruit set which has attained to 0.023% at 93 days after fruit set as higher level. Although a highest level of acidity of longan fruit juice was recorded as 0.026% at 121 days after fruit set which remained same upto 128 days after fruit set after which it has reduced to 0.025% at 135 days after fruit set.

TSS-Acidity ratio: A drastic increase in TSS-acidity ratio of juice of longan fruits has been observed in the present experiment with advancement of fruit growth and maturity of longan in the 2021 and 2022 as well as pooled value (Table 2). The pooled initial ratio was recorded as 219.37 at 51 days after fruit set which has increased to 402.04 at 93 days after fruit set during the first phase of fruit growth. On the other hand the TSS-acidity ratio was started to increase from 402.04 at 100 days after fruit set to 686.68 at 135 days after fruit set of longan. Thus a double sigmoid nature of fruit growth and development with respect to TSS-acidity ratio was recorded in can of fruit development of longan upto fruit maturity.

Total sugar: The total sugar content of longan fruits have shown an increasing pattern with advancement of growth and attaining maturity into two distinct phases (Table 2 and Fig. 2). At the beginning the total sugar content of longan fruit juice was recorded as 5.24% at 51 days after fruit set which was increased to 11.97% at 79 days after fruit set which is clear from the pooled data of both the years. Again during second phase of fruit growth of longan, the total sugar content has increased from 13.59% at 86 days after fruit set to 16.46% as highest at 128 days after fruit set after which reduced to 16.29% at 135 days after fruit set. It was thus clear that the longan fruit also has shown double sigmoid pattern of growth with respect to total sugar content of fruit juice.

Reducing sugar: A gradual increase of reducing sugar content of longan fruit juice have been observed in the present investigation while experimenting the biochemical changes pattern of longan fruits with advancement of fruit growth towards maturity (Table 2 and Fig. 2). The statistical analysis of data on both the years as in pooled data indicated 3.48% of reducing sugar content in juice in longan fruits at 51 days after fruit set while it has been reached to a steady state after attaining reducing sugar content of 12.62% at 121 days after fruit set. Although, a little increase of reducing sugar has also been observed upto 12.79% at 128 days after fruit set which has reduced to 12.59% at 135 days after fruit set of longan.

In the present experiment the fruit growth and development took place in two different phases after fruit set. First stage of fruit growth was from fruit set to 93 days after fruit set i.e. 14 week age of the fruit. On the other hand, the second phase of fruit growth was observed from 100 days after fruit set to 128 days after fruit set with respect to most of all fruit physical and biochemical parameters. As per the findings of various workers on phonological studies of longan fruit that the fruit development can be divided into two main growth stages (Chen *et al*., 1995; Xu *et al*., 1997).

In the first stage, pericarp and aril development started and the seed coat, embryo and endosperm can be observed with the naked eye. The second stage is characterized by the embryo filling the seed

cavity, hardening of the seed coat, thinning of the pericarp, development of the fleshy aril and the maturation process. Two waves of fruit maturation in longan fruits in China was observed by Zee *et al*. (1998). The first cycle occurred 43-70 days after fruit set, and the second 80-120 days before harvest. In southern Spain, the first sigmoid curve appears 8 weeks after anthesis (Davenport and Stern, 2005). Thus the findings of the present experiment have conformity with findings of above mentioned scientists.

The fruits are ready for harvest when the pericarp is thin, smooth, tough and leathery, and its color changes from green yellow to yellow-brown (Subhadrabandhu and Stern, 2005). The same applies for fruit maturity that in present experiment occurred at 121 to 128 days after fruit set and similar maturity stage has been reported in China in July-September (Wong, 1991), in Thailand in June-August (Stern, 2005; Wong, 2000), in Queensland from January/February to March/April (Stern, 2005; Wong, 2000), in Florida in August-September (Jonathan *et al*., 2013), and in northern Vietnam from July to August (FAO, 2004). This little variability of fruit growth, development and maturity of longan fruits around the world may be due to the differences in growing conditions and genotypic variability of the plants.

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Study on performance of different elite clones of jamun in top working under Hisar condition

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ABSTRACT

A study was carried out to study on performance of different elite clones of jamun in top working under Hisar condition. Scions of elite plants were collected from 18-different locations and budded on 25 years old jamun trees by top working during the year 2016. The results of two consecutive years of study revealed significant increase in growth, yield and quality of jamun. The maximum plant height was recorded from the plant, raised from the scion collected from Khera-1 Yamunanagar (7.6 m), whereas, the maximum stem girth was recorded from the collection taken from Dhani Chanderpal, Rohtak (26.4 cm). The maximum yield was recorded under collection from Milakpur, Hisar (18.4 kg/plant), maximum fruit weight was recorded from Milakpur, Hisar collection (9.2 g), whereas, minimum stone weight was recorded under collection from Chilkana-2 Saharanpur (1.0 g), maximum TSS was found in collection from Chhachrouli fruit nursery (14.0°Brix) and minimum acidity was recorded under collection from Dhani Chanderpal, Rohtak (1.00 %). Thus, it can be concluded that different scions collected from different sites showed variability in growth, yield and fruit quality of jamun.

Keywords **:** Fruit quality, fruit yield, scion, top working

INTRODUCTION

Jamun (*Syzygium cuminii*) is an indigenous tropical fruit tree of India belonging to family myrtaceae. It is also known by other common names like black plum, java plum, Indian blackberry etc. (Sartaj Ali *et al*., 2013). It is liked both by poor and rich but much liked by those who cannot afford to buy costly fruits. The tree bear fruits once in a year and the berries are sweetish sour in taste (Ghosh *et al*., 2016). Jamun is also appreciated as windbreaks in fields (Ud din *et al*., 2020). Jamun has gained commercial importance as a minor fruit in tropical and subtropical conditions. It is one of the most hardy fruit crops and can easily be grown even in marshy areas where other fruits fail to establish (Singh *et al*., 2007). Jamun has been attributed in the Indian folklore medicine system to possess several medicinal properties (Inamdar, 2000). Apart from consumption of its fruit pulp, its seeds are used in Ayurvedic medicine against diabetes, heart and liver trouble. The jamun fruits are abundant source of anthocyanins, phenols, pectin, protein and also rich in antioxidant properties. They are rich in anthocyanin pigments and are good source of natural food colourants (Chaudhary and Mukhyopadhyay, 2012). The seed contains jambosin and jambolin, which reduces diastatic conversion of starch into sugars. Besides its use as dessert fruit, jamun is used for preparation of delicious beverages, jellies, jam, squash, wine, vinegar etc.

Jamun is highly cross pollinated crop; hence wide variability is common in the species (Swamy *et al*., 2017). In nature, lot of variation with respect to fruit shape and size, TSS, acidity and earliness in bearing of this crop is evident. Advantages of these variations can be taken to evolve selections of superior types. Survey and selection are the best procedures to evolve suitable genotypes for a particular area of its natural existence (Swamy *et al*., 2017). Keeping in view, this experiment has been planned for "Study on performance of different elite clones of jamun in Top working under

Hisar condition" with the objective to evaluate the elite types for recommending under Hissar condition.

MATERIALS AND METHODS

The present investigation was conducted at Experimental orchard of Department of Horticulture, CCS Haryana Agricultural University, Hisar during the year 2018-22. Hisar falls within a typical semi-arid climatic region characterized by scorching, arid summers and extremely frigid winters. This area experiences substantial fluctuations in temperatures, both in terms of monthly maximum and minimum readings, during the summer and winter months. In the summer, from May to June, temperatures soar to a peak of approximately 48°C, while winter brings frigid conditions with temperatures occasionally dropping to freezing points in December and January. Rainfall exhibits significant variability in both its total quantity and distribution. Of the overall annual rainfall, which amounts to about 450 mm, a substantial 80% occurs during the monsoon season from July to September. There are sporadic, light showers from December to February due to western disturbances. The rainfall in this region is notably unreliable, with annual fluctuations ranging between 20-30%, and seasonal variations spanning 30-50%.

Twenty five years old 60 nos. unproductive trees were taken for top working study. There were 18-treatments (scion of 18-sites) with three replications of each (three top-worked trees from each site taking one plant as a unit) following randomized block design. The scions from 18-locations were collected and budded on old jamun trees by top working for evaluation of elite planting material of jamun. The criteria used for selection of scions from the trees of 18-sites was on the basis of yield, fruit size and quality of fruits. During the previous year of collection of scions i.e. July-August 2015, the trees were ear marked and then in the following year, the scions were collected.

The top working was done in July, 2016, taking scions from 18 locations of Hissar district. The 18-sites were i. Chhachrouli fruit nursery, ii. Khera-1 Yamunanagar, iii. Khera-2 Yamunanagar, iv. Chilkana-1 Saharanpur, v. Chilkana-2 Saharanpur, vi. Parkash Farm-1 (Chilkana, Saharanpur), vii. Parkash Farm-2 (Chilkana, Saharanpur), viii. Karam Sain Farm-1 (Chilkana, Saharanpur), ix. Rakesh Mehta Farm house (Yamunanagar), x. R.R.S Buria (Yamunagar), xi. Khijarpura (Kurukshetra), xii. Hansala (Kurukshetra), xii. Udarsi (Kurukshetra), xiv. Dhera Bhath Majra (Kurukshetra), xv. Dadlu (Kurukshetra), xvi. Dangli (Kurukshetra), xviii. Milakpur (Hisar) and xviii. Dhani Chanderpal (Rohtak). Success of top working 4-months after operation has been presented in the table.

The observations included plant height (m), stem girth (cm), yield (kg/plant), fruit weight (g), stone weight (g), TSS (°Brix) and Acidity (%). The height of the trees were measured with the help of measuring pole, up to maximum point of height. For measuring the stem girth a marking place using a black circular one-centimeter-wide band, the tree trunk was marked 10 cm above the graft union on scion part and girth was measured in centimeter with the help of measuring tape. Plant height and stem girth were recorded on first fortnight of February 2020, February 2021 and February 2022 and pooled data is presented in the paper.

To calculate total fruit yield, the total number of fruits per tree was multiplied with average fruit weight and the value was expressed in kilograms (kg/tree). For recording fruit weight five randomly selected fruits from the tagged branch of the tree were picked and weighed on top pan electric balance. To calculate the average fruit weight, the total fruit weight was divided by total number of fruits taken and expressed in grams (g).The total soluble solids (TSS) was measured by hand refractometer and fruit acidity was estimated by using the method given in A.O.A.C. (1990). The yield and fruit quality parameters were recorded in July-August of 2020, 2021 and 2022 and pooled data is presented in the Tables.

RESULTS AND DISCUSSION

Variation was observed in the success of top working and growth of scion of jamun germplasm collected from different sites (Table 1). Budding success, recorded 4-months after operation varied from 50 % to 68 % among the collection sites. The plant height ranged from 5.6 m to 7.6 m. The maximum plant height was recorded by the collection from Khera-1 Yamunanagar (7.6 m) and the minimum plant height was recorded by the collection from Parkash Farm-2, Chilkana,

Table 1: Growth of scion of jamun germplasm collected from different sites (Pooled data of 2020, 2021 and 2022)

Treatment (Site of collection)		Plant height (m)	Stem girth (cm)	Budding Success (%)
1.	Chhachrouli fruit nursery	7.0	24.0	58 %
2.	Khera-1 Yamunanagar	7.6	21.0	60 %
3.	Khera-2 Yamunanagar	6.2	22.5	56 %
4.	Chilkana-1 Saharanpur	5.8	23.0	50 %
5.	Chilkana-2 Saharanpur	6.5	23.5	54 %
6.	Parkash Farm-1, Chilkana, Saharanpur	6.8	21.2	51 %
7.	Parkash Farm-2, Chilkana, Saharanpur	5.6	22.5	68 %
8.	Karam Sain Farm-1, Chilkana, Saharanpur	7.4	24.6	66 %
9.	Rakesh Mehta Farm house, Yamunanagar	6.8	22.8	60 %
10.	R.R.S Buria, Yamunagar	6.5	23.4	54 %
11.	Khijarpura, Kurukshetra	6.7	23.0	57 %
12.	Hansala, Kurukshetra	6.6	23.4	50 %
13.	Udarsi, Kurukshetra	6.5	22.5	55 %
14.	Dhera Bhath Majra, Kurukshetra	6.0	24.6	62 %
15.	Dadlu, Kurukshetra	6.8	22.5	64 %
16.	Dangli, Kurukshetra	6.2	24.5	56 %
17.	Milakpur, Hisar	6.7	24.6	59 %
18.	Dhani Chanderpal, Rohtak	6.6	26.4	63 %
CD (p=0.05)		0.3	1.0	

Table 2: Yield and fruit quality of jamun germplasm collected from different sites (Pooled data of 2020, 2021 and 2022)

Site of Collection		Yield (kg/plant)	Fruit weight (g)	Stone weight (g)	TSS (^{0}Brix)	Acidity (%)
1.	Chhachrouli fruit nursery	15.5	8.7	2.1	14.0	1.05
2.	Khera-1 Yamunanagar	6.6	5.8	1.5	12.0	1.12
3.	Khera-2 Yamunanagar	6.4	6.5	1.8	11.8	1.20
4.	Chilkana-1 Saharanpur	5.8	5.6	1.4	12.2	1.15
5.	Chilkana-2 Saharanpur	3.5	5.0	1.0	10.6	1.25
6.	Parkash Farm-1, Chilkana, Saharanpur	3.6	5.2	1.1	11.4	1.22
7.	Parkash Farm-2, Chilkana, Saharanpur	5.5	5.8	1.5	11.8	1.18
8.	Karam Sain Farm-1, Chilkana, Saharanpur	6.0	6.0	1.6	12.0	1.20
9.	Rakesh Mehta Farm house, Yamunanagar	8.5	8.1	2.0	13.2	1.10
10.	R.R.S Buria, Yamunagar	3.0	5.2	1.1	10.0	1.22
11.	Khijarpura, Kurukshetra	5.0	5.0	1.0	11.4	1.16
12.	Hansala, Kurukshetra	9.2	8.5	2.0	12.6	1.12
13.	Udarsi, Kurukshetra	8.6	8.6	2.0	13.0	1.08
14.	Dhera Bhath Majra, Kurukshetra	10.7	9.0	2.2	12.5	1.10
15.	Dadlu, Kurukshetra	9.0	8.2	2.1	13.2	1.12
16.	Dangli, Kurukshetra	16.8	8.8	2.2	14.0	1.02
17.	Milakpur, Hisar	18.4	9.2	2.5	13.8	1.04
18.	Dhani Chanderpal, Rohtak	13.5	8.0	2.0	13.6	1.00
CD (p=0.05)		0.4	0.3	0.07	0.6	0.06

Saharanpur (5.6 m). Whereas, stem girth of scions varied from 21.0 cm to 26.4 cm. The maximum stem girth was recorded by the collection from Dhani Chanderpal, Rohtak (26.4 cm) and minimum stem girth was recorded by the collection from Khera-1 Yamunanagar (21.0 cm).

Similarly, variation was also observed in fruit yield and fruit quality of jamun germplasm collected from different sites. Table 2 shows that the yield ranged from 3.0 to 18.4 kg/plant. The maximum yield was recorded under collection from Milakpur, Hisar (18.4 kg/plant) and minimum yield was recorded under collection from R.R.S Buria, Yamunagar (3.0 kg/plant). On the other hand, fruit weight varied from 5.0 to 9.2 g. The maximum fruit weight was recorded from Milakpur, Hisar collection (9.2 g) and the minimum fruit weight was recorded from Chilkana-2 Saharanpur collection (5.0 g). Whereas, stone weight ranged from 1.0 to 2.5 g. The minimum stone weight was recorded under collection from Chilkana-2 Saharanpur (1.0 g) and the maximum stone weight was recorded under collection from Milakpur, Hisar (2.5 g). The TSS ranged from (10.0 - 14.0°Brix) and acidity varied from 1.25 to 1.00 %. The maximum TSS was found in collection from Chhachrouli fruit nursery (14.0°Brix) and minimum TSS was found in collection from R.R.S Buria, Yamunagar (10.0°Brix). Whereas, minimum acidity was recorded under collection from Dhani Chanderpal, Rohtak (1.00 %) and maximum acidity was recorded under collection from Chilkana-2 Saharanpur (1.25 %). Similar findings were reported by Jai Prakash *et al*. (2010) and Shahnawaz and Sheikh (2011).

CONCLUSION

The performance of different elite clones of Jamun in top working under Hisar conditions showed variability in growth, yield and fruit quality. However, the top worked plants raised from the scions of Milakpur, Hisar resulted in higher fruit yield, maximum sizeable fruits along with better fruit quality and these plants can be used as mother plants for further multiplication for cultivation in this region.

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Exploring *in vitro* efficacy of roots of *Bergenia ligulata* for urolithiasis management

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ABSTRACT

Urolithiasis is a metabolic disorder associated with formation of stones within the urinary tract. The physiochemical processes underlying stone formation involve precipitation, growth, aggregation and concretion of lithogenic salts in urine. An approach for the management of urinary stones, in lieu of surgical procedures, includes usage of herbal plants. Bergenia ligulata also known as "Paashanbheda" has been used for dissolving kidney stones traditionally. The objective of the present study is to investigate the calcium oxalate (CaOx) crystallization inhibition efficacy of aqueous root extract of Bergenia ligulata (BLAE) through in vitro experimental procedure- nucleation and aggregation assays. The inhibitory activity of the BLAE was determined by spectrophotometric assay for which percentage inhibition of calcium oxalate crystallization was calculated. BLAE showed dose dependent inhibition, i.e. more percentage inhibition of calcium oxalate crystallization was observed with the increase in the concentration of BLAE. Present study indicated significant efficacy of BLAE in inhibiting the formation of urinary stones displaying its inhibitory action in calcium oxalate formation in both nucleation and aggregation assays.

