

Phyllotaxic diversity as a means of assessing variations in mulberry (*Morus* spp.)

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ABSTRACT

Plant architecture is characterised by the regular spacing of lateral organs along stems and branches, an arrangement known as phyllotaxis. Leaf arrangement pattern is a specific trait for a given species and in the majority of angiosperms, it is relatively stable and primarily represented by the Fibonacci pattern. However, diversity has been noted within and amongst the studied mulberry species. The orders of spiral phyllotaxy were $\frac{1}{2}$, $\frac{1}{3}$, $\frac{2}{5}$, $\frac{3}{7}$ or $\frac{3}{8}$. Most of the genotypes studied like Thar Harit, Thar Lohit, MI-315, Saharanpur Local-2, Delhi Local, Gurgaon Local, CIAH-3 and Ajmer Local showed $\frac{1}{2}$ phyllotaxy pattern, while genotypes MI-380 and Anand Local exhibited $\frac{1}{3}$ leaf arrangement. Genotype Saharanpur Local-1 showed $\frac{3}{8}$ pattern. Only genotype MI-172 deviated from the Fibonacci series and exhibited $\frac{3}{7}$ phyllotaxy. Thus, phyllotaxic divergence can be utilized for differentiating the mulberry varieties.

Keywords: Morphological characterization, phyllotaxic variation, mulberry, distichous

INTRODUCTION

Mulberry (*Morus* spp.; Family- Moraceae) is widely distributed in the temperate, subtropical, or tropical regions of the world and can grow in a wide range of climatic, topographical, and soil conditions. Mulberry is considered to be originated in the border area of the Indo-Chinese region and distributed in the lower slopes of the sub-Himalayan zone up to an elevation of 3300m (Tikader and Vijyan, 2010). In India, there are many species, of which *Morus alba* and *M. indica* are fully domesticated while other important species are *M. laevigata*, *M. rubra*, *M. nigra* and *M. serrata* (Vijayan *et al.*, 2011). Mulberry is a multipurpose tree and has huge potential economic value other than sericulture owing to its several unique and special features. Apart from being the sole food plant of mulberry silkworm (*Bombyx mori*), mulberry can also be utilized for catering diversified needs such as food, fodder, fuel and fibre. The ethno-botanical usage of mulberry involves consumption of ripe fruits, which are highly appreciated for their delicious taste and are consumed either fresh or after extraction of juice. Immature fruits are used for *chutney* preparation (Jalikap *et al.*, 2011; Krishna and Chauhan, 2015). Mulberry fruit is used to treat weakness, dizziness,

tinnitus, fatigue, anemia and incontinence (Tikader and Vijyan, 2010).

The effective utilisation of germplasm depends on systematic characterisation and identification of genotypes with specific traits (Dandin and Jolly, 1986; Fakir *et al.*, 2018). Further, selection of suitable genotypes from gene pool requires a thorough knowledge of morphological characteristics of different genotypes for utilizing them in breeding (Yilmaz *et al.*, 2012). Traditional methods for cultivar identification are based on the observation of phenotypic characteristics as they aid fast and simple evaluation of variability and; hence, are considered as an effective means of preliminary evaluation of assessing genetic diversity among morphologically distinguishable accessions (Èoliæ *et al.*, 2012; Rumana *et al.*, 2016). Furthermore, morphological characterization is the official method accepted for registration and protection of new cultivars (Ertan, 2007).

Phyllotaxy (arrangement of leaves) can be described as a fraction with the numerator being the number of turns around the stem and the denominator being the number of leaves it takes to return to original leaf position. Phyllotaxy indices (like many other spirals in nature) follow a

Fibonacci series (1, 1, 2, 3, 5, 8, 13, 21, 34, 55, 89 ...). Phyllotaxy follows a numerator and denominator set of offset Fibonacci numbers such as $\frac{1}{2}$, $\frac{1}{3}$, $\frac{2}{5}$, $\frac{3}{8}$... (Prusinkiewicz and Lindenmayer, 2012). Leaf arrangement in the majority of angiosperms is relatively stable and primarily represented by the Fibonacci pattern (ca. 95%), with additional patterns occurring only infrequently and most times appearing as the bijugy of the main Fibonacci pattern and the Lucas pattern (Jean, 1994; Pennybacker *et al.*, 2015). Despite the enormous range of phyllotactic patterns both between and within taxa, some patterns are strongly associated with and preserved in specific taxon (Gola and Banasik, 2016). Leaf arrangement pattern is a specific trait for a given species (Pennybacker *et al.*, 2015). For instances in hazel, the leaves are separated by one-third of a revolution ($\frac{1}{3}$ phyllotaxis). Likewise, the apricot, and the cherry tree exhibit $\frac{2}{5}$ phyllotaxis and the pear $\frac{3}{8}$, while that of the almond is $\frac{5}{13}$. However, diversity has been noted within and amongst the mulberry species. This trait can be utilized for differentiating the varieties. Much less attention is paid to study the phyllotaxis diversity in fruit crops, as in mulberry, and only meagre information is available. Here, we focus on the patterns of leaf arrangements in mulberry germplasm available at ICAR-CIAH, Bikaner and discuss the possible factors involved in the deviation in observed phyllotactic patterns.

