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High yielding mulberry production through controlled pollination for enhanced vegetative growth and early sprouting suitable for tropical agroclimatic regions

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Abstract
Mulberry is an important crop for the sericulture sector because they are the main source of nutrition for silkworms. Sericulture is the practice of raising silkworms to produce silk, which is a valuable and opulent textile in many industries. Mulberry is the primary food source for silkworms, hence the availability and quality of mulberry are crucial to the sericulture industry. Therefore, development of improved varieties is essential for enhancing silk production. Attempts were made to develop high yielding mulberry varieties with vigorous vegetative growth and early sprouting suitable for tropical agroclimatic regions through controlled pollination. The importance of developing a high-yielding variety with early sprouting is emphasized, as it can lead to increased silk production with improved profitability to farmers. The review also describes methods of controlled pollination, which are used to produce hybrid seeds with desired traits. The various factors that influence the success of controlled pollination, including timing, temperature, humidity, and nutrition, are discussed. Finally, the review summarizes the research on the development of high-yielding mulberry varieties with vigorous vegetative growth and early sprouting. Overall, the information provides valuable insights into the development of improved mulberry varieties for tropical regions and highlights the potential for increased silk production and profitability to farmers.

Keywords: Mulberry, hybridization, sericulture, silk industry

1. Introduction

Mulberry is an important crop in the sericulture industry because it serves as the primary food source for silkworms (Altman & Farrell, 2022) [3]. Sericulture is the process of rearing silkworms for the production of silk, which is considered as a valuable and luxurious textile (Guo *et al*., 2019) [12]. Therefore, the success of the sericulture industry largely depends on the availability and quality of mulberry as the main food source for silkworms. Cross-pollinated and extremely heterozygous, the mulberry plant (Genus: *Morus*; family: Moraceae) is a woody perennial tree or shrub. It has a strong root system, displays quick growth, and can generate a lot of biomass (Alipanah *et al*., 2020) [2]. Mulberry leaves are an essential part of the sustainable manufacturing of silk since they are abundant in proteins, carbs, and moisture, which are essential for the upbringing of silkworms. A reliable source of adequate amounts of high-quality mulberry leaves is crucial for the silk industry development (Ruth *et al*., 2019) [20]. Mulberry farming has been genetically enhanced in several nations to fulfill this need, notably in China and India, which rank first and second in the world in terms of mulberry silk output. The majority of the world's mulberry silk is consumed in India. There is a need to improve the production of high-quality silk by breeding high-yielding mulberry types that can adapt to various temperatures and changing environments (Vijayan *et al*., 2012) [33]. In order to ensure a consistent supply of high-quality mulberry, the development of high yielding mulberry varieties with vigorous vegetative growth and early sprouting is crucial, especially in tropical agroclimatic regions (Umarov *et al*., 2021) [29]. This is because the quality and quantity of mulberry production can be affected by various environmental factors such as temperature, humidity and rainfall, which are often unpredictable in tropical regions. Therefore, it is essential to have mulberry varieties that are adaptable to these environmental conditions and can produce high yields of quality leaves throughout the year (Ram, 2016) [16]. Controlled pollination is an important technique used in the development of such high-yielding mulberry varieties. Controlled pollination involves manual transfer of pollen from the male to the female parent plant under controlled conditions, with the aim of obtaining desired traits in the offspring. This technique allows plant breeders to selectively breed for specific traits such as high yield, early sprouting, and disease resistance, among the existing. In conclusion, mulberry is a crucial crop in the sericulture industry, and the development of high yielding mulberry varieties is essential for the success of this industry, particularly in tropical agroclimatic regions. Controlled pollination is an important technique used in the development of such varieties, as it allows for the selection and breeding of desirable traits in the offspring.