***Keywords*:** Aggregation assay*, Bergenia ligulata*, calcium oxalate, nucleation assay, urolithiasis

INTRODUCTION

Urolithiasis is a metabolic disease that is associated with multiple etiologies which occur as a result of complex interaction between genetic and environmental factors which leads to stone/urolith formation in the urinary tract (Yasui *et al*., 2008). It is a wide spread condition that can affect individuals across all geographical regions and demographics (Bawari *et al*., 2017; Thongprayoon *et al*., 2020; Li *et al*., 2023). About 12% of world population is affected by kidney stone disease and it is more common in males as compared to females (Sofia *et al*., 2016). Several risk factors are associated with occurrence of urolithiasis which included consumption of ground water, obesity, lack of physical activity, high intake of salts of calcium, phosphorus, magnesium *etc*., high intake of sugar, coffee and tea, increased consumption of red meat, less frequency of urination/ day and working in higher environmental temperature (Sequira *et al*., 2023). Changes in urinary salt excretion are associated with calcium oxalate (CaOx) stone formation. Two processes of stone formation are described in *Sushruta Samhita*, one of which include stagnation and supersaturation of urine and the other is by crystallization of crystalloids in the urine (Das *et al*., 2022). The conventional management of urolithiasis is based on surgical procedures that include external shockwave lithotripsy, ureteroscopy, percutaneous extraction but they are associated with certain drawbacks such as they are invasive procedures, there is higher rate of recurrence of the disease, associated with lifetime medical complications such as chronic kidney disease, hypertension *etc*. and involvement of high cost of treatment (Khan *et al*., 2021). Along with surgical management, medical management of urolithiasis include usage of certain medicine such as diuretics, urinary alkalizer *etc*. but no such satisfactory treatment is available

(Jamshed *et al*., 2022). One of the cheaper and safer alternatives for management of uroliths is the use of natural anti-urolithiatic agents derived from herbal plants (Khan *et al*., 2021). Traditional herbal plants are known to have several beneficial effects in the urinary system such as aqueous extract of *Urticadioica* was helpful in mass reduction of calcium oxalate stones in *in vitro* studies (Belmamoun *et al*., 2022). Another such important Himalayan plant is *Bergenia ligulata* also known as "*Paashanbheda*" in the Indian traditional system of medicine which is a perennial Himalayan herb belonging to the family Saxifragaceae and in the Indian subcontinent it is distributed along the high altitude of Himalayan regions (Goswami *et al*., 2013). The roots and rhizomes of the plant are commonly consumed for management of wounds, sepsis, asthma, cough and cold, inflammation, stomach disorders and urinary related issues (Ruby *et al*., 2012; Gurav *et al*., 2014). *Ayurvedic* formulations are known to use *Bergenia* species since centuries to deliquesce kidney and bladder stones (Ahmad *et al*., 2018). Some of the most important active phytoconstituents of *Bergenia ligulata* include bergenin, leucocyanidin, catechin, gallic acid, tannic acid, afzelechin, ß-sitosterol (Roychoudhury *et al*., 2022) which are responsible for the medicinal properties of the plant (Sadat *et al*., 2015). Aqueous extract of *Bergenia ligulata* showed curative effects against ethylene glycol induced urolithiasis in rats as there was significant decrease in serum and urine markers, decreased CaOx deposits during histological examination and minimum damage in the kidney cells (Sharma *et al*., 2017). The present study was conducted to evaluate the antilithiatic potential of aqueous roots extract of *Bergenia ligulata* (BLAE) by using different *in vitro* assays *i.e*. nucleation assay, aggregation assay, simultaneous flow static and dynamic model and reservoir static and dynamic model.

MATERIALS AND METHODS

Collection of plant material

The plant material (roots) utilized in this study was collected from Berinag, District Pithoragarh situated in the Kumaon region of Uttarakhand, India, located within 29.80° of North latitude and 80.07° of East longitude and authenticated by Dr. D.S. Rawat, Assistant Professor, Department of Biological Sciences, College of Basic Sciences and Humanities, G.B. Pant University of Agriculture & Technology, Pantnagar, Uttarakhand, India (Voucher specimen GBPUH-1437).

Chemicals used in the study

The study employed usage of analytical research grade chemicals that include calcium chloride, calcium acetate, sodium carbonate, sodium oxalate, sodium chloride, trisodium phosphate and Tris. These chemicals were purchased from Himedia (India) and SRL (India). Autoclaved distilled water was used to prepare different buffers and solutions.

Preparation of the extract

The collected plant material was thoroughly rinsed with running tap water and subsequently by distilled water. The cleansed plant material was then shade dried and after complete drying, it was ground into fine powder. 50 grams quantity of the dried powder was soaked in 250 mL of double-distilled water which was then homogenised at a temperature of 37^{o}C for duration of 72 hours in an incubator-shaker unit. Subsequently, the mixture was first strained through the muslin cloth and then passed through Whatmann filter paper No. 1. The resulting aqueous extract of *Bergenia ligulata* (BLAE) was then kept on water bath at 37^{o}C-40^{o}C to evaporate the water followed by lyophilisation and then it was stored at -20^{o}C till further use**.**

Evaluation of anti-urolithiatic activity of the plant extract by using *in vitro* assays

Different *in vitro* assays were conducted to study the anti-urolithiatic activity of BLAE *viz*. nucleation, aggregation and CaOx mineralization inhibition assays. Crystallization or inhibition assays were conducted with or without BLAE to evaluate the inhibitory potential of BLAE.

Nucleation assays

The nucleation assay was conducted by the method as described by Patel *et al*. (2012) with slight modifications. A solution consisting of 5 mmol/L calcium chloride ($CaCl_2$) and 7.5 mmol/L sodium oxalate ($Na_2C_2O_4$) was prepared in a buffer containing 0.05 mol/L Tris-HCl and 0.15 mol/L sodium chloride (NaCl) buffer adjusted at a pH of 6.5. Different dilutions of BLAE ranging from 25-

1000 µg/ml were prepared in distilled water. 100 µl of different dilution of BLAE was then mixed with 950 µl of $CaCl_2$ solution, which was followed by addition of 950 µl of $Na_2C_2O_4$ solution which initiated the crystallisation. The final mixture was incubated at a temperature of 37ºC for a period of 1 hour. 100 µL of buffer was added to $CaCl_2$ solution which was used as a control in the experiment, and was similarly incubated at 37ºC for a period of 1 hour. Subsequently, the nucleation of the crystals was observed under microscope at 40X magnification.

Aggregation assay

The method used to perform aggregation assay was as described by Hess *et al*. (1989) with some modifications. To create 'seed' CaOx crystals, a solution was prepared by mixing calcium chloride, $CaCl_2$ (6.0 m mol/L) and sodium oxalate, $Na_2C_2O_4$ (6.5 mmol/L) in a buffer containing Tris-HCl (0.05 mol/L) and NaCl (0.15 mol/L) at a pH of 6.5. Then, 950 µL of $CaCl_2$ solution was mixed with 100 µL of different dilution of BLAE ranging from 100-1500 µg/mL to examine the degree of inhibition of aggregation by comparing the turbidity of samples in the presence of the plant extract at different concentrations. After this step, 950 µL of $Na_2C_2O_4$ was added to it which initiated crystallization. 100 µL of buffer was added to $CaCl_2$ which was used as control in the experiment. The resulting mixture was then incubated for a period of 1 hour at 37ºC. The optical density (OD) of the crystallized suspension was measured at a wavelength of 620 nm.The inhibition of percentage aggregation was then calculated by comparing the turbidity observed in presence of the extract with that in the control.

The following formula was used to calculate percent aggregation inhibition:

% Inhibition = [1 - (Turbidity of the sample / Turbidity of the control)] × 100

Inhibition of calcium oxalate mineralization

To evaluate the i*n vitro* inhibition of CaOx mineralization by BLAE was examined using four different experimental models *viz*., 'simultaneous flow static model' (S.S.M.), 'simultaneous flow dynamic model' (S.D.M.), 'reservoir static model' (R.S.M.) and 'reservoir dynamic model' (R.D.M.).

Simultaneous flow static model (S.S.M)

The method used for S.S.M.was as described by Farook *et al*. (2004) with slight modifications. Three different burettes were filled with 50 mL of 0.01M of $Na_2C_2O_4$, 50 mL of 0.01M of calcium acetate, $Ca(C_2H_3O_2)_2$ and 50 mL of the BLAE (1500 µg/ml). Subsequently, all the solutions were then allowed to fall simultaneously into a 250 mL beaker in a controlled drop wise manner and with equal speed. After the process was completed, the mixture was digested in a hot water bath for a period of 10 minutes and then cooled down to room temperature. The precipitate was then collected into a pre-weighed centrifuge tube by centrifuging small volumes at a time and discarding the supernatant liquid. The tube with the precipitate was subjected to drying in a hot air oven at 120^0C, cooled to room temperature and weighed.

Simultaneous flow dynamic model (S.D.M.)

The procedure was similar as S.S.M except that the reaction mixture in the beaker was continuously stirred on a magnetic stirrer during the flow of salt forming solutions as well as the inhibitor (Farook *et al*., 2004).

Reservoir static model (R.S.M)

In this particular model, the entire volume of BLAE (50 mL) was placed in the beaker. Subsequently, the two salt forming solutions were allowed to run into it gradually through burettes and thus forming a reservoir of inhibitor into which the salt forming solutions ran down and the rest procedure was similar to that of simultaneous flow static model (Farook *et al*., 2004).

Reservoir dynamic model (R.D.M)

The procedure in the R.D.M. was similar to that of reservoir static model except that the reaction mixture was continuously stirred on a magnetic stirrer during the experiment (Farook *et al*., 2004).

For the analysis of the data the observations were recorded in triplicates, and their mean ± standard deviation (SD) values were calculated for statistical analysis. Then the results were analysed using a one factorial Completely Randomized Design and to assess the significance of difference between different treatment means, a critical

difference was determined at a 5% level of significance.

RESULTS AND DISCUSSION

Effect of BLAE on nucleation assay

In the present study it was observed that as the concentration of BLAE was increased, there was a reduction in the aggregation of CaOx crystals and the percentage of inhibition of CaOx crystallization was also increased as presented in Figure 1.

Effect of BLAE on calcium oxalate crystallization through aggregation assay

A dose dependent inhibition of CaOx crystallization was shown by the plant extract in the *in vitro* aggregation assay. As the concentration of BLAE was increased, there was an increase in the percent inhibition of aggregation. The highest percentage of inhibition of aggregation was obtained at a concentration of 1500 µg/ml which was 85.13%. The results are shown in Table 1 and Figure 2.

Effect of BLAE on Inhibition of calcium oxalate mineralization

The inhibitory effect of BLAE on the formation of CaOx crystal has been studied in various dynamic models such as simultaneous flow static model, simultaneous flow dynamic model, reservoir static model and reservoir dynamic model. For this, BLAE at a concentration of 1500 µg/ml was taken and the above-mentioned procedure was followed. The formed precipitate was quantified at last and then percentage of inhibition of crystallization was calculated in various models. The percentage of inhibition of crystallization was found to be highest in reservoir static model (53.88%), followed by simultaneous flow dynamic model (32.27%), reservoir dynamic model (26.10%) and least in simultaneous flow static model (14.78%). The result showing the percent of inhibition by BLAE in various dynamic models is presented in Figure 3.

The oxalates can initiate a vicious cycle of oxidative stress in the renal epithelial cells which can lead to cellular injury and deposition of the crystal of CaOx on the injured cells which further cause oxidative damage and inflammation which can further lead to irrevocable damage of kidney cells (Singh *et al*., 2022). To prevent this kind of damage *Bergenia ligulata* is a traditional medicinal plant rich in antioxidants which is used for the management of kidney stones. Several reports suggest that the mechanism involved behind the antilithiatic effect of *Bergenia* sp. is mainly by diuresis, inhibiting formation and aggregation of CaOx crystals, antioxidant activity and hypermagnesemic effects (Nagal *et al*., 2013). Aqueous extract of *Bergenia ligulata* showed inhibition of calcium oxalate monohydrate (COM) crystals (Joshi *et al*., 2005). Methanolic extract of *Bergenia ligulata* as well as bergenin is known to inhibit, show significant anti-oxidant effect against 1, 1-diphenyl 2-picrylhydrazyl free radical as well as prevented lipid peroxidation in *in vitro* condition and prevented deposition of CaOx crystals in the renal tubules of rats (Bashir and Gilani, 2009). Ethanolic extract of *Bergenia ligulata* showed a dose dependent inhibition of nucleation and aggregation process of CaOx crystal formation and also showed a cytoprotective effect on renal epithelial NRK-52E cells against oxalate injury. It was also found that when the renal cells were exposed to the extract the COM crystals were converted to CaOx dihydrate crystals that are known to be less injurious (Singh *et al*., 2021). Bergenin isolated from rhizome of *Bergenia ligulata* when given at a dose of 10 mg/kg body weight to hyperoxaluric rats-maintained oxidant/ antioxidant balance, improved creatinine clearance and decreased kidney damage and thus helpful in managing the CaOx calculi (Aggarwal *et al*., 2014). *Bergenia ligulata* shows it antiurolithiatic effect by decreasing oxidative stress, modulating structure of crystals and preventing adhesion of crystal thus, exhibiting cytoprotective effect (Singh *et al*., 2022). *Bergenia ligulata* is one of the major ingredients of a polyherbal formulation named cystone which is commonly used in the management of uroliths. *Bergenia ciliata* is another species of the genus *Bergenia* which was found to inhibit the nucleation as well as aggregation of COM crystals in a dose dependent manner. 70% methanolic extract of rhizomes of *Bergenia ciliata* also prevented the histopathological changes in animal model of ethylene glycol induced hyperoxaluria (Saha and Verma, 2011). A crude phenolic compound when

Table 1: Percentage inhibition of calcium oxalate crystallization by BLAE in aggregation assay

S. No.	Different concentration of BLAE	Percent inhibition of Aggregation (Mean±S.D.)
1.	25 µg/ml	18.85±1.57
2.	50 µg/ml	31.91±2.10
3.	100 µg/ml	37.09±1.87
4.	250 µg/ml	43.38±1.77
5.	500 µg/ml	52.35±1.30
6.	750 µg/ml	58.77±1.95
7.	1000 µg/ml	72.93±2.06
8.	1250 µg/ml	75.74±1.72
9.	1500 µg/ml	85.13±1.74
	Critical Difference	3.112
	Coefficient of Variation	3.402

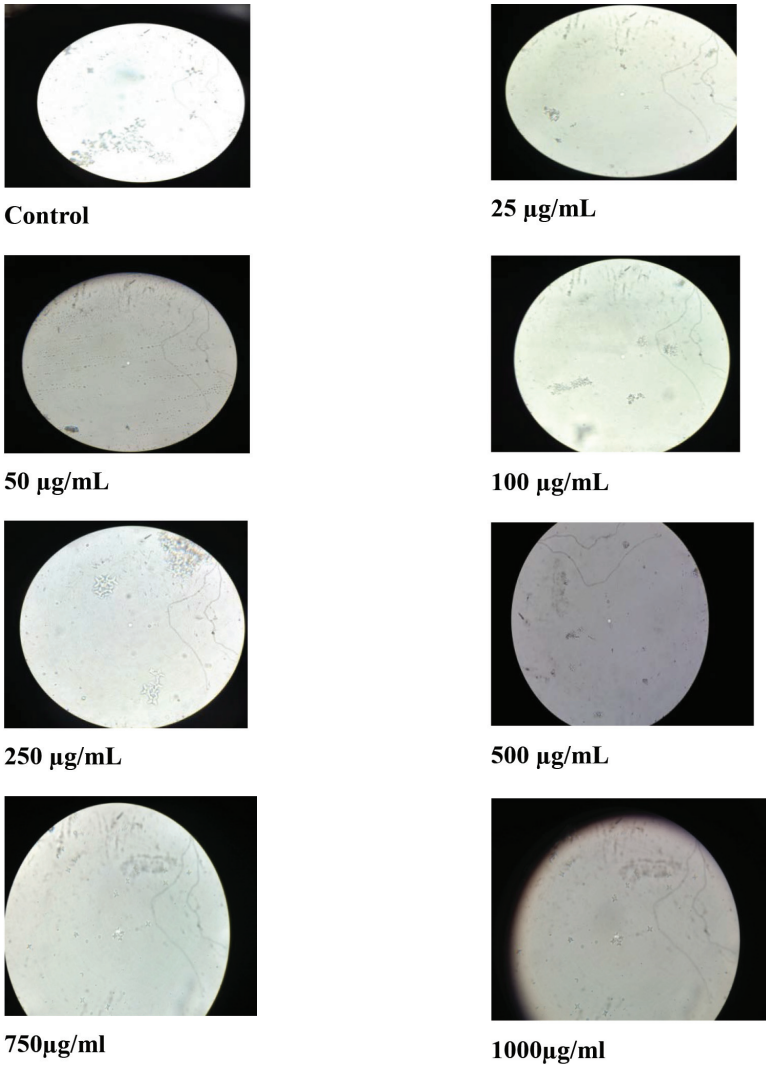

Fig. 1: Nucleation assay at different concentration of BLAE observed under Microscope (40X)

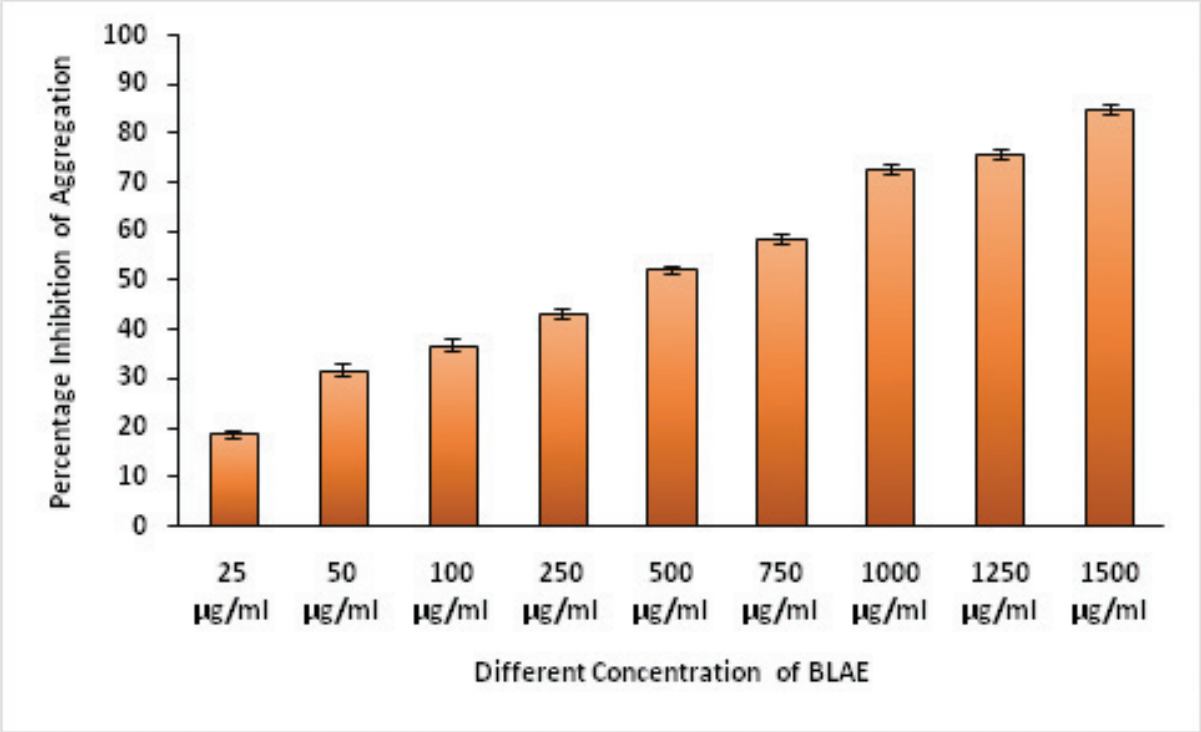

Fig. 2: Percentage inhibition of calcium oxalate crystallization by BLAE in aggregation assay