MATERIALS AND METHODS

Thirteen mulberry genotypes, grown at Mulberry Germplasm Block of ICAR-Central Institute for Arid Horticulture, Bikaner, India ($73^{\circ}35'E$ and $28^{\circ}1'N$) were selected for the studies. These genotypes were obtained from various sources such as research institutes or through survey and collection (Table 1), including two recently identified varieties (Thar Lohit and Thar Harit) for commercial cultivation by the ICAR-Central Institute for Arid Horticulture, Bikaner. Three 'single tree', raised through stem cuttings and trained on modified leader system, replicates of each genotype were planted 6 x 6 m apart in randomized block design (Panse and Sukhatame, 1967). The soil of experimental site

was loamy sand in texture with pH 8.23, EC 1.59 dS/m, and organic carbon 0.27%. During the experiment, field was fertilized annually with [50kg N, 50 kg P, 50 kg K, and 10 t Farm Yard Manure per hectare].

Species of different *Morus* genotypes were ascertained (Table 1) by distinguishing them for the morphological characters as suggested by Vijayan *et al.* (2011). Phyllotaxy was recorded in the middle portion of the longest shoot leaving emerging young leaves from the top and the lower portion of the shoot. When first and third leaf is in the same direction on the stem, it was referred as distichous ($\frac{1}{2}$). However, when first and fourth leaf were in same direction on the stem and first and sixth leaf were in same direction on the stem, they were termed as tristichous ($\frac{1}{3}$) and pentastichous ($\frac{2}{5}$), respectively (Ahuja and Jain, 2017). Similarly, when first and seventh and first and eighth leaf is in the same direction on the stem, they are said to be $\frac{3}{7}$ and $\frac{3}{8}$, respectively.

RESULTS AND DISCUSSION

Plants display a wide variety of three dimensional forms, or architectures, which are critical for their survival in competitive environments or, in the case of crops, for their productivity. Architecture is generated after embryogenesis through the activities of apical meristems. Leaves are the principal lateral organ that determines the plant shoot morphology and they normally develop in very regular patterns in time and space (Lee *et al.*, 2009). There is a definite pattern according to which the leaves are arranged on the stem of a particular species, which may vary depending on the type of the element and the taxonomic position of the species. The most common pattern is spiral phyllotaxy. Spiral phyllotaxis is formed when organs successively initiated at the apex are circumferentially displaced from one another by an angular distance known as the divergence angle, where a single leaf grows out at each node and successive leaves form a spiral pattern with an approximately 137° displacement from the previous leaf. A wide variation was recorded regarding leaf arrangement (phyllotaxy), which was noted to be either alternate or spiral (Fig. 1; Table 2). The orders of spiral phyllotaxy were