2. Botanical description of mulberry

Mulberry is a woody deciduous perennial plant that grows quickly and has an extensive root system. Simple, alternately arranged, with stipules and petioles and with one to five lobes, its leaves can either be whole or lobed (Acharya *et al*., 2022) [1]. Usually dioecious, the plant has a catkin-shaped inflorescence with unisexual blooms on a dangling or drooping peduncle. There is usually an auxiliary place for inflorescence. The male flowers are loosely distributed, and male catkins are often longer than female catkins. Male inflorescences that have released their pollen dry out and fall off, leaving behind four stamens that are inflexed in bud and four persisting perianth lobes (Taylor *et al*., 2006) [27]. The female inflorescence is typically short and produces flowers with four persistent perianth lobes that are placed closely together. The stigma is bifid, and the ovary is one cell in size. For mulberry plants, wind is the main pollinator. The mulberry's fruit is called a sorosis and it typically has a violet-black tint. Although triploids (2n=3x=42) are also commonly grown for their adaptability, strong growth, and superior leaf quality, the majority of species and cultivated cultivars of the genus *Morus* are diploid in nature with 28 chromosomes (Shafiei *et al*., 2018) [24].

3. Cultivated species and varieties of mulberry

There are 68 species in the genus *Morus*, most of which are found in Asia, mainly in China (24 species) and Japan (19 species) (Table 1). Moreover, a wide variety of *Morus* species may be found throughout North America. In contrast, the genus is missing from Australia and is poorly found in Africa, Europe, and the Near East (Zeng *et al*., 2015) [37]. *Morus* species that grow naturally in the Himalayas, such as *Morus alba*, *M. indica*, *M. serrata*, and *M. laevigata*, are found across India. In addition, a number of newly imported species, including *M. multicaulis*, *M. nigra*, *M. sinensis*, and *M. philippinensis*, of which *M. indica* is the most widespread Indian mulberry variety, have been discovered (Tikader & Vijayan, 2017) [28]. Four of the 15 *Morus* species are found in China. They are *Morus alba*, *M. multicaulis*, *M. atropurpurea*, and *M. mizuho*, which are grown for sericulture. *M. multicaulis*, *M. alba*, *M. tartarica*, and *M. nigra* are all found in the former Soviet Union. While mulberry cultivation is done in a wide range of climates, the preponderance of the regions which include the states of Karnataka, Andhra Pradesh, and Tamil Nadu which are in the tropical zone and account for around 90% of the cultivation (Table 2). West Bengal, Himachal Pradesh, and north-eastern states have substantial regions under mulberry farming in the subtropical zone.

Table 1: A brief description of major species of mulberry

Mulberry species	Origin/widely cultivated	Ploidy level	Salient features
M. notabilis	Yunnan and Sichuan Provinces in China	n = x = 14	It has basic chromosomes, suggesting that it is basal to all the other species in its genus.
Red mulberry (*Morus rubra*)	Eastern North America	2n = 2x = 28	The plant's leaves can either be unlobed, two-lobed, or three-lobed, and it produces dark purple fruits that are edible.
White mulberry (*M. alba*)	Asia	2n = 2x = 28	It is so called because of the white fruits it bears; its leaves are used as food for silkworms.
M. indica	India, Southeast Asia	2n = 2x = 28	The tree's leaves are typically attached to petioles and come in varying lengths of 4 to 12.5 cm and widths of 2.5 to 7.5 cm. They are usually ovate or lobed with a caudate shape. *M. indica* is frequently cultivated for its medicinal qualities and like many other berries, the mulberries from this tree possess powerful antioxidant properties.
M. australis	East and Southeast Asia	2n = 2x = 28	This plant has edible fruits and leaves and is commonly utilized as feed for raising silkworms. Additionally, it is extensively cultivated for its fiber, which is used in the production of cloth and paper.
M. cathayana	China, Japan, Korea	2n = 4x = 56	This tree, commonly found in secondary forests and scrubland, is deciduous in nature. It can grow up to a maximum height of 15 meters and blooms during the months of May and June. The tree bears edible berries and its leaves can be used to make tea.
M. serrata	Tibet, Nepal, north western India	2n = 6x = 84	This is a compact deciduous tree that can reach up to a height of 15 meters. The leaves are 10-14 cm long and 6-10 cm wide, and covered with dense hair on the veins beneath while the upper surface is smooth. The fruit is a cluster of small drupes that measure 2-3 cm in length and turn red when ripe. These drupes are edible.
Black mulberry (*M. nigra*)	Western Asia	2n = 22x = 308	The primary reason for its cultivation is its sizable, succulent purple-black fruits, which surpass the flavour of red mulberry fruits.