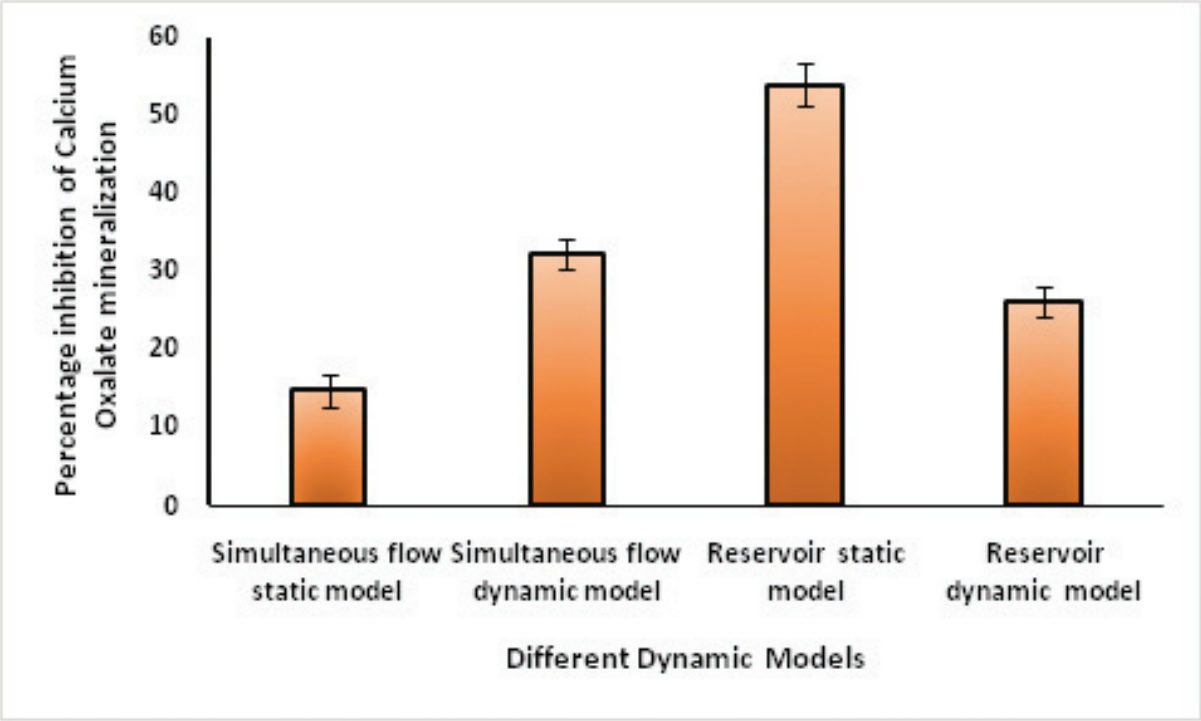

Fig. 3: Percentage inhibition of Calcium Oxalate mineralization by BLAE in different models

isolated from *Bergenia ciliata* showed highest dissolution of calcium phosphate and CaOx stones (Byahatti *et al*., 2010).

CONCLUSION

Thus, the *in vitro* assays in the present study suggest that aqueous extract of roots of *Bergenia ligulata* has a significant anti-urolithiatic activity on the basis of results of nucleation assay, aggregation assays and inhibition of CaOx mineralization as seen in different dynamic models. There was inhibition of nucleation as well as aggregation of CaOx crystals which is a common component of kidney stones/uroliths. Significant efficacy was found in the highest concentration of the plant extract *i.e.,* 1500 µg/ml. Further *in vivo* studies are required to explore the mechanism of action of aqueous extract of *Bergenia ligulata*. The active phytoconstituents present in the plant extract are responsible for the anti-urolithiatic activity which will require further characterization and isolation of the active compounds.

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SHORT COMMUNICATION

Ethnobotanical study of medicinal plants used to treat human ailments in hilly areas of District Kupwara, Jammu and Kashmir

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ABSTRACT

Many rural groups in impoverished nations still use plant-based medicines today, and contemporary healthcare systems depend on components from plants. In our nation, the Kashmir Himalayas are home to a great trove of medicinal plants. The present study focused on the use of medicinal plants for the treatment of human diseases. Data was collected by performing surveys in the study area. Semi structured interviews and group discussions were conducted with local communities preferably in their local language in order to collect appropriate and reliable information. Throughout the course of the study, interviews with 60 families made up of villagers, medical professionals, tribal members, and traditional healers were conducted. Local experts and area herbal healers were mostly consulted for the collecting of data. The traditional primary healthcare system in the investigated area includes herbal medicine as a crucial component. It was observed that people use different plant parts for treatment of their ailments i.e., leaves, whole plant, fruit, seed, root, flower, rhizome, bark etc. In the study area, residents use a total of 33 species from 21 families to treat various illnesses. Asteraceae was found to be the dominant family in which maximum 7 species were found.

Keywords: Family interview, medicinal plants, treatment of human diseases, traditional plant knowledge

INTRODUCTION

Subtropical, intermediate, temperate, and cold desert zones are among the diverse agroclimatic zones found in the Union Tertiary of Jammu and Kashmir, which is located at the westernmost tip of the Himalayan mountain range. Due to the state's varied climate and altitude, which has created an excellent habitat for the growth of a diverse range of forests, it is endowed with a rich floral diversity. Within just 2.15 percent (15,948 km^2) of the total land area, the Kashmir Himalaya alone is responsible for almost 2,000 (20%) plant species. In most nations, medical plants—also known as herbs, herbal remedies, pharmacologically active plants, or phytomedicinals—remain the principal source of medication. For their everyday healthcare needs, more than three-fourths of the world's population rely mostly on unprocessed plant products (Barrett and Kieffer, 2001). The usage of plant-based health products has significantly increased in both developed and developing nations as a result of recent advances in plant sciences. Around 70–80% of individuals rely on medicinal herbs for their daily health care. In Kashmir Valley, medicinal plants are also a major source of revenue for thousands of families. (Mushtaq *et al*, 2020).

From the very ancient times, People living here in this chunk of world are admiring the plant resources for various purposes including healthcare and food, which is also mentioned in Kalhans's Rajtarangini (1149-50 AD). Till date a number of ethnobotanical studies have been carried out by researchers throughout the Kashmir Himalaya (Dar *et al*., 1984), but most of the studies are related to general assessment and documentation of medicinal plants. A very few studies have been carried out merely on the medicinal uses of plants to cure human diseases. But no such systematic study has been carried out in District Kupwara Jammu and Kashmir (J&K). India is blessed with a variety of ecological conditions, abundant natural resources, and a long history of traditional farming practices that are in keeping with its ethnic diversity and old culture. It traditionally provides vast amounts of plant-based raw materials that are used all over the

world in the pharmaceutical, cosmetics, fragrance, and allied industries. More than 2,000 species of therapeutic plants have been documented in its extensive medicinal plant flora. 1,100 species are used in various medical systems, and of them, 600–700 species are extensively used throughout the nation, primarily by local enterprises. Commercial use includes about 150 species. Many of these are exported to different nations throughout the world. The western Himalayas are said to have 50% of the plant medicines listed in the British pharmacopoeia. It serves the following medical systems: 30% Ayurvedic, 46% Unani, and 33% Allopathic (Meena *et al*. 2009).

Kashmir In our nation, the Himalayas are home to a great trove of therapeutic plants. According to Phondani *et al.* (2010), forests are crucial to the viability and survival of indigenous households in India. The goals and purposes of ethnobotany and ethnomedicine are the interactions between human behavior and the plant communities with which they interact. Ethno botanical studies look on how these plants' resources are used for food, medicine, fuel wood, agriculture, housing, crafts, fodder, and religious rituals (Khan *et al*. 2003). Rural inhabitants in the Himalayan region, particularly those living close to forested areas; rely more heavily on the use of forest resources. About 800 plant species are thought to be used as food and medicine in India, primarily by the country's tribal people (Tantray *et al*., 2009; Bhat *et al*., 2012; Hassan *et al*., 2013).

In order to record the traditional uses of medicinal plants for treating human ailments, the current study was done in the mountainous areas of district Kupwara in the Kashmir Himalayas. The following objectives were studied

1. Taxonomic evaluation of the medicinal plants found in the concerned district

2. Effect of medicinal plants on human Ailments

MATERIALS AND METHODS

The current study was carried out in district Kupwara, during the year 2022 and 2023. The details of the technique followed and materials used during the course of investigations are described below.

Study area

The Himalaya is known for its loftiest and longest mountain ranges. Kashmir is one of the provinces of Jammu and Kashmir Union Territory in the laps of Himalayas. The study area is located in Kupwara district of North Kashmir. The northern frontier district of Kupwara, which was established in 1979, has a total size of 2379 square kilometers and more than 240 kilometers of LOC (Line of Control). With its vibrant culture, varied past, distinctive folklore, and expansive meadows, the district of Kupwara is breathtakingly lovely. Between the Pir Panchal and Shams Bari mountain ranges is where you'll find Kupwara district. These mountain ranges are surrounded by lovely pastures and meadows that serve as grazing land for sheep and cattle. In addition, they serve as vacation locations for healthy travelers and daring spirits.

Taxonomic evaluation of the medicinal plants found in the concerned District:

Data was collected with respect to medicinal plants scientific name, local name, Family and Life-form.

Sampling :

Number of districts surveyed: 1; **Total Number of Blocks surveyed:** 3 (Kralgund, Langate, Qalamabad); **Number of informants surveyed in each Block:** 20;

Total number of informants surveyed in the district: No. of district × No. of blocks × No. of informants from each block = 1×3×20 = 60 informants

Effect of medicinal plants on human diseases

The present study focused on the use of medicinal plants for the treatment of human diseases. Data was collected by performing surveys in the study area. Semi structured interviews and group discussions were conducted with local communities preferably in their local language in order to collect appropriate and reliable information. Information was collected with respect to plants used in treating diseases, local name, mode of administration, preparation and application of the remedy. Throughout the course of the study, interviews with 60 individuals, including members of several tribes, physicians, traditional healers, and locals, were conducted. Local experts and area

Table 1: Taxonomic evaluation of medicinal plants in district Kupwara

S.No	Scientific name	Local name	Family	Life form
1	*Arnebia euchroma* Royle	Ratanjot	Boraginaceae	Herb
2	*Achillea millefolium* Linn.	Pahel-ghass	Asteraceae	Herb
3	*Skimmia anquetilia* Taylor	Inga	Rutaceae	Herb
4	*Jurinea dolomiaea* Royle	Guggal dhoop	Asteraceae	Herb
5	*Juglans regia* Linn.	Doon	Juglandaceae	Tree
6	*Saussurea costus* Falc.	Kouth	Asteraceae	Herb
7	*Hypericum perforatum* Linn.	Shin-chae	Clusiaceae	Herb
8	*Aconitum heterophyllum* Royle	Patrees	Renanculaceae	Herb
9	*Artemisia absintum* Linn.	Tethwan	Asteraceae	Herb
10	*Ajuga parviflora* Benth.	Jan-i-adam	Lamiaceae	Herb
11	*Rheum webbianum* Royle	Pambchalan	Polygonaceae	Herb
12	*Prunella vulgaris* Linn.	Kal-wyoth	Lamiaceae	Herb
13	*Arnebia benthami* Wall.	Kahzaban	Boraginaceae	Herb
14	*Cichorium intybus* Linn.	Waare hand	Asteraceae	Herb
15	*Datura stramonium* Linn.	Datur	Solanaceae	Herb
16	*Iris nepalensis* D.Don	Mazarmunji	Iridaceae	Herb
17	*Euphorbia royleana* Boiss.	Sochal	Euphorbiaceae	Herb
18	*Polygonum plebium* R.Br.	Drabb	Polygonaceae	Herb
19	*Rumex nepalensis* Spreng.	Abij	Polygonaceae	Herb
20	*Taraxicum officinale* Linn.	Hand	Asteraceae	Herb
21	*Thymus linearis* Benth	Jayind	Lamiaceae	Herb
22	*Urtica diocia* Linn.	Soi	Urticaceae	Herb
23	*Polygonatum verticillatum* Linn	Salam mishri	Liliaceae	Herb
24	*Ficus carica* Linn.	Anjeer	Moraceae	Tree
25	*Salix alba* Linn.	Veer	Salicaceae	Tree
26	*Abies pindrow* Royle	Budul	Pinaceae	Tree
27	*Berberis lyceum* Royle	Kawdach	Berberidaceae	Shrub
28	*Centauria iberica* Spreng.	Kreach	Asteraceae	Shrub
29	*Cannabis sativa* Linn.	Bhang	Cannabinaceae	Herb
30	*Indigofera heterantha* Wall.	Zand	Fabaceae	Shrub
31	*Mentha arvensis* Linn.	Pudneh	Lamiaceae	Herb
32	*Punica granatum* Linn.	Daen	Punicaceae	Shrub
33	*Viola odorata* Linn.	Bunfsha	Violaceae	Herb

herbal healers were mostly consulted for the collecting of data. Local herbal healers confirmed and double-checked the information learned from locals.

Research Findings : The research findings of the present study obtained have been presented objective wise as under:

Taxonomic evaluation of the medicinal plants found in the concerned blocks

The results pertaining to taxonomic evaluation of medicinal plants in the study area are shown in Table 1 and 2. Studies conducted revealed that a total of 33 species belonging to 21 families were used by locals to treat different diseases in the study area. Asteraceae was found to be the dominat family in which maximum 7 species were found followed by Lamiaceae (4), Polygonaceae (3), Boraginaceae (2), Rutaceae (1), Juglandaceae (1), Clusiaceae (1), Renanculaceae (1), Solanaceae (1), Iridaceae (1), Euphorbiaceae (1), Urticaceae (1), Liliaceae (1), Moraceae (1), Salicaceae (1), Pinaceae (1), Berberidaceae (1), Cannabinaceae (1), Fabaceae (1), Punicaceae (1) and Violaceae (1) (Fig 1). It was also revealed that 25 species were herbs, shrub (4) and trees (4) (Fig 2).

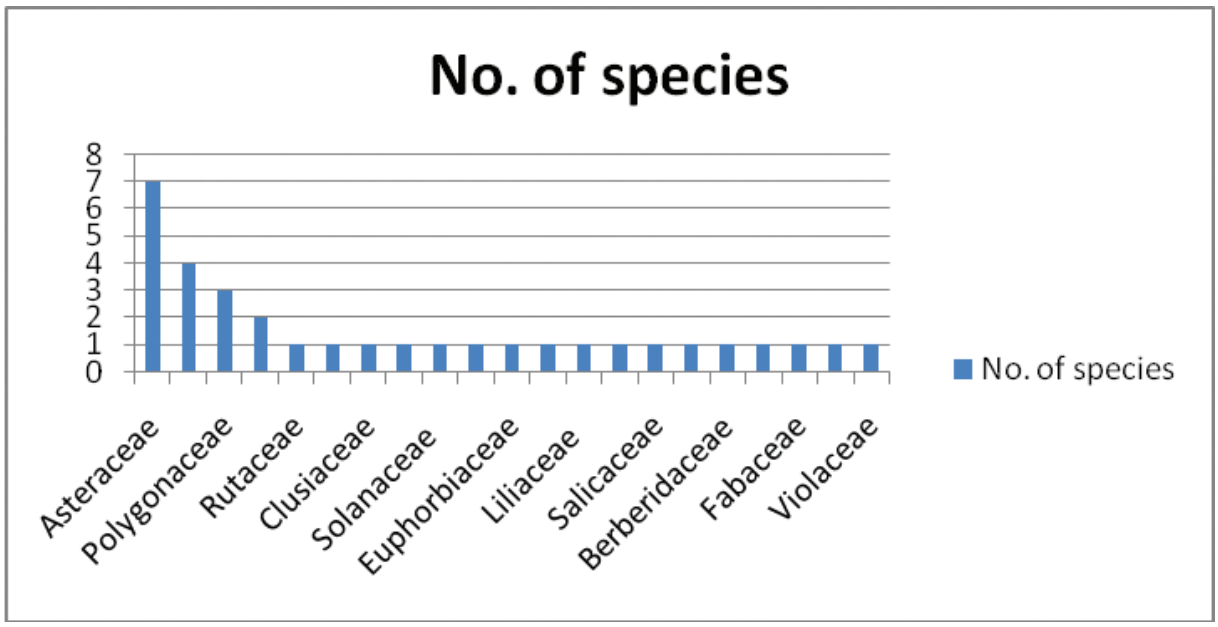

Fig 1: Distribution of species among Families

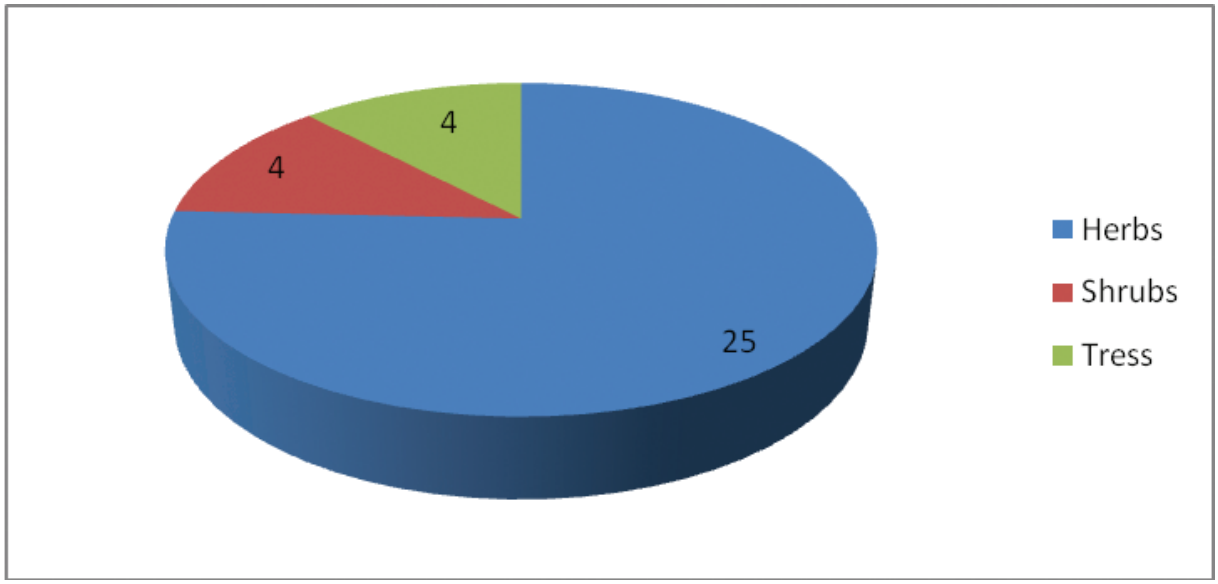

Fig 2: Distribution of species according to their Life-form

Effect of medicinal plants on human ailments

The current study found that residents of the study region use medicinal plants to treat a variety of illnesses and ease pain. The traditional primary healthcare system in the investigated area includes herbal medicine as a crucial component. People have been observed using a variety of plant parts for therapy, including leaves, the entire plant, fruit, seeds, roots, flowers, rhizomes, and bark. Table 3 shows the use of various medicinal plants for curing different diseases in district Kupwara of J&K.