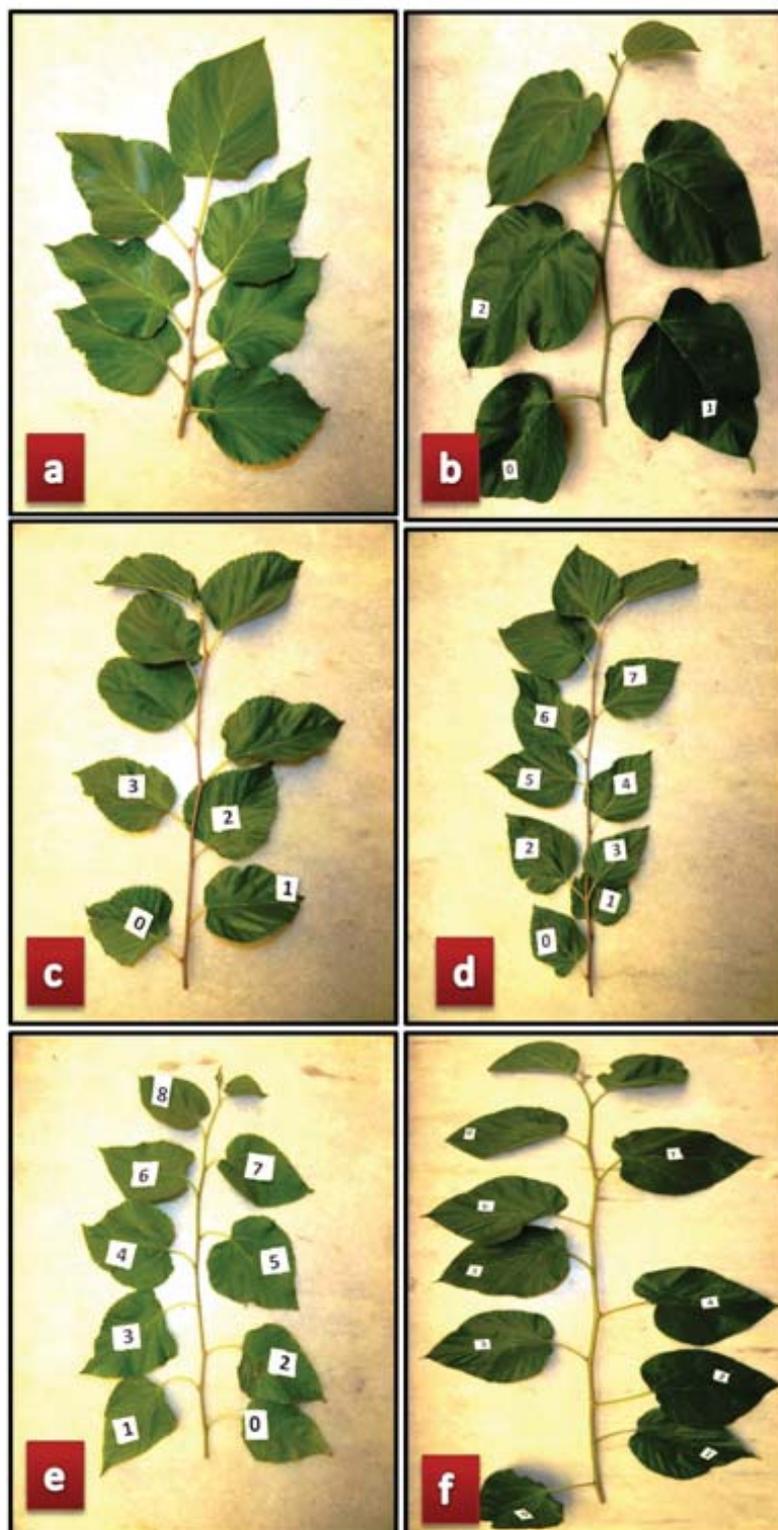


Fig. 1. Divergence in phyllotaxy as seen in mulberry genotypes; (a) & (b) Distichous or $\frac{1}{2}$ Phyllotaxy (c) Tristichous or $\frac{1}{3}$ Phyllotaxy (d) Pentastichous or $\frac{2}{5}$ Phyllotaxy (e) $\frac{3}{7}$ Phyllotaxy (f) Octastichous or $\frac{3}{8}$ Phyllotaxy

Table 1: List of mulberry (*Morus* spp.) genotypes and their corresponding source

S. No.	Genotypes	<i>Morus</i> species	Source
1.		Thar Lohit	<i>Morus rubra</i> Chance seedling spotted at ICAR-Central Institute for Arid Horticulture, Bikaner (Rajasthan)
2.	Thar Harit	<i>Morus alba</i>	Pushkar (Rajasthan)
3.	CIAH-3	<i>Morus alba</i>	Hisar (Haryana)
4.	Delhi Local	<i>Morus laevigata</i>	New Delhi
5.	Ajmer Local	<i>Morus laevigata</i>	Ajmer (Rajasthan)
6.	Gurgaon Local	<i>Morus laevigata</i>	ICAR-National Bureau of Plant Genetic Resources, New Delhi
7.	Saharanpur Local-1 (SL-1)	<i>Morus laevigata</i>	ICAR-National Bureau of Plant Genetic Resources, New Delhi
8.	Saharanpur Local-2 (SL-2)	<i>Morus laevigata</i>	ICAR-National Bureau of Plant Genetic Resources, New Delhi
9.	Anand Local	<i>Morus laevigata</i>	ICAR-National Bureau of Plant Genetic Resources, New Delhi
10.	MI-172*	<i>Morus indica</i>	Central Sericultural Germplasm Resources Centre
11.	MI-300*	<i>Morus indica</i>	Central Sericultural Germplasm Resources Centre
12.	MI-315*	<i>Morus laevigata</i>	Central Sericultural Germplasm Resources Centre, Hosur (Tamil Nadu)
13.	MI-380*	<i>Morus laevigata</i>	Central Sericultural Germplasm Resources Centre, Hosur (Tamil Nadu)