Table 2: A brief overview of Indian mulberry varieties

Variety	Region/ soil type	Developed at	Pedigree	Yield (MT/ha/year)
S-1	Eastern and NE India, Irrigated	CSRTI, Berhampore	Introduction from Myanmar	28-29
Goshoerami	Temperate	CSRTI, Pampore	Introduction from Japan.	15-20
Kosen	Hills of Eastern India	CSRTI, Berhampore	Introduction from Japan	4-5 (hills) 10-12 (foot hills)
Chak Majra	Subtemperate	RSRS, Jammu	Selection from natural variability	20-22
Kanva-2	South India, Irrigated	CSRTI, Mysore	Selection from natural variability	20
MR-2	South India, Rainfed	CSRTI, Mysore	Selection from open pollinated hybrids.	35
S-7999	Eastern and NE India, Irrigated	CSRTI, Berhampore	Selection from open pollinated hybrids	30
S-146	N. India and Hills of J and K, Irrigated	CSRTI, Berhampore	Selection from open pollinated hybrids	30-35
G2	South India, Irrigated	CSRTI, Mysore	selection from controlled pollinated hybrids of *M. multicaulis* and S-34	36-38
G4	South India, Irrigated	CSRTI, Mysore	Selection from cross pollinated hybrids of *M. Multicaulis* and S-30	12-15
DD	South India, Irrigated	KSSRDI, Thalaghattapura	Clonal selection	40
China White	Temperate	CSRTI, Pampore	Clonal selection	15-20
S-1635	Eastern and NE India, Irrigated	CSRTI, Berhampore	Triploid selection	40-45
Tr-10	Hills of Eastern India	CSRTI, Berhampore	Triploid [T-4 (4x) x Philippines (2x)]	7-8 (hills) 12-14 (foot hills)
Tr-23	Hills of Eastern India	CSRTI, Berhampore	Triploid [T20 (4x) × S162 (2x)]	11-12 (hills) 24-25 (foot hills)
C-1730	Eastern and NE India, Irrigated	CSRTI, Berhampore	Triploid [T25 (4x) × S162 (2x)]	15-16
S-36	South India, Irrigated	CSRTI, Mysore	Developed through EMS treatment of Berhampore Local	35-45
S-54	South India, Irrigated	CSRTI, Mysore	Developed through EMS treatment of Berhampore Local	17
$BC_2$59	Hills of Eastern India	CSRTI, Berhampore	Back crossing of Kosen with the hybrid of *M. indica* var Matigara Local x Kosen	9-10 (hills) 15-16 (foot hills)
S-13	South India, Rainfed	CSRTI, Mysore	Selection from polycross (mixed pollen) progeny	18
S-34	South India, Rainfed	CSRTI, Mysore	Selection from polycross (mixed pollen) progeny	17