Discussion

The people in the study area use medicinal plants for treatment of various diseases in order to alleviate sufferings and ailments. Herbal medicine forms an essential part of the traditional primary medical system in the studied area. Different researchers have reported traditional uses of medicinal plants

Table 2: Distribution of species among Families

S.No.	Family	Number of species
1	Asteraceae	7
2	Lamiaceae	4
3	Polygonaceae	3
4	Boraginaceae	2
5	Rutaceae	1
6	Juglandaceae	1
7	Clusiaceae	1
8	Renanculaceae	1
9	Solanaceae	1
10	Iridaceae	1
11	Euphorbiaceae	1
12	Urticaceae	1
13	Liliaceae	1
14	Moraceae	1
15	Salicaceae	1
16	Pinaceae	1
17	Berberidaceae	1
18	Cannabinaceae	1
19	Fabaceae	1
20	Punicaceae	1
21	Violaceae	1

throughout Globe. According to Abdullah and Andrabi (2021), tribal tribes in the Ramhal Forest Division in Kupwara used ethnomedicinal herbs. In total, 65 medicinal plants from 40 different families were reportedly obtained from the research region. The majority of them are members of the families Asteraceae and Lamiaceae (9 species each), followed by the Apiaceae (3 species), with the other families each contributing just one or two species. Herbs made up the majority of the medicinal plants, followed by leaves, the entire plant, seeds, aerial parts, fruits, and stem latex. An ethnobotanical assessment of medicinal plants was carried out by Tangjang *et al.* (2010) in the Eastern Himalayan region of Arunachal Pradesh, India. The locals used a total of 74 medicinal plant species, spread across 41 families and 61 genera, in their traditional medical practices to treat at least 25 various diseases and ailments. Different modes of remedy preparation, routes administration and dosage for treating various ailments were observed during the study. Most species were prepared using a hot water decoction (32 species), a paste (23 species), a vegetable (9 species), or by eating them raw (7 species). Other preparations, including as essential oils, alkaloids, and ash powder, were typically made from freshly harvested plant material right before usage.

Table 3: Medicinal plants used by local people in District Kupwara

S. No	Scientific name	Local name	Family	Part used	Ailments treated	Mode of application
1	*Arnebia euchroma*	Ratanjot	Boraginaceae	Roots	Toothache, cuts and wounds, abdominal swelling and eye infections	Root used as pieces or in powdered form.
2	*Achillea millefolium*	Pahel-ghass	Asteraceae	Leaves	Leaves and floral heads are crushed and used to treat inflamated gums and toothache	Paste, External
3	*Skimmia anquetilia*	Inga	Rutaceae	Leaves, bark	Diabetes, Rheumatism, clearing of nose, flu, small pox, burns, snake and scorpion bites, body ache, headache	Leaf extract used to treat diabetes. Powder of plant bark for wounds and burns. Cold infusion of fresh leaves for smallpox, headache and fever. Whole plant also used as an anaesthetic.
4	*Jurinea dolomiaea*	Guggal dhoop	Asteraceae	Roots	Fractures, boils, fever	Decoction of roots is made
5	*Juglans regia*	Doon	Juglandaceae	Bark	Bark of the plant is obtained and rubbed on teeth to get relief from tooth ache	Raw, External

S. No	Scientific name	Local name	Family	Part used	Ailments treated	Mode of application
6	*Saussurea costus*	Kouth	Asteraceae	Roots	Arthritis, insecticide, cough, cold and asthma, fertility enhancer	The extract of root is used in preparation of rice and fed to arthritic patients.
7	*Hypericum perforatum*	Shin-chae	Clusiaceae	Leaves	Tea is made from crushed leaves and taken orally to treat joint pain and urinary disorders	Decoction, Internal
8	*Aconitum heterophyllum*	Patrees	Renanculaceae	Roots	Abdominal problems, fever, tooth ache, anti helminthic, gastric disorders	The root is used in powdered form or as pieces.
9	*Artemisia absintum*	Tethwan	Asteraceae	Whole plant	Anti helminthic, antiseptic, cardiac stimulant	Decoction of dried leaves used as wormicide.
10	*Ajuga parviflora*	Jan-i-adam	Lamiaceae	Leaves	Hepatitis C virus, cancer, jaundice, arthritis, fever, asthma and wounds	Powdered form of dried leaves taken with water. Fresh leaves can also be chewed or eaten as whole.
11	*Rheum webbianum*	Pambchalan	Polygonaceae	Leaves	Anti cancerous, memory booster, bowel abnormalities	Leaves are crushed into a paste.
12	*Prunella vulgaris*	Kal-wyoth	Lamiaceae	Flower, leaf	Frost bite, wound healing, joint pain, cough and cold, headache	Extract obtained from leaves and flowers. Dried leaves and flowers along with stem boiled and the water is used to soak feet and legs to relieve joint pain.
13	*Arnebia benthami*	Kahzaban/ gaw zaban	Boraginaceae	Flower	Fever, cough, throat diseases	Flowers boiled in water to make decoction.
14	*Cichorium intybus*	Waare hand	Asteraceae	Leaves	High fever, wound healing, ulcers, rheumatic pain	The entire plant is ground up and transformed into a paste. Moreover, it is a vegetable.
15	*Datura stramonium*	Datur	Solanaceae	Seeds, leaves	Boils, frost bites, skin infections	Boiled in water to cure frost. Leaves used in the form of paste on skin.
16	*Iris nepalensis*	Mazarmunji	Iridaceae	Roots	Rheumatic pain	Dried root is powdered and made into a paste
17	*Euphorbia royleana*	Sochal	Euphorbiaceae	Leaves	Wounds, boils	Leaves crushed to make a paste. Also used as a vegetable.
18	*Polygonum plebium*	Drabb	Polygonaceae	Leaves	Pmeumonia , bowel irregularities	Leaves used in the form of a decoction
19	*Rumex nepalensis*	Abij	Polygonaceae	Leaves	Cough, constipation, wounds, skin problems	Leaves cooked as a vegetable. Also used as a paste on skin.
20	*Taraxicum officinale*	Hand	Asteraceae	Leaves	Stomach cramps and ulcers, swelling, cough and asthma, urine irritation.	Leaves used as vegetable mostly in dried form.
21	*Thymus linearis*	Jayind	Lamiaceae	Whole herb	Cough, cold and fever	Dried plant is powdered and taken orally with milk.
22	*Urtica diocia*	Soi	Urticaceae	Whole herb	Dandruff , skin infections, wounds , fractures , fever	Extract is obtained by crushing the herb
23	*Polygonatum verticillatum*	Salam mishri	Liliaceae	Roots	Backache, leucorrhoea, menstrual troubles, appetizer	In leucorrhoea, root powder is mixed with water and taken daily.
24	*Ficus carica*	Anjeer	Moraceae	Fruits	Extract obtained from fruits is taken orally for indigestion, loss of appetite and diarrahoea	Juice, Internal

Contd.

Contd. Table 3

S. No	Scientific name	Local name	Family	Part used	Ailments treated	Mode of application
25	*Salix alba*	Veer	Salicaceae	Leaves	Boiling leaves in water and applying them to the legs and other body parts acts as an analgesic	Decoction, External
26	*Abies pindrow*	Budul	Pinaceae	Leaves	As a treatment for rashes on the skin, leaf paste is applied	Decoction, External
27	*Berberis lycium*	Kawdach	Berberidaceae	Fruits	For faster healing of wounds, fruit paste is administered externally	Decoction, External
28	*Centauria iberica*	Kreach	Asteraceae	Leaves	Skin Rashes, Wound healing	Decoction ,External
29	*Cannabis sativa*	Bhang	Cannabinaceae	Leaves	Dried leaf mixture smoked through a pipe called Hukkah is used to treat depression	Internal, External
30	Indigofera heterantha	Zand	Fabaceae	Rhizome, Bark	Abdominal pain, Toothache, Cough	Internal, External
31	*Mentha arvensis*	Pudneh	Lamiaceae	Leaves	To treat diarrhoea and low blood pressure, dried leaves are ground up and used with curd.	Internal
32	*Punica granatum*	Daen	Punicaceae	Fruit	Fruit juice can be used to cure diarrhoea and as a general body tonic	Internal
33	*Viola odorata*	Bunfsha	Violaceae	Flowers	Khambir, a type of jaggery made from crushed lowers and sugar, is used in the winter to treat throat infections	Internal

Studies conducted by Kaif *et al*. (2023) revealed that *Atropa acuminata* is an important medical plant used in traditional healthcare system in the area studied. A total of 15 ailments were found to be treated by *Atropa acuminata.* The ethno medicinal survey confirmed that different parts (roots, leaves, berries and whole plant) of investigating plant has medicinal values and is used for treatment of different diseases under traditional system of medicine. Studies conducted also revealed that 47% of the roots followed by 28% of whole plant, 20% of leaves, and 5% of berries were used for treatment of different diseases under ethno medicines. According to research done by Jan *et al.* (2021), the Gujjar and Bakerwal communities used a total of 60 plant species from 56 genera and 35 different families to treat a variety of illnesses. The area's leading plant family, Asteraceae, was identified; leaves were the most often used plant component, and decoction was the main method for preparing herbal recipes. Herbs made up the majority of the known medicinal plant species (72%), followed by shrubs (13%), and trees (15%).

CONCLUSION

The results of the present investigation, revealed that people in the study area use medicinal plants for treatment of various diseases in order to alleviate sufferings and ailments. It was found that the tribal tribes living at high altitudes rely heavily on herbal medicine to address their main healthcare needs. Traditional plant knowledge is only being passed down orally from one generation to the next, and it is quickly becoming extinct.

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International Journal of Minor Fruits, Medicinal and Aromatic Plants. Vol. 9 (2) : 205-208, December 2023

SHORT COMMUNICATION

Pratap Raj Dhania-1 (RKD 18): Promising coriander (*Coriandrum sativum* L.) variety for farmers of South Eastern Rajasthan

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ABSTRACT

Coriander (Coriandrum sativum L.) is an important seed spice crop cultivated throughout the world for its seed and leaves, used in flavouring and seasoning of the food and food products.The coriander variety RKD 18 (Pratap Raj Dhania 1), developed by Agricultural Research Station, Agriculture University, Kota (Rajasthan) possess many desirable traits like earliness, high essential oil, semi dwarf stature for lodging resistance, tolerance to pests and diseases along with high yield. Due to earliness of the variety along with many other desirable characters, the frontline demonstrations on coriander variety RKD 18 have been conducted at farmers' fields by different Krishi Vigyan Kendras (KVKs) of Agriculture University, Kota during the past years for dissemination of improved technology and popularization of the variety. In this status paper, salient features of this coriander variety, its production and distribution scenario has been discussed.

***Keywords*:** Coriander, quality**,** RKD 18, yield

Coriander (*Coriandrum sativum* L.), a member of family Apiaceae is an important seed spice crop of India cultivated for its seed and leaves, used in flavouring and seasoning of the food and food products. India is the largest producer, consumer and exporter of coriander in the world. The total production of coriander in the country was 811 thousand tons from an area of 640 thousand hectare (2021-22). The major coriander producing states are Rajasthan, Gujarat and Madhya Pradesh. It is cultivated in an area of 124286 ha in Rajasthan and produces 182705 m tonnes (2020-21) of coriander seeds. Rajasthan contributes nearly 19 and 20 percent to the country's total area and production, respectively. The south eastern part of Rajasthan *i.e*., humid south eastern plain zone or zone V is the largest producer of coriander in the state where it is cultivated in an area of 118030 ha contributing the production of 173764 m. tonnes (2020-21), thereby contributing around 95 percent to the total area and production of the state.

A perusal of Table 1 depicting coriander statistics (Vital Agricultural Statistics, Government of Rajasthan) indicates considerable decline in area and production of coriander in Rajasthan state and zone V during the past years, the reasons of which needs to be examined critically for reviving the area and production of this export oriented crop. In order to sustain the highest production of coriander in zone V of the state, high yielding varieties of coriander having other desirable traits like high quality in terms of high essential oil, early maturity, resistance against pests and diseases, suitable to the prevailing agro-climatic conditions are required. Pratap Raj Dhania-1 (RKD 18) having IC no. 594870 is a high yielding coriander variety notified in 2015 vide S.O.268 (E) for the state of Rajasthan. It's a selection from local germplasm of south eastern humid plain zone (Zone V) of Rajasthan. Since its notification, more than1200 quintals of seed of the variety has been produced by Agriculture University, Kota for conducting frontline demonstrations at farmers' fields and sale to the farmers, government and private institutions as well.

The salient features of the variety **Pratap Raj Dhania-1** are as under:

- **Plant height:** 70-80 cm
- **Days to 50% flowering:**55-58 days
- **Days to maturity:** 100-105 days

- **Test weight:** 12-15 g
- **Seed yield:** 18-20 q/ha
- **Essential oil:** > 0.4 %
- **Disease and pest reaction:** Moderately resistant to aphids, stem gall and tolerant to powdery mildew.

The details of yearwise production of the variety by the university and the revenue generated through sale have been provided in Table 2.

The frontline demonstrations on coriander variety Pratap Raj Dhania-1 have been conducted at farmers' fields by different Krishi Vigyan Kendras (KVKs) of Agriculture University, Kota during the past years for dissemination of improved technology and popularization of the variety. The variety has given higher yield as compared to local ones to the tune of 11 to 29 per cent. The farmers have also liked the variety due to its earliness, high yield, good aroma and tolerance towards diseases and pest.

Coriander is valued for its quality in terms of essential oil content as the essential oil is responsible for its aroma. The essential oil is volatile in nature and hence its concentration goes on diminishing with the rising temperature. The temperature in south eastern Rajasthan starts rising with the end of February and reaches as high as 35^0C by the month of March that adversely affects not only the essential oil content in coriander seed but decreases the seed yield as well. Giridhar *et al*. (2014) reported that the low productivity of coriander under rainfed situation is mainly due to terminal moisture stress that affects growth and productivity. Growing coriander in rainfed vertisol farming situation demands highly productive types with short (75 days) to medium (85-100 days) duration for cultivation. The optimum time of sowing of coriander in south eastern Rajasthan is last week of October to first week of November (Verma *et al*., 2017), therefore, in order to harvest high quality coriander, the demand is of an early variety that matures and becomes ready to harvest by the end of February or first week of March i.e. before the temperature starts rising. The another advantage of an early variety of coriander or any other winter (*rabi*) crop is that if the field is vacated by the month of February, the farmers can go for cultivation of spring (*zaid*) *mungbean* or other short duration vegetable crops so that they can take up three crops in a year adding to their annual income. Therefore, the farmers' preference is for early variety of coriander in which the third order umbels also mature by February so as to harvest higher yield with good aroma and also for cultivation of *zaid* crop during spring season.

The desirable attributes like earliness in days to flowering, days to maturity, higher number of umbellets per umbel and high essential oil (0.51%) in genotype RKD-18 was also reported by Giridhar *et al*. (2014). Doshi *et al*. (2014) also evaluated the coriander varieties popularly cultivated in zone V for their quality traits and reported coriander variety RKD 18 to be most suitable for processing for the production of value added products owing to its high percent essential oil and oleoresin content as compared to other checks. Earlier, Meena *et al*. (2013) also observed RKD 18 to be superior to other varieties in terms of seed yield, test weight, essential oil and in showing highest net return and B: C ratio and therefore, reported it to be most suitable and profitable for the south eastern humid plains of Rajasthan.

Verma *et al.* (2015) performed stability analysis among coriander genotypes and found RKD 18 to be highly stable over the environments for essential oil content. Based on the stability parameters, it was concluded that the stability of yield is imparted in the genotype RKD 18 through the number of secondary branches, number of umbels and umbellets per plant. This stable genotype can be further used for varietal development programme in coriander.