* MI = *Morus* Indigenus**Table 2: Phyllotactic diversity in mulberry germplasm**

S. No.	Genotype	Phyllotactic arrangement
1.	Thar Lohit	1/2
2.	Thar Harit	1/2
3.	CIAH-3	1/2
4.	Delhi Local	1/2
5.	Ajmer Local	½
6.	Gurgaon Local	1/2
7.	Saharanpur Local-1	3/8
8.	Saharanpur Local-2	1/2
9.	Anand Local	1/3
10.	MI-172	3/7
11.	MI-300	2/5
12.	MI-315	½
13.	MI-380	1/3

$\frac{1}{2}$, $\frac{1}{3}$, $\frac{2}{5}$, $\frac{3}{7}$ or $\frac{3}{8}$. Most of the genotypes studied like Thar Harit, Thar Lohit, MI-315, Saharanpur Local-2, Delhi Local, Gurgaon Local, CIAH-3 and Ajmer Local showed $\frac{1}{2}$ phyllotaxy pattern, while genotypes MI-380 and Anand Local exhibited $\frac{1}{3}$ leaf arrangement. Distichy ($\frac{1}{2}$) phyllotactic pattern is common to the family Poaceae, as well as representatives of a few other families, including the Orchidaceae. Although, generally rare in dicots, distichy is a typical phyllotactic pattern of *Pisum sativum*. On the other hand, Tristichy ($\frac{1}{3}$) is a rather rare pattern in angiosperms, although it may be a characteristic phyllotaxis of the Cyperaceae, as three regular orthostichies typically occur in this family (Gola and Banasiak, 2016). Furthermore, MI-300 showed $\frac{2}{5}$ phyllotaxy sequence. Phyllotaxy pattern like $\frac{1}{2}$, $\frac{1}{3}$ or $\frac{2}{5}$ have been reported in genotypes of different mulberry species naturally available in India (Tikader *et al.*, 2011; Tikader and Kamble, 2008); however, subsequent phyllotaxy sequences have not been reported. Genotype Saharanpur Local-1 showed $\frac{3}{8}$ pattern, which is a next numeric sequence in Fibonacci series.

All these aforementioned genotypes followed Fibonacci series except MI-172 (Table 2), which deviated from the Fibonacci series and exhibited $\frac{3}{7}$ phyllotaxy. Gola and Banasiak (2016) reported that if the spiral counts do not belong to the regular Fibonacci sequence, they are most likely consecutive members of the double Fibonacci sequence 2, 2, 4, 6, 10, 16, 42, . . . or the Lucas sequence 1, 3, 4, 7, 11, 18, 29, . . . According to one account, of the plants with spiral phyllotaxis, 91% have counts in the regular Fibonacci sequence, 5% in the double Fibonacci sequence, and 2% in the Lucas sequence. In this study, the leaf arrangement pattern displayed by the MI-172 seems to follow Lucas sequence. Earlier, Keller (2015) also reported $\frac{3}{7}$ phyllotaxy sequence in grapevines, which is known to have $\frac{2}{5}$ phyllotaxy when raised from seeds. Sharma *et al.* (2010) also noted phyllotaxic variation in guava varieties. The phyllotaxic pattern was found to be either decussate or superimposed depending upon the varieties. The level of phyllotaxis diversity corresponds with shoot apical meristem organization; there is a much greater pattern diversification in plant species which have layered meristems than in species that have segmented meristems and a single apical cell (Wiss and Zagórska-Marek, 2012).

Phyllotaxic diversity seems to be an important character, which can be utilized for distinguishing mulberry genotypes. This attains more importance in view of DUS (Distinctness Uniformity Stability) test of varieties, which primarily rely on morphological traits. The outcome of the present variety can help a breeder to distinguish a variety based on phyllotaxic diversity, which is a stable character and remain uninfluenced by environmental variations.

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