4. Breeding objectives to develop high yielding mulberry varieties

Research projects involve the evaluation of various mulberry genotypes for their suitability in producing high yields of foliage biomass, which is important for the production of silkworms. Specifically, the research aims to identify mulberry genotypes that sprout early during winter, which could provide an advantage in terms of growth and yield. The evaluation process will involve several criteria, including the measurement of foliage biomass (leaf yield/plant/year, weight of branches/plant, no of branches/plant, length of the longest branch, stem diameter, fresh weight of 100 leaves, no of nodes/meter length of shoot, leaf weight/meter length of longest branch, and leaf shoot ratio), as well as other agronomic traits that could affect the growth (leaf shedding, earliness of sprouting, and rooting capacity) and health of the plants (Figure 1). The desirable leafy traits for mulberry varieties for sericulture include the leaf shape and size, texture, colour, nutrient content, absence of diseases and pests (Caccam & Mendoza, 2015) [7]. The leaves of mulberry plants should be large enough to provide sufficient food for silkworms. Larger leaves are preferred as they can provide more nutrition to silkworms. The texture of mulberry leaves is also an essential factor in determining their suitability for sericulture. The leaves should be smooth and soft, making them easy for silkworms to consume. Mulberry leaves with a dark green colour are preferred because they contain higher levels of chlorophyll, which is essential for the growth and development of silkworms. The shape of mulberry leaves can also influence their suitability for sericulture. Leaves with a rounded or ovate shape are preferred as they are easier for silkworms to consume (Murthy *et al.*, 2011) [15]. The nutrient content of mulberry leaves is critical for the growth and development of silkworms. The leaves should be high in protein, carbohydrates, and vitamins to support the growth and development of silkworms. Mulberry varieties with good disease resistance are preferred as they can minimize the risk of disease outbreaks, which can be detrimental to sericulture. High yield is also an essential factor in determining the suitability of mulberry varieties for sericulture. Mulberry varieties with high yields can provide a steady supply of leaves for silkworms throughout their lifecycle. The project will use a PYT (Participatory Yield Trial) approach, which involves working closely with local farmers and stakeholders to ensure that the research is relevant and applicable to the needs of the community. In addition to measuring quantitative traits, such as biomass yield, the research will also involve a qualitative assessment of the genotypes, which will include biochemical tests to analyze the chemical composition of the foliage, propagation tests to determine the ease of propagation, and silkworm moulting tests to evaluate the suitability of the foliage for feeding silkworms (Snapp *et al.*, 2019) [26]. To ensure the production of high-quality leaves, factors such as leaf moisture content, moisture retention capacity after six hours of harvest, total soluble protein, and total soluble sugar content must be considered, alongside the severity of disease (measured as percent disease incidence or PDI) and insect infestation. Leaf quality can also be assessed using a silkworm bioassay, which takes into account metrics such as mature larval weight, leaf-to-cocoon ratio, and silk recovery. The ultimate goal of the research is to identify promising mulberry seedlings that can be further utilized for silkworm production, which could have significant economic benefits for the community (Giora *et al.*, 2022) [11].

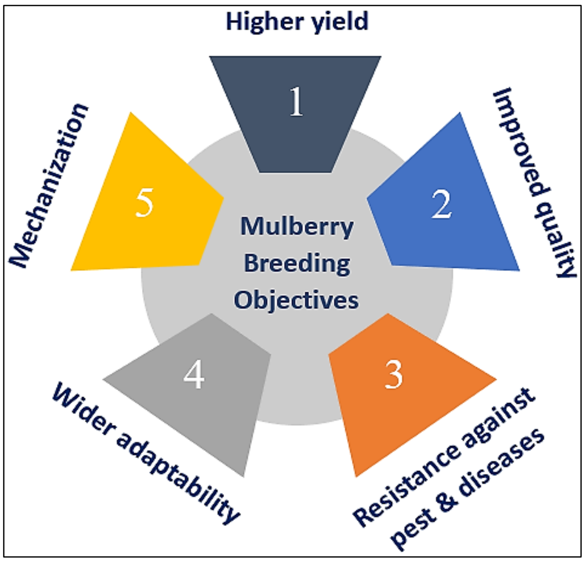

Fig 1: Pictorial overview of mulberry breeding objectives

5. Steps in mulberry hybrid development

In this review paper, we will discuss the various methods used for the development of high yielding mulberry varieties with vigorous vegetative growth and early sprouting through controlled pollination. The review will cover the following areas:

1. Identification of desirable traits for mulberry varieties.
2. Selection of parent plants and controlled pollination.
3. Evaluation of the resulting progenies (Vijayan *et al*., 2014) [32].