Suman *et. al.* (2018) evaluated the seed quality parameters in coriander and observed minimum seed metabolic efficiency in the genotype RKD18. Seed Metabolic Efficiency (SME) of the seed is the amount of dry seed weight that is required for producing one gram of dry root and shoot. Thus higher the value of seed metabolic efficiency, lower the efficiency of the seed as more seed reserve would be used for producing root and shoot. Sharma *et al.* (2019) studied twenty-three coriander genotypes to find out genetic variability using random amplified polymorphic DNA (RAPD) marker and found unique band in RKD-18 suggesting that unique sequences further can be

Table 1: Area, production and productivity of coriander in Rajasthan and zone V (South Eastern Humid Plain Zone)

Year	Area (ha)		Percent share of Zone V	Production (MT)		Percent share of Zone V	Productivity (kg/ha)	
	State	Zone V		State	Zone V		State	Zone V
2010-11	197891	193449	97.76	218899	215422	98.41	1106	1114
2011-12	267827	263881	98.53	329427	324837	98.61	1230	1231
2012-13	158678	156024	98.33	231925	229585	98.99	1462	1471
2013-14	182726	178210	97.53	117084	113898	97.28	641	639
2014-15	243910	242870	99.57	198764	195133	98.17	797	803
2015-16	212725	203020	95.44	227203	216721	95.39	1068	1067
2016-17	181712	173515	95.49	206960	197875	95.61	1139	1140
2017-18	97797	92293	94.37	130197	123654	94.97	1331	1340
2018-19	68784	65269	94.89	89306	84773	94.92	1298	1299
2019-20	60216	56054	93.09	89605	82934	92.55	1488	1480
2020-21	124286	118030	94.97	182705	173764	95.10	1470	1472

Source: Vital Agricultural Statistics, Government of Rajasthan

Table 2: Seed production of coriander variety RKD 18 under Agriculture University, Kota through CSS-MIDH (Centrally Sponsored Scheme-Mission for Integrated Development of Horticulture, Directorate of Arecanut and Spices Development, Kozhikode) funding

Year	Class of seed	Total coriander production (q)	Production of RKD 18 (q)	% Share of RKD 18	Approx. Revenue generated @ Rs. 7000/ qtl (Rs. in lakh)	Seed lifting agencies
2015-16	TL	318.86	35.80	11.22	2.51	Govt. of Raj. and farmers
2016-17	TL	375.72	265.86	70.76	18.61	Govt. of Raj. and farmers Private firms
2017-18	TL	172.83	150.07	86.83	10.50	Private firms, farmers
2018-19	TL	247.40	243.90	98.58	17.07	Private firms, farmers
2019-20	TL	185.81	185.56	99.86	12.99	Private firms, farmers
2020-21	TL	172.40	168.15	97.53	11.77	Private firms, farmers
2021-22	TL	250.19	247.04	98.74	17.29	Private firms, farmers
Total		**1723**	**1296**	**75.23**	**90.75**	

Source: University's internal official reports, annual progress reports of CSS-MIDH.

cloned to get the nucleotide sequences linked to a trait of interest. This genotype could be efficiently utilized in crop genetic improvement and breeding programs. Later, Choudhary *et al*. (2022) while studying genetic divergence in coriander genotypes also observed this genotype diverse from other genotypes and suggested inter cluster crossing among diverse genotypes for exploitation of heterosis for economically important traits.

Thus, the superiority of variety Pratap Raj Dhania 1 in various aspects has been reported by several other researchers also highlighting its suitability for mass multiplication. It can be hoped that increased seed production of this variety in larger area will help to fulfil the quality seed requirement of coriander farmers of Rajasthan.

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International Journal of Minor Fruits, Medicinal and Aromatic Plants. Vol. 9 (2) : 209-214, December 2023

SHORT COMMUNICATION

Status of tree genetic resources conserved at the Botanic Garden, Bangladesh Agricultural University

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Abstract

A total of 410 tree species under 66 families are conserved at the Botanic Garden, Bangladesh Agricultural University, which is about 40% of the total tree genetic resources of Bangladesh. According to IUCN Red List Categories and Criteria (Version 3.0) among the conserved tree species, 190 species are categorized as not evaluated (NE), 10 as data deficient (DD), 125 as least concern (LC), 25 near threatened (NT), 59 threatened and 1 extinct from wild (EW). These collections could be used as a source of materials for both species and habitat restoration programmes.

***Keywords*:** Bangladesh, IUCN red list, tree,

INTRODUCTION

A tree could be defined as a woody plant usually with a single stem growing to a height of at least two metres, or if multi-stemmed, then at least one vertical stem five centimetres in diameter at breast height (BGCI, 2021). Trees form the major structural components of forest ecosystems that cover approximately 31% of the world's land surface (FAO and UNEP, 2020), and are of vital importance ecologically, culturally and economically. They have many direct and/or indirect benefits for example, providing habitat for innumerable plant and animal species, and provide a host of other ecosystem services – ranging from timber to food and water, environmental processes like climate regulation, pollination, and air quality improvement, soil formation and rehabilitation, photosynthesis, and nutrient cycling, reduce flood damage and erosion, filter rainwater and improve water quality. Rainforests are some of the largest reservoirs for sequestering atmospheric carbon (Harris *et al*., 2021). People also enjoy a variety of aesthetic experiences, recreational activities, and associated well-being and health benefits in green areas (especially areas with trees). More importantly,the restoration of trees remains among the most effective strategies for climate change mitigation (Bastin *et al*., 2019). The total number of tree species on Earth is *ca*. 73,000, including roughly 9,000 yet not known to science (Gatti *et al*., 2022). Bangladesh lies in a transition between the Indo-Gangetic plains and the eastern Himalayas and, in flip, a part of the Indo-Chinese language sub-vicinity of the oriental realm (MoEF, 2004). This country thus acts as an important merging and sharing habitat, land bridge, and biological corridors of flora and fauna between these sub-regions, and is rich in tree genetic resources.The number of tree species has been reported from Bangladesh territory varied from 694 (BGCI, 2021) to a total of 1048 tree species (comprising both indigenous and introduced trees) under 432 genera in 99 families (Basak and Alam, 2015).

Botanic gardens are institutions holding documented collections of living plants and play major roles in plant conservation globally (Edwards and Jackson, 2019.). These have four common fundamental functions, including plant introduction and conservation, scientific research, recreational display, and popular science. Over the last few decades, the number of botanic gardens and their activities has grown remarkably worldwide.Recently, the botanic garden has been opinioned as one of the solutions to the plant

extinction crisis (Westwood *et al*., 2021). The Botanic Garden, Bangladesh Agricultural University (BAUBG) has emerged as a unique centre for plant conservation, education, scientific research, and information relating to plant biodiversity in the national and regional context (Rahman *et al*., 2017). Approximately 1800 species of vascular plants (including Pteridophytes, Gymnosperms, and Angiosperms) are conserved here. In this paper, I have reported a glimpse of the tree genetic resources harboured at the BAUBG and analyzed their present status according to IUCN Red List Categories and Criteria (Version 3.0) based on the "Developing National Red List of Plants" (Anon., 2023).

METHODOLOGY

The Botanic Garden, Department of Crop Botany, Bangladesh Agricultural University (BAUBG) is situated on the west bank of the Old Brahmaputra River and it lies between 24°43′27.9″ N and 90°26′28.2″ E. Living collections of the BAUBG tree genetic resources were illustrated with a total of 410 species. All the species had been physically verified and the nomenclature of each species has been updated by consulting "Plants of the World Online"<https://powo.science.kew.org/>. Their present status according to IUCN Red List Categories and Criteria (Version 3.0) based on the Fact Sheet of the "Developing Bangladesh National Red List of Plants and Developing Management Strategy of Invasive Alien Species (IAS) of Plants in Selected Protected Areas (PAs) Programme" implemented by the Forest Department (FD) and the Bangladesh National Herbarium (BNH) with the technical assistance of IUCN Bangladesh (Anon., 2023).

RESULTS AND DISCUSSION

A total of 410 species under 66 families are conserved at the BAUBG (Table 1), which is about 40% of the total tree genetic resources of Bangladesh (Basak and Alam, 2015). Among the families, Fabaceae was the largest represented by 33 species followed by Aracaceae by 30 species, and 18 families were represented by only single species each (Table 1). According to IUCN Red List Categories and Criteria (Version 3.0) out of these 410 species, 190 were not evaluated (NE), 10 data deficient (DD), 125 least concern (LC), 25 near threatened (NT), 59 threatened (of different categories) and 1 extinct from wild (EW) (a complete list available on request). The recent works of the FD and the BNH evaluated only 1000 selected plant (forest) species, out of approximate 6000 species in total, where 271 species are categorized as LC, 258 species DD, 69 species NT while the other 394 species are collectively termed Threatened, i.e., CR 5, EN 127 and VU 262 species (Anon. 2023). Many of tree species conserved at the BAUBG have not been under current assessment and categorized as NE. Among the threatened species, 46 are vulnerable (VU), 11 endangered (EN) and 2 critically endangered (CR) (Table 2). Although trees generally act as the source of wood or timber for different commercial uses for example furniture, construction materials, fuel & fibre, gum, dye, paper & pulping materials, etc., most of the threatened tree species are used in traditional medicine (Table 2).The GSPC (Global Strategy for Plant Conservation) target #8 required that "At least 75% of threatened plant species in *ex-situ* collections, preferably in the country of origin" (CBD, 2012). It emphasized both the evaluation of the status (and threat assessments) of biodiversity according to IUCN Red List Categories and Criteria, and the conservation of threatened plant species. Bangladesh is far behind the target (Anon., 2023). As an *ex-situ* conservatory, more priority should be given to the collection and conservation of threatened plant (tree) genetic resources at the BAUBG. These collections not only preserved plant genetic resources from extinction but also can provide a source of material for both species and habitat restoration.

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Table 1: Family-wise distribution of tree genetic resources conserved at the BAUBG.

Sl. No.	Family	Species (no.)	Sl. No.	Family	Species (no.)
1.	Alangiaceae	1	34.	Malvaceae	5
2.	Anacardiaceae	12	35.	Meliaceae	16
3.	Annonaceae	9	36.	Mimosaceae	18
4.	Apocynaceae	11	37.	Moraceae	28
5.	Aquifoliaceae	1	38.	Moringaceae	2
6.	Araucariaceae	4	39.	Myristicaceae	2
7.	Arecaceae	30	40.	Myrtaceae	17
8.	Bignoniaceae	11	41.	Ochnaceae	1
9.	Bixaceae	1	42.	Oxalidaceae	2
10.	Bombacaceae	4	43.	Phyllanthaceae	1
11.	Boraginaceae	3	44.	Pinaceae	2
12.	Burseraceae	4	45.	Poaceae	13
13.	Caesalpinaceae	1	46.	Podocarpaceae	3
14.	Capparaceae	1	47.	Primulaceae	1
15.	Casuarinaceae	1	48.	Proteaceae	1
16.	Celastraceae	1	49.	Punicaceae	1
17.	Clusiaceae	9	50.	Rhamnaceae	2
18.	Combretaceae	7	51.	Rhizophoraceae	6
19.	Cupressaceae	2	52.	Rosaceae	5
20.	Cycadaceae	4	53.	Rubiaceae	11
21.	Dilleniaceae	2	54.	Rutaceae	7
22.	Dipterocarpaceae	5	55.	Santalaceae	1
23.	Ebenaceae	5	56.	Sapindaceae	8
24.	Elaeocarpaceae	2	57.	Sapotaceae	6
25.	Euphorbiaceae	26	58.	Simaroubaceae	1
26.	Fabaceae	33	59.	Sonneratiaceae	3
27.	Fagaceae	5	60.	Sterculiaceae	8
28.	Flacourtiaceae	2	61.	Tetramelaceae	1
29.	Lauraceae	10	62.	Theaceae	2
30.	Lecythidaceae	3	63.	Thymelaeaceae	1
31.	Loganiaceae	1	64.	Tiliaceae	4
32.	Lythraceae	5	65.	Ulmaceae	1
33.	Magnoliaceae	5	66.	Verbenaceae	9

Table 2: List of threatened species conserved at the BAUBG. VU Vulnerable, EN Endangered, CR Critically Endangered, EW Extinct in the Wild.

Sl. No.	Local Name	*Botanical Name*	Family	Status	Ethnobotanical Uses*
1.	Khalisha, Khalshi	*Aegiceras corniculatum* (L.) Blanco	Primulaceae	VU	Medicinal. wood for knife handles, fuel
2.	Boilam, Boilsur	*Anisoptera scaphula* (Roxb.) Kurz.	Dipterocarpaceae	VU	Timber (general) light construction
3.	Bankanthal, Dewa	*Artocarpus lacucha* Buch.-Ham.	Moraceae	VU	Fruit edible, medicinal, tannin, fibre, termites-resistant timber
4.	Mitings bash	*Bambusa burmanica* Gamble	Poaceae	VU	Roofing, thatching, construction, basket making
5.	Rakta Kanchan	*Bauhinia variegata* L.	Fabaceae	VU	Medicinal, buds as vegetables,*wood wool board, tannin, oil, gum, fibre*
6.	Bixa, Doirong	*Bixa orellana* L.	Bixaceae	VU	Colourant, medicinal
7.	Mailaam	*Bouea oppositifolia* (Roxb.) Meissner	Anacardiaceae	VU	Fruits food & vegetables, ornamental, Timber & Products
8.	Maj, majot	*Brownlowia elata* Roxb.	Tiliaceae	VU	Medicinal
9.	Kumbhi	*Careya arborea* Roxb.	Lecythidaceae	VU	Fruits edible, medicinal, dye, gum, tannin, wood
10.	Fish Tail Palm	*Caryota mitis* Lour.	Arecaceae	VU	Ornamental, making sago, construction materials
11.	Mon kata	*Catunaregam spinosa* (Thunb.) Tirveng.	Rubiaceae	VU	Medicinal
12.	Ram-tejpata	*Cinnamomum bejoghota* (Buch-Ham.) Sweet	Lauraceae	VU	*Condiment, medicinal, source of essential oil and wood.*
13.	Tejbal, Gondroi	*Cinnamomum glaucescens* (Nees) Meiss.	Lauraceae	VU	Timber, essential oil for perfumery and medicine
14.	Adagach, Dudhia	*Croton persimilis* Müll.Arg.	Euphorbiaceae	VU	Medicinal, ornamental
15.	Ashphal, Katlitchu	*Dimocarpus longan* Lour.	Sapindaceae	VU	Fruits food & vegetables, medicinal, tough, very hard, highly durable wood, fuel
16.	DhuliGarjan	*Dipterocarpus gracilis* Blume	Dipterocarpaceae	VU	Commercial grade plywood, *resin used* in paint oils, coat for waterproofing paper
17.	Ban Bokul	*Drypetes assamica* (Hook.f.) Pax & K. Hoffm.	Euphorbiaceae	VU	Fruits food, wood-making charcoal
18.	Rangirata, pitraj	*Dysoxylum binectariferum* (Roxb.) Hook.f. *ex* Beddome	Meliaceae	VU	Medicinal
19.	Khuskadumur	*Ficus hirta* Vahl	Moraceae	VU	Fruit edible, medicinal
20.	Ficus	*Ficus variegata* Blume	Moraceae	VU	Fruit edible, medicinal, latex, fuel wood
21.	Jarul	*Lagerstroemia macrocarpa* Wall.	Lythraceae	VU	Medicinal, timber of excellent quality
22.	Jarul (Nil)	*Lagerstroemia parviflora* Roxb.	Lythraceae	VU	Medicinal, edible gum, very hard and durable timber
23.	Ruffled Fan Pam	*Licuala grandis* (T.Moore) H.Wendl.	Arecaceae	VU	Ornamental, construction materials
24.	Batna, Kalibatna	*Lithocarpus fenestratus* (Roxb.) Rehder.	Fagaceae	VU	Fuel wood
25.	Raktan	*Lophopetalum wightianum* Arn.	Celastraceae	VU	Medicinal, timber *used* as perupok
26.	Dulichampa	*Magnolia pterocarpa* Roxb.	Magnoliaceae	VU	Wood suitable for tea boxes, firewood
27.	Khirni	*Manilkara hexandra* (Roxb.) Dubard	Sapotaceae	VU	Fruit edible, medicinal, rootstocks for sapodilla
28.	Phul Kadam	*Mitragyna diversifolia* (Wall. *ex* G. Don) Havil.	Rubiaceae	VU	Medicinal
29.	Phuti/Keli Kadam	*Mitragyna parvifolia* (Roxb.) Korth. var. *microphylla* (Kurz) Ridsdale	Rubiaceae	VU	Medicinal, increase quantities of breast milk in lactating mothers, lactodepurant, wood
30.	Bontejpata	*Neolitsea cassia* (L.) Kosterm.	Lauraceae	VU	Medicinal, building houses, planks, and rafters
31.	Duddahgach, Tali	*Palaquium polyanthum* (Wall. *ex* DC.) Engl.	Sapotaceae	VU	*House building, making boats, plywood flush doors & black boards, latex*
32.	Kuki-tetui	*Parkiatimoriana* (DC.) Merr.	Mimosaceae	VU	Medicinal, ornamental, firewood and lumber
33.	Ban-kanchan	*Piliostigma malabaricum* (Roxb.) Benth.	Fabaceae	VU	Medicinal

Continued.