A particular process is required for mulberry genetic improvement by conventional breeding, as indicated in Figure 2. Germplasm accessions are first characterized by their morphological, biochemical, physiological, rooting, leaf yield, moisture, protein, sugar and photosynthetic efficiency properties. Parents with desirable qualities are chosen, and controlled hybridization is carried out, based on statistical analysis. The mature fruits of a controlled and natural hybridization of chosen mother plants are harvested to obtain the seeds (Sadia *et al*., 2021) [21]. In progeny row trials (PRT), seedlings are nurtured in nurseries before being transferred to the field to be tested for growth, branching, leaf texture, and disease susceptibility. Traditional breeding emphasizes the development of F_1 hybrids because of the high heterozygosity and hybrid vigour. Primary yield trials (PYT) are used to further assess promising hybrids from PRT for critical agronomic, biochemical and silkworm feeding characteristics. During final yield trials (FYT), which consist of 3-5 replications and 25-49 plants for each replication. In the trial, the top 5-10% hybrids from PYT are chosen for further consideration. In FYT, the leaf production, quality, adaptability, sensitivity to pests and diseases, rooting capacity, responsiveness to agronomic methods and silkworm feeding attributes are all thoroughly assessed. The top hybrid is vegetatively mass-produced for additional testing in various areas (MLT). In most cases, regional multi-location studies employ 8-9 hybrids. To assess their performance in various agro-climatic conditions in India, the MLT's consistent performers are chosen and tested further in the All India Coordinated Experiments on Mulberry (AICEM) for at least four years. AICEM releases its top performers for commercial cultivation and as a result, several high-yielding mulberry types have been developed (Vijayan *et al*., 2012) [33].

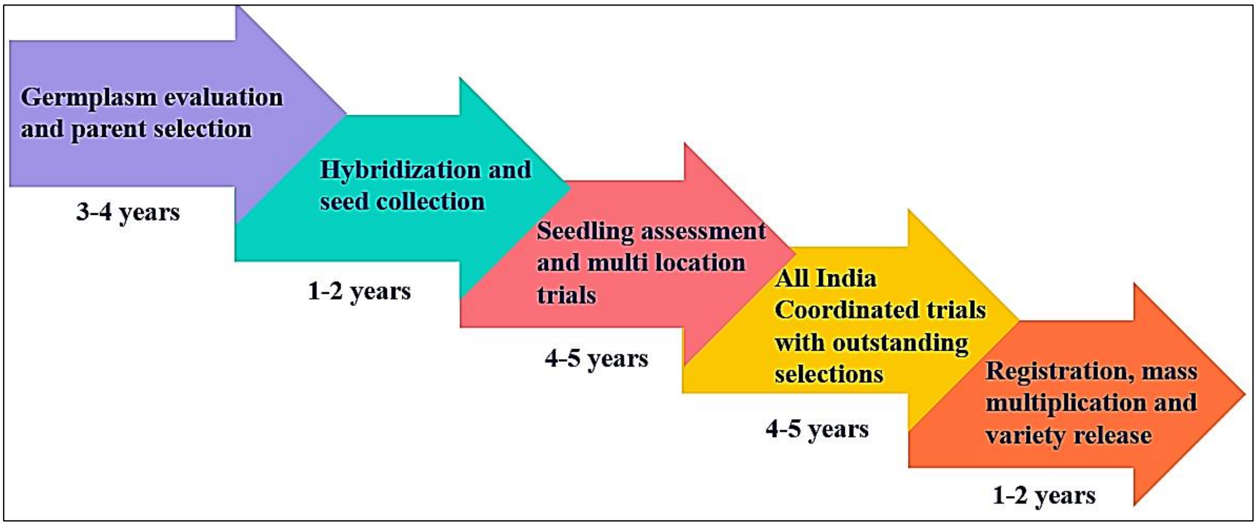

Fig 2: Flow diagram of variety development and release in mulberry

6. Case studies

Desirable traits for mulberry varieties include high yield, rapid vegetative growth, early sprouting, and resistance to biotic and abiotic stresses. Conventional varieties have fewer no of these qualitative traits and also could not break the leaf yield plateau. That is why scientists are going for developing varieties having a combination of good leaf yield as well as qualitative traits suitable for silkworm feeding (Table 3). Parent plants with these traits can be selected using various techniques such as mass selection, pedigree selection, and then hybridization is performed. Controlled pollination techniques such as hand pollination, bagging and emasculation can be used to control the pollination process and ensure the desired traits are passed on to the progenies. Evaluation of the resulting progenies can be done using various methods such as field trials and laboratory analysis (http://www.csrtiber.res.in).

A total of nine novel genotypes (C-11, C-01, C-384, C-05, C-45, C-212, C-09, C-02, C-108) with promising traits, as well as the well-known variety S1635 as a control, were studied across four sites in Eastern and North-Eastern India to evaluate leaf yield and quality across various seasons as final yield trials (FYT) cum multilocation trials (MLT). Seasonal production of 7.05 (C-45) to 11.41 (C-11) t/ha/year on average was produced. C-11, C-01 and C-02 among them showed yields greater than 10 t/ha/crop. The typical leaf yield for the winter harvests (Agrayani and Falguni) ranged from 6.95 to 10.45 t/ha. Interestingly, S1635 exhibited much lower winter leaf yields than C-11, C-01, C-384, C-05, C-212 and C-02. Seven genotypes exhibited considerably greater annual leaf yields, which varied from 35.26 to 57.06 t/ha/year. In irrigated circumstances, these genotypes outperformed the S1635 variety in terms of yield by 9% to 35%. In three different rainfed environments over several seasons, the leaf yield and quality of nine test genotypes were evaluated. With the exception of days to sprout, primary branches, leaf wetness and moisture retention capacity, genotype differences for all examined variables were found to be highly significant, according to the results of the combined analysis of variance. Growth characteristics such as days to sprout (9-12 days), 100 leaves weight (250-289 g), leaf wetness (74-77.44%), retention capacity (79-81.50%), length of tallest shoot (124-147 cm) and total shoot length all showed significant variance (600-763 cm). Particularly, the growth rates of C-11, C-01, C-384, C-108 and C-05 were greater than those of the check (Annual report 2020-21, CSRTI, Berhampur).

Due to Kashmir's temperate environment, mulberry trees can thrive and silkworms can be raised, producing raw silk that satisfies international standards. Study in the area has outlined the advantages and disadvantages of regional and imported types, particularly those from Italy and Japan. These types have strong stem canker resistance, although they sprout later in the spring, which can affect paddy farming and raising. Also, many types require time-consuming, expensive root grafting since they cannot be reproduced using stem cuttings. On the other hand, while early sprouting types that may be grown from stem cuttings have lower yields, they are somewhat resistant to stem canker. Controlled pollination, open pollination, and mixed pollination were used to induce diversity in order to establish new high-yield types that can withstand cold and stem canker and are ideal for the rain-fed conditions of Kashmir. The selections were assessed and shortlisted based on their average Evaluation Index after pooling data from two years for various yield-attributing factors (Mano *et al*., 1993). Superior selections, including S-38, S-85, S-106, S-140, S-145, S-152, S-158, S-186, S-213, S-217, and S-113, yield between 2.9 and 4.3 kg of product per plant each year as a result of the study. All of these selections, with the exception of S-38, have noticeably bigger leaves than Chinese White, which yields 1.7 kg of product per plant each year and has leaves that weigh 0.5 kg. They all have a weight of about 1.0 kg per 100 leaves. These cultivars, which resemble Chinese White, may be produced by stem cuttings, and their winter buds develop quickly. They are tolerant to stem canker (Bindroo & Kirsur, 2013) [6].

Table 3: Developed mulberry hybrids in India

Hybrid	Region/soil type	Developed at	Pedigree	Yield (MT/ha/year)
Victoria-1	South India, Irrigated	CSRTI, Mysore	Hybrid from S30 x Berc 776	35-40
Sahana		CSRTI, Mysore	Hybrid from K2 x Kosen	25-30
RC1 (Resource constraint-1)	South India, Irrigated	CSRTI, Mysore	Hybrids from Punjab local x Kosen	45-50
AR12 (Alkalaine tolerant)	Region having alkaline soils	CSRTI, Mysore	Hybrid from S-41 (4x) x Ber. C-776	23-25
C-2038	Eastern and NE India, Irrigated	CSRTI, Berhampore	Hybrid from $CF_1$10 × C763	53-54
C-2028	Eastern and NE India, Irrigated	CSRTI, Berhampore	Hybrid from China White x S-1532	36-37
C-2058 (C-9)	Eastern and NE India, Irrigated	CSRTI, Berhampore	Hybrid from Berhampore-A × Shrim-2	34-35
C-2060 (Gen-1)	Eastern and NE India, Irrigated	CSRTI, Berhampore	Hybrid from Kajli OP × V-1	58-60
C-1360 (Ganga)	Eastern and NE India, Irrigated	CSRTI, Berhampore	Hybrid from Philippines × Vietnam-2	57
MSG2	South India, semi-arid tropics	CSRTI, Mysore	Hybrid from BR-4 and S13	22-23
AGB8	South India, semi-arid tropics	CSRTI, Mysore	Hybrid from (Sujanpur-5 x Phillipines) and (K-2 x Black cherry)	47-50