Sl. No.	Local Name	*Botanical Name*	Family	Status	Ethnobotanical Uses* *Continued. Table 2.*
34.	Ram Gua	*Pinanga gracilis* Blume	Arecaceae	VU	Fruit as masticator
35.	Pine	*Pinus kesiya* Royle *ex* Gordon.	Pinaceae	VU	Fruit edible, medicinal, green dye, woods forboxes, paper pulp, temporary electric poles
36.	Ceylon cherry	*Prunus ceylanica* (Wight) Miq.	Rosaceae	VU	Medicinal (in Sri Lanka)
37.	Putrajhiva	*Putranjiva roxburghii* Wall.	Euphorbiaceae	VU	Medicinal
38.	Momchina	*Sapium sebiferum* (L.) Roxb.	Euphorbiaceae	VU	Medicinal, fat for soap and candle, wood for*making furniture and incense*
39.	Joyna	*Schleichera oleosa* (Lour.) Oken.	Sapindaceae	VU	Edible leaves, shoot & fruits, Medicinal, oil, tannin, dye, wood
40.	Bhela	*Semecarpus anacardium* L.f.	Anacardiaceae	VU	Food, medicinal, oil, gum, making permanent marking ink
41.	Kuchila	*Strychnos lucida* R.Br.	Loganiaceae	VU	Medicinal, fish poison
42.	Khudi jam	*Syzygium cymosum* DC.	Myrtaceae	VU	Dye from bark,wood for making panels
43.	Hatiyal, Harra	*Terminalia citrina* (Gaertn.) Roxb. *ex* Fleming	Combretaceae	VU	Medicinal, source of tannins and wood.
44.	BelatiTentul	*Vangueria madagascariensis* Gmel.	Rubiaceae	VU	Fruit food, medicinal, wood
45.	Dudkarach, Dudhi	*Wrightia arborea* (Dennst.) Mabb.	Apocynaceae	VU	Medicinal, Timber
46.	Dhdhi, Pallam	*Wrightia coccinea* (Roxb.) Sims.	Apocynaceae	VU	Ornamental, medicinal, fire wood
47.	Chatim (Big)	*Alstonia macrophylla* Wall. *ex.* G. Don.	Apocynaceae	EN	Medicinal, construction materials
48.	Berria	*Berrya cordifolia* (Willd.) L.Laurent	Malvacea	EN	High-quality timber, fibre
49.	Dhup	*Canarium resiniferum* Brace *ex* King.	Burseraceae	EN	Medicinal, resin
50.	Mussanda (White)	*Canthium glabrum* Blume	Rubiaceae	EN	Medicinal
51.	Cycad- Moniraj	*Cycas pectinata* Buch.-Ham	Cycadaceae	EN	Food & vegetable, medicinal, making sago, glue
52.	Monther	*Erythrina suberosa* Roxb.	Fabaceae	EN	Ornamental, medicinal, fuel
53.	Swarna khiri	*Garcinia morella* (Gaertn.) Desr.	Clusiaceae	EN	Fruits food & vegetable, medicinal, resin, rootstock for mangosteen
54.	Thoikar	*Garcinia pedunculata* Roxb. *ex* Buch-Ham.	Clusiaceae	EN	Fruits food & vegetables, medicinal, timber used for planks, beams, etc.
55.	Bharal	*Intsia bijuga* (Colebr.) O. Kuntze.	Fabaceae	EN	Medicinal, constructing the main hull, masthead, maststep and steering oar of ocean-going canoes.
56.	Bongajari	*Miliusa tomentosa* (Roxb.) J. Sinclair	Annonaceae	EN	Fruits food, medicinal
57.	Boncula	*Pterospermum lanceifolium* Roxb. *ex* DC.	Malvaceae	EN	Wood, fuel
58.	Chalmogra	*Hydnocarpus kurzii* (King) Warb.	Flacourtiaceae	CR	Medicinal
59.	Banspata	*Podocarpus nerifolius* D. Don	Podocarpaceae	CR	Fruit edible, medicinal, wood used for construction, making furniture, musical instruments, carvings, paper
60.	Tali palm	*Corypha taliera* Roxb.	Arecaceae	EW	*Leaves as writing paper, ornamental*

* <https://mpbd.cu.ac.bd/index.php>; <https://tropical.theferns.info/>; <https://www.wikipedia.org/>; <https://indiabiodiversity.org/>;<https://www.natureinfo.com.bd/mpb/>

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SHORT COMMUNICATION

Effect of Indole-3- butaric acid on air-layering in jackfruit under the Sub-Himalayan Terai region of West Bengal

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ABSTRACT

The present experiment was conducted to study the effect of various concentrations of Indole-3- butaric acid (IBA) on air-layering in jackfruit during 2020-21.Five IBA concentrations,viz.,T_1:Control (0 ppm), T_2: 1000 ppm, T_3: 5000 ppm, T_4: 10000 ppm and T_5: 15000 ppm were used. Results showed that,T_4(10000 ppm IBA) had the highest, percentage of rooted layers (82.50 %), length of the root (6.17 cm), diameter of longest root (1.50 mm), and number of roots (primary and secondary). Although 10000 ppm IBA had the minimum number of days taken to rooting (33.40 days), but it also had the maximum fresh (0.98 g) and dry weight (0.48 g) of roots. Therefore, it is concluded that use of 10000 ppm IBA showed the highest performance of air-layers in jackfruits.

***Keywords*:** Air layering, Indole-3- butaric acid (IBA)**,** Jackfruit, primary roots.

INTRODUCTION

Jackfruit (*Artocarpus heterophyllus* Lam.) is one of the indigenous fruits of India and comes under the family Moraceae. The immature or mature unripe fruits are used as popular vegetable and the ripe fruits are used as table purpose fruit. Jackfruit is a good source of nutrients such as starch, protein, and minerals (Ocloo *et al*., 2010). Jackfruit is considered as the national fruit of Bangladesh, and is a plant of multiple uses as food for all the ages, quality timber for furnitures, fodder for cattle, fuel, preparation of medicinal and industrial products. Due to its long seasonal availability as well as low price, it is called "*Poor Mans' Fruit"* in India. A mature tree produces up to 700 fruits per year, each weighing 0.5 to 50 kg. Despite the region having enormous potential for commercial cultivation of jackfruit, however, it is still considered an underutilized fruit crop, and its commercial adoption is still at a primitive stage. Universally, jackfruit is propagated mostly by the fresh seeds and seedlings may take 8-10 years to bear fruits with great variability due to cross pollination behavior of the plant. On contrary, the vegetatively propagated jackfruit plants require only 5-6 years to come into commercial bearing stage. Air layering is one of the easy and quick methods among all vegetative methods of propagation (Tomar, 2011).

Layering is a method of propagation where generation of adventitious roots is forced in the plant parts while they are still attached to mother plants. Indole-3- butaric acid (IBA) is a most commonly used hormone used to induce formation of adventitious roots in cuttings or air layers that helps in quick and better field establishment, huge growth of the plants and early floral bud formation (Singh, 2002).The media used for rooting in air layers and wrapping materials causes variation in the time required for root emergence, number of adventitious roots, root thickness and root length in air layers (Alam *et al.,* 2004). Considering beneficial effect of IBA in rooting in air layering and to generate uniform planting material in jackfruit, an experiment was undertaken to find out the effect of different concentrations of Indole-3-butaric acid (IBA) on success of air-layering in jackfruit under the Sub-Himalayan Terai region of West Bengal.

The experiment has been carried out at the Instructional Farm, Department of Pomology and Post-Harvest Technology, Uttar Banga Krishi Viswavidyalaya, Pundibari, Cooch Behar, West Bengal, India during June-July, 2020. The Instructional Farm comes under the eastern Sub-Himalayan plains at 81°66′73′′ E longitude

crossing 28°58′86″ N latitude which remains at an elevation of 42m above mean sea level. The climate of the region is subtropical with high humidity, high rainfall, and a prolonged winter. Broadly, there are two dominant seasons in a year: the long rainy season and dry rabi, or extended winter season. The minimum and maximum temperature of this location varies from 7.1–8.0°C and 24.8–32.2°C respectively. The soil is coarse textured sandy loam in nature with poor water holding capacity, rich in organic carbon and contains high available nitrogen.

A shoot length of 30-45cm and 1.0-1.25 cm thick (pencil thickness) of 1-2year old branches were selected. Defoliated the base of the shoot and then the stem was girdled by removing a bark about 2-3cm wide at the base. The upper bark cut portion was treated with five different IBA concentrations, viz., T_1-Control (0 ppm), T_2- 1000 ppm, T_3- 5000 ppm, T_4 - 10000 ppm and T_5 - 15000 ppm. The open wood is then covered with hand full of moist soil mixture (soil: well rotten cow dung:: 3:1) and wrapped with 20-25 cm polyethylene sheet (200-300 gauge). The two ends were then tied using jute thread. The technical procedure for air layering was followed as suggested by the method of Bhowmick *et al*. (2023).

The required quantity of IBA hormonal powder was weighed with the help of an electronic balance. Then it was dissolved in the required amount of rectified spirit in a beaker. This material was poured and thoroughly mixed with the glass rod. After mixing, the mixture was kept in the air for a few hours, which helped evaporate the alcohol/spirit. The dried talc, along with the hormone was ground into a fine powder. This fine powder was stored in an air tight container to avoid moisturizing, and it was applied to selected air layered shoots of the jackfruit plant. The concentration of IBA was expressed as ppm (parts per million) which is equivalent to milligram of solute per litre of water or per kilogram of powder. Properly rooted air layers have been detached from the mother plant at two months after performing the air-layering and transferred to plastic pots kept under partial shade in the fruit nursery. Majority of the leaves were removed from the layers to reduce the transpiration loss. The layers were irrigated immediately after planting. After detaching the rooted layers, 10 layers are randomly selected from each replication for recording the data (Ezekiel *et al*., 2016; Kumar, 2000).

The experiment has been carried out in a Randomized Block Design (RBD) with five treatments and four replications. Each replication has consisted 30 air-layers and a total of 120 air-layered jackfruit plants has been considered per treatment. The data collected from different treatments were analysed with the help of OPSTAT statistical software as designed for randomized block design (RBD) as described by Panse and Sukhatme (2000).

The results indicated that the application of various concentrations of IBA significantly enhanced the rooting percentage and rooting attributes (Table 1). The application of 10000 ppm IBA significantly gave highest rooting success of 82.50 % followed by 5000 ppm IBA (75.00 %), while the minimum rooting success was in control(63.33). The longest root was recorded in T_4 (6.17 cm) followed by T_3 (5.62 cm). Whereas, the lowest root length was recorded in T_1 (4.36 cm). The results from the Table 1 indicates that the maximum diameter of the longest root was recorded in T_4 (1.50 mm) followed by T_3 (1.45 cm) as compared to control (T_1). The maximum numbers of primary and secondary adventitious roots were observed in T_4 followed by T_3 and minimum was observed in T_1. This result may be for the use of Indole-3-butyric acid which may stimulate the translocation of photosynthates from the leaves to root growing zone and thus encourage the growth and development of air-layered jackfruit plants to produce quality planting materials (Rymbai and Reddy, 2010). The combination of IBA @ 5000 ppm and NAA @ 5000 ppm has also showed the best effect on the rooting of the air layers of jackfruit as reported by Singh and Singh (2004). Previously, it has been shown that the application of IBA at different concentrations significantly increases the rooting, number of primary, secondary, and tertiary roots as compared to the control (0 ppm IBA) treatment. Tomar (2011) has found that the use of IBA @ 10000 ppm has resulted higher percentage of rooted air layers and a maximum survival percentage in jackfruit when layering done during the month of July.

Table 1: Effect of different IBA concentrations on rooting success and root growth in air-layering of jackfruit

Treatments (Concentration of IBA)	Success percentage of rooted layers (%)	Length of the longest root (cm)	Diameter of the longest root (mm)	Number of primary and secondary roots	Days to rooting	Root fresh weight (g)	Root dry weight (g)	Length of the new growth (cm)
T_1 -Control (0 ppm)	63.33	4.36	1.28	34.75	56.45	0.68	0.22	1.50
T_2-1000 ppm	67.50	5.21	1.34	43.00	50.90	0.75	0.31	2.02
T_3 -5000 ppm	78.33	5.62	1.45	52.25	37.10	0.96	0.45	2.88
T_4 -10000 ppm	82.50	6.17	1.50	62.75	33.40	0.98	0.48	3.38
T_5 -15000 ppm	75.00	5.44	1.39	48.50	45.40	0.81	0.34	2.39
S.Em.(±)	**0.98**	**0.82**	**0.01**	**0.77**	**0.88**	**0.02**	**0.01**	**0.05**
CD (0.05)	**3.05**	**2.54**	**0.03**	**2.41**	**2.73**	**0.05**	**0.03**	**0.15**

The minimum days taken to produce rooting was recorded in T_4 (33.40 days) followed by T_3 (37.10 days). Whereas, maximum days taken to produce rooting was recorded in T_1 (56.45 days). The results indicated that root fresh and dry weight was significantly affected by different concentration of IBA in jackfruit (Table 1). The maximum root fresh weight (0.98 g) was recorded in T_4 followed by T_3 (0.96 g) as compared to T_1 (0.68 g). The maximum root dry weight (0.48 g) was recorded in T_4 followed by T_3 (0.45 g), T_5 (0.34 g), T_2 (0.31 g). Whereas, the minimum root dry weight (0.22 g) was recorded in T_1 (0 ppm IBA).The maximum length of the new growth (3.38 cm) was recorded in T_4 followed by T_3 (2.88 cm), T_5 (2.39 cm), T_2 (2.02 cm). Whereas, the minimum length of the new growth (1.50 cm) was recorded in T_1 (0 ppm IBA).The increase in shoot and root biomass with the use of auxins is consistent with the findings of Chander and Kumar (2023). Application of Indole-3-Butyric Acid (IBA) and using proper rooting for air layering of wax apple has been resulted the maximum root initiation, growth of the layers and their survivability. Khandaker *et al.* (2022) reported that the application of IBA with rooting media, promoted the formation of adventitious roots, increased chlorophyll content in leaves, higher vegetative growth and better survival rate of air layers of wax apple.

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SHORT COMMUNICATION

Structure, area distribution and condition of pistachio forests in Kyrgyzstan

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ABSTRACT

The article provides information on the distribution of the area of pistachio forests by altitude, slope exposures, age classes is given. Growing on slopes of different exposures, pistachio and sparse forests fulfill soil-protective, water-protective and water-regulating role. The diversity of pistachio forms allows selecting the best forms for introduction into culture. Pistachio forests serve as a source of valuable nuts for the local population. In harvest years, the collection of pistachio fruits brings good income to the local population. However, it is necessary to work together with the local population on sustainable use of pistachio plantations and preservation of their biodiversity, as there is a negative impact of anthropogenic factors. The data on the impact of major pests and diseases of pistachio plantations are presented. Due to the influence of pests and forest diseases, the yields are reduced and the condition of pistachio forests deteriorates. Timely and effective control measures against forest pests and diseases are necessary. Analysis of the current state and solution of the issues of biodiversity conservation of pistachio plantations is of great scientific and practical importance.

***Keywords*:** Biodiversity, forest areas, pests, pistachio diseases. pistachio forests, yield

INTRODUCTION

The total area of forests in the world is 4.06 billion hectares, or 31% of the total land area and contain more than 80% of terrestrial biodiversity (FAO, 2020). According to the Global Forest Resources Assessment (FAO, 2020) 398 million hectares of protected forests cover 12 percent of the world's total forest area. In four countries, the share of total forest area with soil and water protection functions exceeds 90 percent - Kiribati and Kuwait (both at 100 percent), Cape Verde (98 percent) and Kyrgyzstan (92 percent). The Kyrgyz Republic is a mountainous low-forest country and about 90% of the area is at an altitude of more than 1500 meters above sea level (Okruzhaiushaia sreda v Kyrgyzskoi Respublike- 2015-2019, 2020). All forests of the Kyrgyz Republic are under state ownership and are national wealth. The total area of the state forest fund is 2,619.7 thousand hectares. These forests store the gene pool of many species of not only tree and shrub species, but also herbaceous plants, as well as a source of income for the local population and play a key role in the economy of the country.

Of the 4,500 species of plants in the State Forest Fund of Kyrgyzstan, 300 wild plants are rare and endangered, 125 species are endemics, 200 are medicinal plants, and more than 180 species are woody and shrubby plants that make up the forests of the Kyrgyz Republic. In addition, more than 65% of the total composition of endemic plants grows in the forest area. They constitute the main core of plant communities of forest ecosystems of the Republic, and they cannot be replaced by foreign species (Murzakmatov *et al.,* 2016). According to remote zoning data obtained as a result of space images processing, about 1.2 million people live at a distance of up to 5 km from the forest and use its resources. About 200 thousand people live inside the forest and are fully dependent on forest resources. The population's needs for firewood and construction material are growing every year, it was found that one family uses about 5-10 cubic meters

of firewood for heating and cooking per year (Bikirov *et al*., 2018).

One of the most characteristic for the lower part of the nut-fruit forest belt are sparse forests formed by pistachio (*Pistacea vera* L.) and almond (*Amygdalis* sp.) with a total area of 45840 hectares (Okruzhaiushaia sreda v Kyrgyzskoi Respublike-2015-2019, 2020). Dry growing conditions are the main reason for the sparseness of pistachio plantations (Mamadzhanov and Kenzhebaev, 2022). In harvest years forestry enterprises and local people collect mountain fruits. Walnut over 2000 tons; pistachio 52 tons; wild apples within 5000 tons (Toktoraliev and Attokurov, 2009). Currently, the state of nut and fruit forests is relatively stable, but there are localized threats such as unsystematic grazing (Wilson *et al*., 2019), excessive harvesting of medicinal and technical plants, still lead to forest degradation.

METHODS

Field surveys were conducted to assess the structure, area distribution and phytosanitary status of pistachio forests in Zhalal-Abad, Osh and Batken regions of the Kyrgyz Republic. The coordinates of the sample area points were determined using a Global Position System (Garmin GPS) device.

RESULTS

According to FAO (FAO, 2022), the world production of pistachios was more than 915717 tons. The leading pistachio producing countries are USA, Iran, Turkey and account for 84.9% of the total volume. According to statistics, Turkey produces 144000 tons of pistachio annually (Mikdat Simsek, 2018). Pistachio, *Pistacia vera* L, is one of the main forest-forming species of walnut-fruit forests. The total area of pistachio forests is 34403.3 ha (of which 4622 ha is outside the territory of the state forest fund) or 3.4% of the total forest area of Kyrgyzstan and is concentrated on the south-western slopes of the Fergana Range and Chingir-Tash mountain massif. Pistachio forests of Kyrgyzstan have a huge gene pool that allows selecting the most productive, stable and economically valuable forms, which are not inferior in nut quality to cultivated pistachio varieties. The study of form diversity of pistachio forests of Kyrgyzstan makes it possible to select high-yielding economically valuable forms promising for creation of highly productive plantations. The form diversity of pistachio can be classified according to the shape of the crown, fruit brushes and nuts (fruits), pericarp coloring, yield, nut size and openness, etc.