7. Miscellaneous aspects on mulberry

Mulberry plants are of great importance in the sericulture industry as they provide food for silkworms. The development of high yielding mulberry varieties with vigorous vegetative growth and early sprouting is a time-consuming and labour-intensive process, as it involves controlled pollination techniques. This process requires skilled labour, proper selection of parent plants, and strict monitoring of hybridization processes to ensure desired traits are passed on to the offspring. However, the benefits of having high yielding mulberry varieties are enormous. High yielding varieties lead to increased silk production, which can lead to higher profits for sericulture farmers. The silk industry is a major contributor to the economy of many countries, and increased silk production can have a positive impact on the overall economy (Bhalla *et al*., 2020) [5]. In addition to high yields, rapid vegetative growth and early sprouting allow for multiple harvests in a year. This is particularly beneficial in areas with suitable climatic conditions, where farmers can take advantage of multiple growing seasons to increase their productivity and profitability. Moreover, early sprouting also leads to early maturity, which means farmers can start harvesting earlier and have a longer harvesting period. This, in turn, reduces the risk of losses due to unpredictable weather changes (Raza *et al*., 2019) [17]. By traditional breeding techniques that focus on agronomic and morpho-physiological based phenotyping, productive mulberry varieties that can withstand drought, alkalinity, salinity, water logging and cold have been successfully developed (Sarkar *et al*., 2017) [23]. The mulberry germplasm collections, comprising wild spp., alien and native genotypes, have been identified and conserved. These collections display sexual polymorphism (dioecious, monoecious and bisexual), genotypic and phenotypic differences for significant agronomic variables, plant insect/microbe interaction, biotic/abiotic stress tolerance and attributes linked to nutritional and therapeutic benefits (Razdan & Thomas, 2021) [18]. Several of the germplasm collections have been used to create strain-tolerant mulberry cultivars. The development of introgression lines of mulberry with high Water use efficiency (WUE) and root traits has been made possible by physiological trait-based breeding techniques. These lines could subsequently be used for foliage yield evaluation trials, as a pre-breeding resource in breeding programmes, and for the validation of quantitative trait loci (QTLs) linked to mulberry drought adaptive traits (Sarkar *et al*. 2017) [23]. Although a sizable portion of mulberry species used for commerce are diploid in nature, high yielding triploid mulberry species are employed in various regions of India for raising silkworms. A natural or forced tetraploid x diploid hybridization produces a triploid mulberry genotype, or one is chosen from naturally occurring triploid populations. In terms of growth pattern, leaf output and nutritional properties, the majority of triploid mulberry plants outperform diploids. Due to their sterility and inability to set seeds, desired features of triploid mulberries might be preserved by clonal multiplication without any loss of leaf production potential (Vijayan *et al*., 2022) [31]. Moreover, C4-like photosynthetic 'trait' system, improved water, nitrogen and radiation usage efficiency and better resistance to high temperature and drought conditions were physiological features seen in mulberry germplasm. Mulberry is a C3 perennial tree, however, a previous study found that triploids have a C4 photosynthetic "syndrome"; these cultivars fixed more CO_2 in the form of four-carbon compounds than do diploid high-yielding cultivars. Moreover, triploid cultivars displayed higher PEPC (phosphoenolpyruvate carboxylase) activity compared to Rubisco (ribulose 1, 5-bisphosphate carboxylase-oxygenase) (Shi *et al*., 2021) [25]. For the purpose of genetically enhancing mulberries, hybridization-based traditional breeding methods have been used. These methods include a well-defined set of activities like gathering exotic or indigenous germplasm within the nation or abroad, assessing germplasm collections for a trait of interest, choosing suitable parents for use in hybridization based on traits pyramiding, crossing chosen parents (diploid x diploid, tetraploid x diploid), and backcrossing (Arunakumar *et al*., 2021) [4]. Following the collection of F_1 seeds from a cross or backcross, seedlings are raised in nursery beds, F_1 seedlings from a single, multiple cross, or backcross populations are planted in progeny-row-trail (PRT) plots for initial screening, and then a selection of F_1 seedlings or backcross progenies are chosen and evaluated based on traits of agronomic interest and foliage yield (Vijayan *et al*., 2006) [34]. These tactics include open-pollinated cross-pollination techniques including clonal selection, selection from natural variability, and selection of hybrids. Mulberry breeding mainly relies on the generation of F_1 hybrids and subsequent vegetative propagation of hybrids. Primary-yield trials (PYT) are used to assess F_1 hybrids with desired agronomic features, and subsequently final-yield trials are used to assess them (FYT). For additional testing in multi-location trials, the best-performing hybrid offspring from FYT is chosen and replicated by vegetative propagation (MLT). In order to assess how well the chosen mulberry hybrids/triploids perform under various agro-climatic conditions in India, they are further examined in the All India Coordinated Experimental Trials for Mulberry (AICEM). Final recommendation and release of the most prolific, resilient to stress, and climate-tolerant hybrid offspring for industrial culture for silkworm rearing. The passage also discusses capacity building in conjunction with genomic selection and physiological breeding methods using high throughput phenotyping systems in mulberry. This strategy could introduce characteristics linked to stress tolerance, leaf yield, and climatic resistance (Reynolds & Langridge, 2016) [19]. A commercial mulberry cultivar is developed through a lengthy, rigorous and trial-and-error breeding and assessment procedure that generally lasts 15 years.