Pistachio trees are found with spreading, globular, compact and occasionally "weeping" crowns. Pistachio trees with a spreading crown form are found on gentle slopes at an altitude of 800-1400 m above sea level. They are characterized by relatively good growth and yield. Globe-shaped crowns are formed by single-stem (boll) pistachio trees, and they are rarely found. Compact (umbrella-shaped) crown form is found in all types of pistachio trees. Very rarely found trees "weeping" crown form grows on more fertile soils at an altitude of 1000-1200 m above sea level. Basically, the nuts of the true pistachio are divided into 3 groups: globular, oval, and elongated-oval. One of the qualitative indicators is nut openness. The character of nut opening can be up to the base, up to the middle of the knuckle and on the top. The average openness of pistachio nuts is 66% in Southern Kyrgyzstan, and in the north of Kyrgyzstan it is 37-22% (Bolotov and Kenzhebaev, 2008). Pistachio forests in Kyrgyzstan are mainly represented by middle-aged plantations-23.7 thousand ha or 65 % of the total area (Table 1).

Pistachio forests in Kyrgyzstan are distributed at altitudes from 720 meters to 1900 meters above sea level, but sometimes there are isolated pistachio thickets at an altitude of 450 meters above sea level. Optimal altitudes for pistachio are from 700-1500 meters above sea level. At these altitudes (700-1500 meters above sea level) pistachio develops normally and gives a good harvest. Pistachio forests in Kyrgyzstan are mainly distributed in southern exposures, the area of which is 13903.2 hectares or 40.4 percent of the total area of pistachio forests (Fig. 1). The distribution of natural pistachio forests on the slopes of southern exposures indicates its drought resistance and adaptation to harsh soil conditions.

Unfortunately, despite the exceptionally high economic and economic value, pistachio forests are currently in unsatisfactory condition, especially adjacent to populated areas due to anthropogenic

Table 1: Distribution of pistachio plantations in Kyrgyzstan by age classes

Prevailing tree and shrub species	Total (ha)	by age group (ha)					
		Juvenile		Middle aged	Early-mature	Mature and over mature	
		I class	II class			total	Including over mature
Pistachio	**34403.3**	**1989,3**	**5494**	**16350,5**	**5360,3**	**4096,1**	**1108,8**

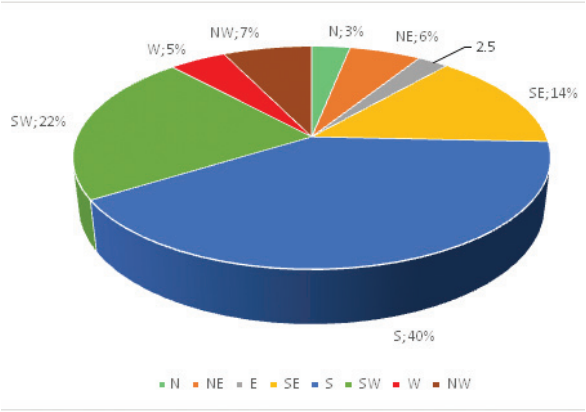

Fig. 1: Distribution of pistachio trees by exposures (%)

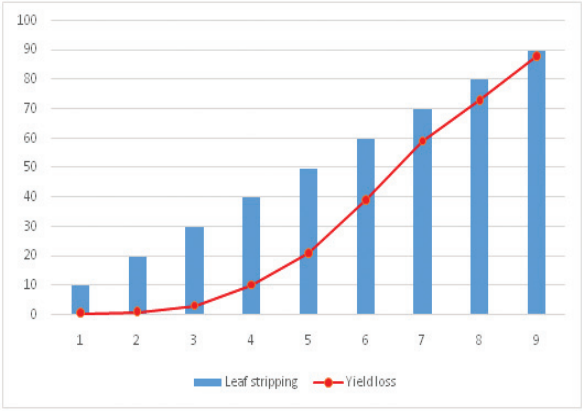

Fig. 2: Yield losses of pistachio as a function of the degree of leaf stripping

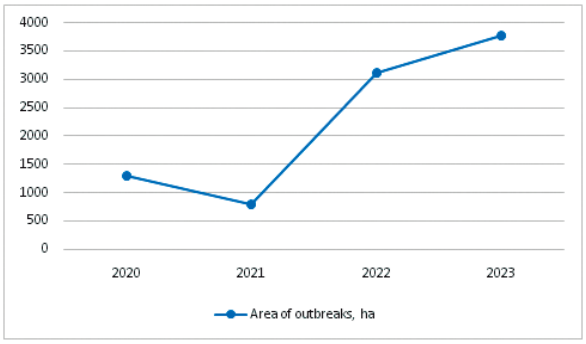

Fig. 3: Dynamics of gypsy silkworm foci in pistachio plantations

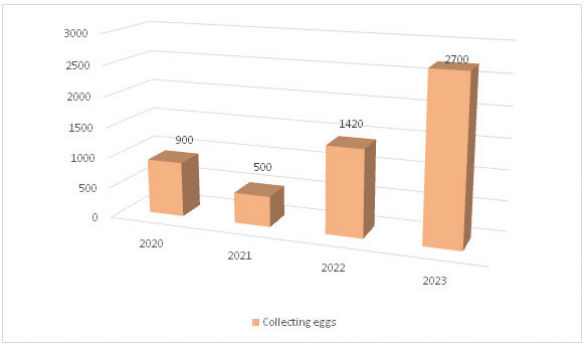

Fig. 4: Collection of gypsy moth egg-layers in pistachios

pressure. Grazing in pistachio forests destroys natural regeneration of pistachio and unsystematic grazing worsens soil water permeability and creates conditions for erosion processes. As a result of unregulated grazing the area subject to erosion expands. Low productivity of pistachio plantations is explained not only by insufficient care, damage from pests and diseases, but also by the ratio of male and female individuals. Male non-fruiting individuals of pistachio occupy more than 50%, *i.e.* the ratio of female and male individuals is close to 1:1.75 sometimes 1:2 (Bolotov and Kenzhebaev, 2008, Kenzhebaev *et al.,* 2019).

Besides, pests and diseases take a noticeable toll on the pistachio forest crop. Some damage the pistachio kernels, and some of them feed exclusively on leaf tissue, and if the leaf laminae are severely damaged, the tree becomes noticeably weaker, which affects yields. The main pests of pistachio are gypsy moth and pistachio moth. The gypsy moth (*Lymantria dispar* L.), in conditions of Kyrgyzstan annually gives mass outbreaks and is the most dangerous pest not only of pistachio

plantations, but also of other forest-forming tree species of nut and fruit forests. From the harmful activity of the gypsy silkworm forestry and local population annually suffer huge economic losses. The gypsy silkworm is found in foothill and mountainous areas at an altitude of 700 to 2000 m above sea level. The most extensive areas of outbreaks are observed in the lower, pistachio zone (Ashimov, 1989; Orozumbekov, 2001; Orozumbekov *et al*., 2005; Teshebaev, 2008, Toktoraliev *et al*., 2021).

Below we present yield losses of pistachio from harmful activity of gypsy moths (Fig. 2). Losses of pistachio yield depending on the degree of leaf stripping up to 30% are not significant, i.e. up to 3%, and with complete stripping the yield is reduced by 90%, and sometimes lost completely (Ashimov *et al*.,1998).

According to the data of Zhalal-Abad forest protection station, in recent years the area of mass outbreaks of gypsy silkworm ranges from 1300 to 3785 hectares (Fig. 3). The palatine silkworm periodically gives outbreaks of mass reproduction migrating along vertical height from pistachio sparse forests to upper nut plantations. Measures to control gypsy moths are carried out by collecting eggs and pupae, using entomophages and biological preparation Virin-ENSh(k).

In order to reduce the number of the pest, forestry workers and forest users annually collect several hundred kg of gypsy silkworm eggs, in 2020 collected 900 kg, and 2023 it was 2700 kg (Fig. 4).

The biological preparation (Toktoraliev and Attokurov, 2009) which is a liquid viral insecticide of domestic production, is also used against the most dangerous pest. The preparation has no toxicity and is not pathogenic for warm-blooded animals, bees, etc. Oviposition of ovipositors is carried out by wetting or spraying. Entomaphages are used against gypsy moths. The composition of natural enemies of entomophages is diverse. Currently, as the main entomophagous against the gypsy silkworm, the forest caterpillar hunter (*Calosoma sycophanta* L.) from the family *Carabidae* L. Annually, more than a thousand beetles (*Calosoma sycophanta* L.) are bred in the Zhalal-Abad forest protection station and they are used to exterminate larvae and pupae of the gypsy silkworm. The beetles and larvae of the odoriferous beetle are very mobile and during the growing season they destroy up to 200-300 caterpillars and up to 20 pupae of the gypsy moths (Sultanov,1998). The use of biological preparations and entomophages to protect pistachios is the safest for forest ecosystems and a promising way to reduce the number of pests.

Pistachio leaf and fruit septoriosis is one of the most common diseases in all pistachio growing areas (Uda Nezar Muhamad,1989; Gusella *et al.,* 2021). Pistachio forests in Kyrgyzstan are often affected by septoriosis (causative agent Septoria pistacina Allescll.), which negatively affects yield and fruit quality. The area affected by septoriosis in 2020 amounted to 1220 hectares, and in 2023 it was 2212 hectares. At first, small brown spots appear on affected pistachio leaves. As the disease develops, the spots gradually grow and cover the entire leaf lamina. In the conditions of Jalal-Abad oblast, the first signs of septoriosis are observed in mid-May. In July, mass infection is observed and leaves shrivel and fall off, as a result, the process of photosynthesis is disturbed, yield (incomplete kernel development) and annual growth is reduced, which in turn affects the future harvest.

Preservation of biodiversity of pistachio plantations, improvement of their condition and sustainable use, increase of yields, expansion of areas through the establishment of industrial plantations, protection and defense against forest pests and diseases are the main tasks of forest science, forestry and forest users.

CONCLUSIONS

1. Pistachio forests of Kyrgyzstan is one of the valuable forest areas and by the occupied area and diversity of forms is one of the large forest areas in Central Asia.
2. The main massifs of pistachio forests are spread below the belt of walnut forests at an altitude of 700 to 1400 m above sea level.
3. In terms of age, middle-aged pistachio plantations prevail, but there are areas of overmature plantations where reforestation measures should be taken.
4. Many areas of pistachio forests are distributed on slopes of southern exposures and therefore pistachio can be cultivated in arid areas.

5. It is necessary to establish new plantations and increase the yield of pistachio plantations.
6. Improve the effectiveness of forest pest and disease control measures using biological and modern control methods.

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SHORT COMMUNICATION

Identifying the effects of climate change on fruit production and creating resilience techniques to reduce environmental challenges

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ABSTRACT

The global fruit business faces considerable obstacles from climate change, which affects the development, growth, and productivity of fruit plants. The main causes of these disruptions are rising temperatures, changing precipitation patterns, and an increase in the frequency of extreme weather events. Temperature changes cause phenological stages to be disrupted, which results in a mismatch in the timing of blooming and pollination, which reduces fruit set and yield. Changes in precipitation patterns have an impact on soil moisture availability, which lowers fruit quality and makes plants more vulnerable to insect and disease infestations. Extreme weather also harms trees physically, reducing their ability to produce fruit and even killing them. The fruit sector is implementing adaptable solutions to deal with these issues, such as the development of climate-resilient cultivars, the application of cutting-edge irrigation methods, and the improvement of pest and disease management procedures. Utilizing remote sensing and data analytics, precision agricultural technologies optimize resource allocation and enhance crop management choices.

***Keywords*:** Adaptive strategies, climate change, fruit industry, phenological stages

INTRODUCTION

Climate change is a serious issue that has repercussions across many industries, including agriculture. As a result of the effects of changing climatic patterns, fruit production is particularly susceptible, demanding proactive efforts to adapt to the changing environment (Smith *et al.,* 2022). Global fruit farming faces substantial problems due to rising temperatures, changed precipitation patterns, and an increase in the frequency of extreme weather events (IPCC, 2021). In order to secure the sustainability and resilience of fruit production in the face of climate change, stakeholders in the fruit sector are increasingly focusing on creating adaptive strategies. The impact of climate change on agriculture, including fruit production, is becoming increasingly evident and urgent.

The creation and application of adaptive solutions are required in light of the effects these changes have on fruit yields, quality, and overall sustainability (Johnson *et al.,* 2023). Rising temperatures are a substantial contributor to the observed effects of climate change on fruit output. Increased temperatures alter phenological stages like flowering and fruit ripening by affecting the growth, development, and productivity of fruit trees. According to Jones *et al.* (2023), this may throw off the timing of plants and pollinators, potentially resulting in decreased fruit set and output. The production of fruit is also hampered by altered precipitation patterns. Fruit quality, size, and flavour are all impacted by variations in rainfall distribution and intensity. Production problems might be made worse by water stress or excessive irrigation needs brought on by changed precipitation patterns. Drought circumstances make fruit more vulnerable to pests and illnesses, which increases the hazards (Smith *et al.,* 2023b). Fruit production is further endangered by the increased frequency and severity of extreme weather phenomena like heat waves, storms, and frosts.

Fruit growers may suffer large financial losses as a result of these occurrences' physical damage to plants, which can induce production reductions and tree death (Brown and Johnson, 2022a). The fruit sector is actively putting adaptive techniques into practice in response to these difficulties. These tactics include the use of cultivars that are climate-resilient, improved irrigation methods, and improved pest and disease management procedures. In order to maximize resource utilization and enhance crop management choices, precision agricultural technologies like remote sensing and data analytics are also being used (Garcia *et al.,* 2023a). To increase the resilience of fruit production systems, orchard management practices must incorporate climate change considerations. To find and spread best practices, create region-specific adaption plans, and guarantee the long-term viability of fruit production in the face of a changing climate, researchers, farmers, and policymakers must work together.

Effect of climate change (Body)

Temperature fluctuation

Fruit trees may experience changes in their phenological stages, such as flowering and fruit ripening, as temperatures rise. A mismatch between pollinators and flowering times might result from this, which may impair fruit set and yield (Jones *et al.,* 2023b). For instance, greater wintertime temperatures in apple orchards may interfere with the necessary period of dormancy, resulting in inconsistent bud break and decreased fruit output (Kumar *et al.,* 2022). High fluctuation of temperature in apple and other temperate fruits causes Bitter pit (calyx zone is more susceptible), Cork Spot (due to high evapotranspiration), Superficial Scald (symptoms are produced by oxidation of α-pharnesen), Sunburn and Sunscald (developed under high solar radiation stress and results in increasing in lipid peroxides), (Colavita, 2008). Blackheart Injury and cambium injury in fruits like (apples, peaches, pears, plums and cherries).

Rainfall fluctuation

The production of fruit is also hampered by shifting precipitation patterns. Changes in rainfall patterns and amounts can have an impact on soil moisture levels, which can result in water stress or over-irrigation needs. Fruit size, flavour, and quality can all be affected by any circumstance. The fruit production can be made much more difficult by drought conditions, which can make fruit more vulnerable to pests and illnesses (Smith *et al.,* 2023d). Farmers are employing cutting-edge irrigation techniques like drip irrigation and precision water management systems in areas where water shortage is becoming more common to maximize water consumption and reduce losses (Garcia *et al.,* 2023b). Banana (Panama wilt) and papaya (Alteration in sex formation during flowering) show a negative impact on their vegetative and reproductive growth during high precipitation.

Harass weather

The increased frequency and intensity of extreme weather events pose threats to fruit production in addition to temperature and precipitation. Heat waves, storms, and frosts can physically harm trees, decreasing their ability to produce fruit and even killing them. For instance, recent studies have emphasized the detrimental effects of strong storms on orchards, which causes fruit growers to suffer large financial losses (Brown and Johnson, 2022b). To lessen the negative consequences of extreme weather occurrences, farmers are putting in place safeguards like windbreaks, hail nets, and enhanced orchard structures (Wilson *et al.,* 2021a).

Adverse effect of climate change on different fruit crops has been mentioned in Table 1.

Mitigation measures

The fruit sector is actively implementing adaptive solutions to deal with the problems caused by climate change. These solutions cover a range of tactics, such as the adoption of cultivars that are climate robust, modification of irrigation methods, and improved pest and disease management techniques. New fruit tree types with characteristics including heat tolerance, drought resistance, and disease resilience are being created by breeders (Cruz *et al.,* 2022a).

Fruit trees are affected negatively by shifting precipitation patterns and water stress, but these effects can be mitigated by using water

Table 1: Impact of climate change on various fruit crops

Fruit	Mango	Banana	Citrus	Pineapple	Custard apple	Avocado	Persimmon	Litchi
Impact of climate changes	Spongy tissue	Neer Wazahi (Water banana)	Granulation	Sun Scald	Stone fruit	Grey Pulp	Millerandage	Fruit Cracking
-Do-	Alternate Bearing	Chock Throat			Woodiness			
-Do-	Jhumka (Cluster bearing)							
-Do-	Jelly Seed							

[*Source: Source:* Singh, J. 2002. Basic Horticulture. Kalyani Publication, New Delhi.]

management techniques such drip irrigation and soil moisture monitoring (Smith *et al.,* 2023c). Fruit farmers can improve the health and production of their orchards by making the best use of their water resources and minimizing the dangers associated with them.

To manage pests and illnesses in fruit production systems sustainably, Integrated Pest Management (IPM) approaches are increasingly becoming more popular. Fruit growers can minimize chemical inputs and lessen the detrimental environmental impact by integrating diverse pest control approaches, such as biological control, cultural practices, and targeted pesticide application (Smith *et al.,* 2023a).

Fruit producers may support the general sustainability of the agriculture industry by reducing greenhouse gas emissions. For fruit growers to make educated judgments and adjust their management practices to changing climatic conditions, they must have access to current, reliable climate information. Weather forecasts, pest and disease alerts, and suggestions for the best times to plant and harvest crops are all provided to growers by climate information services and decision support systems (Wilson *et al.,* 2021b). These tools enable fruit growers to anticipate and respond to climate-related risks effectively.

Additionally, crop management decisions are being improved by applying precision agriculture technology such as remote sensing and data analytics (Garcia *et al.,* 2023c). With the use of these technologies, farmers can accurately apply water and nutrients, measure the health of their plants, and monitor soil moisture, resulting in more effective and long-lasting fruit production systems (Garcia *et al.,* 2023c).

To increase the resilience of fruit production systems, orchard management practices must incorporate climate change considerations. To find and spread best practices, create region-specific adaptation plans, and maintain the long-term viability of fruit production in a given area, researchers, farmers, and policymakers must work together.

To adapt to shifting climatic conditions, fruit growers need also to consider market diversification and crop selection. Growers may look into alternate fruit types or niche markets that may be less affected

Table 2 : While considering the effect of frost, below is the severity of different fruit crops.

Severity	Crops
Most Susceptible	Apricots, Avocados, Bananas, Berries (except cranberries), Lemons, Limes, Peaches, Plums
Moderately Susceptible	Apples, Cranberries, Grapefruit, Grapes, Oranges, Pears berries, Grapefruit, Grapes, Oran berries, Grapefruit, Grapes, Oranges, Pears
Least Susceptible	Dates

[Source: Wang and Wallace, 2003]

Table 3: Resistant and tolerant germplasm/rootstocks of various fruits

Fruit	Rootstock
Citrus	Rough lemon (Citrus jambheri Lush.), Kharna Khatta (Citrus karna), Rangpur lime (Citrus limonia), Gajanimma
Mango	Kurukkan, Olour, Vellaikoloban, Turpentine, Sucary and Sabre, 13-1, Pahutan, Goa
Guava (Seedling rootstocks)	*P. cattleianum, P. cujavillis*, *P. Pumilum* , Pusa Srijan
Apple	Standard (MM.111, Robusta 5), Semi-Dwarf (M.7, Vineland 4 MM.106, Dwarf (M.27, V3, M.9, M.26,
Peach	Lovell, Moderate, Halford, Nemaguard, Nemared, Guardian, Flordaguard, Titan Hybrids, Hansen
Pear	Quince A, Quince B

[Source Handbook of Horticulture, 2019, 2nd Edition, Division of Fruits and Horticultural Technology ICAR- Indian Agricultural Research Institute]

by climate change as a result of how climate change might affect the adaptability of particular fruit crops in particular places. Governmental policy assistance and incentives are essential for promoting climate adaptation in fruit production. Here are some resistant and tolerant germplasm/rootstocks of various fruits in Table 3

Supporting measures

Fruit growers may decrease climate-related risks and improve their resilience with the aid of financial incentives for implementing climate-smart practices, rules to cut emissions and support sustainable agriculture, and access to funding and insurance programmes (Brown and Johnson, 2022c). Promoting sustainable fruit consumption and assisting growers in implementing climate-smart practices need to increase consumer understanding of how climate change is affecting fruit production. Consumer appreciation for locally grown, sustainable fruit can be greatly influenced by educational efforts, labelling initiatives, and farmer-consumer engagement programmes. To solve the problems associated with fruit production caused by the climate, ongoing research and innovation are essential. In response to climate change, improvements in breeding procedures, agronomic practices, crop protection strategies, and post-harvest technology can improve fruit quality, production, and resilience. To foresee and reduce risks related to the climate, fruit growers must increasingly priorities long-term planning and risk assessment. Fruit growers can improve their capacity for adaptation and lessen potential disruptions by analyzing their exposure to climate risks, creating backup plans, and including climate change considerations in business strategies. Given the global character of climate change, international cooperation and policy coordination are essential for effective adaptation in fruit production. Collaboration across nations enables the interchange of information, assets, and technology, resulting in more effective and well-coordinated responses to climatic issues in the fruit business. The fruit business will benefit from ongoing research and knowledge exchange by maintaining production, protecting the world's fruit supply, and preserving farmers' livelihoods (Cruz *et al.,* 2022b; Wilson *et al.,* 2021c).

Demand for action

1. Encourage collaboration and knowledge sharing: Promote cooperation between farmers, academics, and policymakers to share information and best practices for fruit production that take climate change into account. Create forums where people from various places can exchange experiences, success stories, and difficulties. This collaboration will speed up learning and the creation of practical climate change adaptation methods.

2. Government support: Governments and organizations should give investments in climate-smart agriculture practices and infrastructure a high priority. As part of this, irrigation systems should be improved, agroforestry should be encouraged, precision agriculture technology should be used, and fruit production should promote the use of renewable energy sources. With these investments, fruit-producing systems will be more resilient and climate change-resistant.

3. Training and workshop: Farmers should be educated and given the tools they need to adapt to climate change by offering them training programmes and educational materials. Workshops on farming techniques that are climate-resilient, access to meteorological data and forecasting tools, and financial assistance for putting adaptation measures into action are a few examples of what this can include. We can improve farmers' ability to respond to climate change issues by providing them with the tools and knowledge they need.

4. Increase consumer awareness: Inform customers about the effects of climate change on the production of fruit and the significance of promoting locally and sustainably grown produce. By taking into account aspects like the carbon footprint of their food and supporting farmers who use climate-friendly practices, you can encourage consumers to make informed decisions. We may encourage market pressures towards more climate-resilient agricultural systems by increasing customer demand for fruits that are produced responsibly.

CONCLUSION

In conclusion, the worldwide fruit production industry faces considerable hurdles as a result of climate change. The growth, productivity, and quality of fruit trees are already being hampered by rising temperatures, changing precipitation patterns, and extreme weather events. Fruit tree development is impeded, fruit output is decreased, yields are lost, and fruit quality is affected. The fruit sector is using adaptable tactics to deal with these issues, such as creating fruit varieties that are resistant to climate change, employing integrated pest control techniques, and utilizing precision agriculture technologies. Cooperation between academics, farmers, and politicians is necessary for the successful mitigation of the effects of climate change on fruit production. To develop new strategies and technologies that can increase the adaptability of fruit production systems to a changing climate, it is essential to conduct ongoing studies, innovate, and share knowledge. The promotion of sustainable fruit production also heavily relies on market diversification, legislative support, and consumer education.

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SHORT COMMUNICATION

Mallero (*Elaegnus latifolia* L.) : A potential minor fruit of Darjeeling and Kalimpong hills

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ABSTRACT

The Mallerofruit is a very good source of vitamins, minerals, fatty acids and other bioactive compounds. The fruit contains carbohydrate 74.06%, protein 7.80 %, crude fibre 9.3%, sugar 8.3 % and potassium 910 mg/ 100 g. It is consumed raw, and the fruit pulp is used to make jam, jelly, and hydrating beverages. Mallero plants are also used as the fencing due to its thorniness. The shrub has the ability to fix atmospheric nitrogen in the soil. The flowering season of the plant is from November to February and fruits become ready to harvest during April to May. The fruit has many bio-active compounds and has great application in the field of pharmacological and nutraceutical industries.The fruit is used for preparation of many processed products and has a great ceremonial values for the local people in the region.

***Keywords*:** Darjeeling, *Eleagnus*, Kalimpong, mallero, underutilized fruit

INTRODUCTION

The hills of Darjeeling and Kalimpong are endowed with a variety of biological niches that support a range of sub-tropical to temperate area flora and fauna. The region's land use practices for forestry, agroforestry, agriculture, horticulture, and animal husbandry are crucial to the region's ability to sustain its economy. The local natives are well-versed in the utilisation of a variety of plant resources according to their traditional knowledge (Chhetri *et al*., 2022) and other plant parts in their folk and traditional medicines (Tripathi 2021). Some important and highly nutritious fruits consumed by the people in this region are *Baccaurea sapida* (Kusum), *Elaegnus latifolia* (Mallero), *Machilus edulis*(Pumsi), *Eriolobus indica* and *Diploknema butyraceae* (Cheuri). Mallero (*Elaegnus latifolia)*, one of the major wild edible fruit consumed by the people of Darjeeling and Kalimpong and belong to genus *Elaegnus* and family Elaegnacea. It is also nick named as Muslleri, Soh-hang and Basterdoleaster and native to India and southwest Asia. It looks like a tiny plum tomato dusted with silver glitter. Fruit contains an elongated seed inside having an odd striped pattern. It has a slight tomato like taste and are rich in lycopene. The fruit contain vitamins, minerals and essential fatty acid. It is consumed raw, and the fruit pulp is used to make jam, jelly and hydrating beverages. It has the ability to reduce the prevalence of cancer and is a surprisingly good source of essential fatty acids. The crop has the ability to fix atmospheric nitrogen in the soil (Patel *et al*., 2008). Despite having a huge potential for commercial, social, cultural, nutritional, medicinal, and environmental relevance in everyday life, the Mallero is still left as an underutilized fruit crop with little care. There is a vast scope for its utilization for income generation through vale addition.

BOTANICAL DESCRIPTION

Mallero is a large evergreen, heavily branched woody shrub (Fig.1a) having rusty-shiny scales, thorny, spreading in nature and can grow up to the

height of 10 feet and spreading up to 3 feet more with support of nearby stuff. Leaves are alternate, ovate-oblong, glabrous, clothed beneath with shiny silvery or ferruginous scales(Fig.1b). The adaxial surface of leaves are dark green where abaxial surface is silvery white. The maximum leaf length is reported around 14 cm and breadth is 8.5 cm. The flowers are creamy white to light yellow colour, hermaphrodite in nature and pollinated by bees. They are scented, peduncled and very small covered with ferruginous scales. The flowering season of the plant is from November to February and fruits become ready to harvest during April to May. The Fruits are oblong to ovoid, ribbed, glittered with silver dust and turn orange to dark red when ripe. Fruits may only be kept for 3-5 days at room temperature and are often seasonal and very perishable due to their high moisture content (87.31%).The seeds are creamish, elongated with an odd striped pattern, white in colour and is weighing around 2 to 4 gram.

DISTRIBUTION

Mallero is extensively grown in subtropical and sub temperate zone, though it can be found at higher elevation in the tropics. It can be found up to a height of 1900 metres and is typically cultivated in semi-wild conditions. This species has a large geographic range that extends from Northern Asia to the Himalaya and Europe (Ahmadiani *et al*., 2000). The primary centre of genus *Elaeagnus* is believed to be China. It is grown in China, from the Yangtze River Valley to the southern region, while it is also found in northwest China. It has been spread throughout Eastern Asia, reaching as far as South Asia and Queensland in northern Australia. Some species can also be found in southern Europe and North America. In India, it is quite prevalent in Sikkim, Assam, Nagaland, Khasi and Jaintia hills of Meghalaya up to an elevation of 1500 metres above mean sea level (Patel *et al.*, 2008).

USES

Fruits are often consumed raw form when it is fully ripe. In addition, the local people of the region has discovered several other processed product. They are making pulp based refreshing RTS (Ready to Serve) drinks, pickles, jam and jelly. The local farmers are making an alternative source to earn by selling processed product from mallero. The dried wood can be used as a firewood. The fruit is considered an essential item for offering to the Gods during marriage ceremony rituals of Assamese and Manipuri communities.

MEDICINAL USES

Astringent properties can be found in the fruits and flowers. Leaves and fruits may be used in the food processing or pharmaceutical industries as a natural source of antioxidants (Dutta *et al.*, 2018). The fruit extract has potential use in the pharmaceutical and nutraceutical industries for the creation of functional products, as it demonstrates strong antioxidant and antibacterial action and includes bioactive chemicals and minerals (Dasila and Singh, 2022). The fruit may have the ability to reduce the occurrence of cancer as well as slow down or stop the growth of pre-existing tumours (Seal, 2012). The leaves should be a good aid in the production of silver nanoparticles. These nanoparticles have a great application in the field of pharmacological and electronical industries (Phanjom *et. al*., 2012). The fruit is useful in constipation and as a health tonic. Local people consume the root extract of the fruit during pregnancy to prevent miscarriage.

NUTRITIONAL PROPERTIES

The fruit has high nutritional value and considered as a very rich source of minerals, vitamins, essential fatty acid and other bioactive compounds. Because of their abundance in ascorbic acid, phosphorus, potassium, manganese, calcium, sulphur, iron, zinc, boron, and copper, fruits may become more popular with consumers. Toxic elements such as Pb, As, Al, Ti, Ni, and Cr were also discovered to be safe for human consumption (Lepcha, *et. al.*, 2018).

Table 1 summarises nutritional characteristics and bioactive components of *Elaeagnus latifolia* L. It includes protein, carbohydrate, crude fibre, ash, crude fat, phosphorus, magnesium, and zinc, as well as their respective quantities. It also includes sugar, calorific value, vitamin C, calcium, iron, potassium, sodium, and copper levels.

Table 2 illustrates the physical and chemical features of *Elaeagnus latifolia* L. fruits grown at

(a) (b)

Fig. 1: Plant (a) and leaf (b) of *Elaegnus latifolia* L.

(a) (b) (c)

Fig. 2: Immature fruits (a), mature fruits (b) and harvested fruits (c) of *Elaegnus latifolia* L.

Meghalaya. It includes parameters like fruit weight, fruit length, fruit diameter, pulp recovery, seed weight, seed length, and seed diameter, as well as their values. TSS (°Brix), acidity, pH, porosity, TSS: acid ratio, pulp/seed ratio, and moisture content are among the chemical parameters.

PROPAGATION

Seed: Seed should be collected from ripe fruit of healthy mother plants. Cold frame is the best propagation structure to sown seed. Seed germination can be observed after 3 to 4 weeks though it may take 16 to 18 months to transplant

Table 1: Nutritional Properties & Bioactive compound of *Elaeagnus latifolia* L.

Ingredients	Amount	Ingredients	Amount
Protein	7.80%	Sugar	8.3%
Carbohydrate	74.06%	Calorific Value	332.10Kcal/100 g
Crude fibre	9.3%	Vitamin C	12 mg/100 g
Ash	3.6%	Calcium	46.67 mg/100 g
Crude Fat	0.52%	Iron	180 mg/100 g
Phosphorus	228.99 mg/ 100 g	Potassium	910 mg/100 g
Magnesium	42.82 mg/100 g	Sodium	5.58 mg/100 g
Zinc	0.25 mg/100 g	Copper	2.59 mg/100 g

(Source: Kumar and Said, 2018)

Table 2: Physical and chemical fruit properties of *Elaeagnus latifolia* L. grown at Meghalaya

Physical parameters	Value	Chemical parameters	Value
Fruit weight (g)	13.80	TSS (°Brix)	9.31
Fruit Length (cm)	3.10	Acidity (%)	3.46
Fruit diameter (cm)	2.45	pH	3.29
Pulp recovery (%)	70.19	Porosity	37.38
Seed weight/fruit (g)	3.16	TSS: acid ratio	3.29
Seed length (cm)	2.87	Pulp / seed ratio	4.73
Seed diameter (cm)	1.24	Moisture content (%)	87.31

(Souce: Patel *et al*., 2008)

due to slow germination process. Seedling are poked vertically into individual for easy handling and when they are at least 15 cm tall, they are transplanted on the field.

Cutting: Hard wood cuttings are procured from current season growth having pencil thickness with 4-6 nodes of about 8 to 12 cm long are recommended and planted in a polybag. Cutting takes two to three weeks to sprout and ready for transplanting after 2-3 months in a field.

Suggestive action plan for the preservation, reproduction, extension, usage, and promotion of Mallero.

i. Reproduction and propagation: Efficient methods for reproduction and propagation need to be developed to ensure a sustainable and consistent supply of planting materials. This involves optimizing the propagation techniques as well as enhancing the cultivation processes to boost its yield.

ii. Extension and cultivation: Extending the cultivation, particularly in regions beyond its current prevalent areas, could diversify its availability. Research and initiatives aimed at introducing and cultivating in newer geographical locations should be explored to expand its reach.

iii. Utilization and commercialization: Leveraging the diverse utilization, including its nutritional, medicinal, and commercial potential, requires increased awareness and promotion. Educating local communities, farmers, and industries about its value and encouraging its use in various products could enhance its commercial viability.

iv. Promotion and public awareness: Raising public awareness about the significance of the fruit in various aspects of daily life is crucial. Educating farmers, consumers, and industries about its nutritional richness, health benefits, and versatile applications can amplify its demand and utility.

v. Collaborative efforts and research: Collaborative efforts among governmental bodies, research institutions, agricultural organizations, and local communities are pivotal. Investing in research, development, and technological innovations related to mallero can improve its production, processing, and market reach.

CONCLUSION

Nutritional richness, versatile applications, and potential health benefits underscore its significance as an underutilized fruit deserving of further exploration and utilization. Its capacity to enhance living standards and contribute to long-term growth makes it a valuable asset that warrants increased attention and commercialization for the betterment of local communities and the industry at large.

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Society for Minor Fruits, Medicinal and Aromatic Plants

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International Journal of Minor Fruits, Medicinal and Aromatic Plants is the official publication of the Society for Minor Fruits, Medicinal and Aromatic Plants. The journal covers basic and applied aspect of original research on all branches of Minor Fruits, Medicinal and Aromatic Plants and any crops, plants and plant parts having medicinal and aromatic properties. Its goals are to apprise horticultural, agricultural, plant-based pharmaceutical scientists and others interested in any crops having medicinal values specially emphasized on minor or underutilized fruits, medicinal and aromatic plants of scientific and industrial development and extension for betterment of man kinds. The area of research include evaluation of germplasm, breeding, agronomic practices, physiology, biochemistry, phyto-chemicals study, biotechnology, soils and plant nutrition, plant protection, weed control, pesticide residue, post harvest technology, economics, extension, farm machinery and mechanization etc. which facilitate the growth and extension of minor and underutilized fruits, medicinal and aromatic plants.

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Anonymous. 1979. *Mango varieties of West Bengal. Technical Bulletin No. 1*. Department of Horticulture, Faculty of agriculture, Bidhan Chandra Krishi Viswavidyalaya. Pp.52.

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Proceedings: Blake, M.A. 1932. The J.H. Hale as a parent in peach crosses. *Proc. Am. Soc. Hort. Sci*., 29:131-136.

Monet, R. 1979. Transmission génétique du caractère 'fruit doux' chez le pêcher. Incidence sur la selection pour la qualité. *In: Proceedings of Eucarpia Fruit Section Symposium*. Tree Fruit Breeding. INRA, Angers, France, pp. 273-276.

Bulletin: Gray, P. 1914. The compatibility of insecticides and fungicides. Monthly bulletin of California, July, 1914.

Annual meetings: Schenck, N.C. 1965. Compatibility of fungicides with insecticides and foliar nutrients. *57th annual Meeting of American Phytopathological Society*, 3-7 October, 1965.

Reports: Anonymous, 1971. Investigations of insects pests of sorghum and millets. *Final Technical report*, 1965-70, IARI, New Delhi, pp.157.

Annual report: Anonymous, 2010. *Annual Report for 2010-11*, Faculty of Horticulture, Bidhan Chandra Krishi Viswavidyalaya, Monhanpur, Nadia, West Bengal, India. Pp.80-85.

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