8. Limitations and future prospects

Mulberry is an important crop for silk production, and mulberry leaves are the primary food source for silkworms. Early sprouting varieties of mulberry trees have the advantage of producing leaves earlier in the season, but they have a significant disadvantage of early senescence, which means that leaves start to deteriorate and fall off earlier in the season than desirable. The early senescence of leaves is a problem because it reduces the availability of high-quality leaves for silkworms, which ultimately affects silk production (Vijayan, 1970) [30]. To address this issue, breeders should focus on developing mulberry varieties that exhibit stay-green traits. Stay-green traits allow the leaves to remain green and healthy for a more extended period, which increases the availability of high-quality leaves for silkworms and ultimately improves silk production. Stay-green traits are complex genetic traits that allow the leaves to retain their greenness and photosynthetic capacity for a more extended period. These traits are essential for maintaining leaf quality and longevity, especially in environments with a high rate of leaf senescence (Kamal *et al*., 2019) [13]. By breeding for stay-green traits, breeders can develop mulberry varieties that produce high-quality leaves for a more extended period, leading to increased silk production. Advances in molecular biology and genomics have made it feasible to isolate genomic- and genic-SSR markers (microsatellites) (Das *et al*. 2022) [8]. These markers were discovered using a variety of resources, including genomic clones, a genomic library, EST sequences, and mulberry transcriptome data. Moreover, it has been discovered that they can spread to closely related species. Then, utilising molecular markers like RAPD and ISSR, researchers tried to create QTL maps for significant agronomic features in mulberry, such WUE, root traits, and yield-contributing factors. Just a small number of markers, nevertheless, were discovered to be associated with the QTLs that control these important features (Desai *et al*., 2021; Ghosh *et al*. 2022) [9, 10]. Using conventional or marker-assisted breeding techniques, abiotic stress tolerant transgenes from well-characterized transgenic mulberry lines might be introduced into various genetic backgrounds of elite cultivars, as demonstrated in mulberry (Sarkar *et al*., 2017) [23]. As the C4 photosynthesis mechanism is more effective than the C3 photosynthesis system, key genes responsible for C4 photosynthesis, such as those that code for enzymes or transporter proteins or a modified form of the gene coding for Rubisco, can be introduced into mulberry to increase its resilience to climate change (Wang *et al*., 2012; Maity *et al*., 2023) [35, 14]. Moreover, CRISPR/Cas9 systems may be employed as a genome editing technique to improve mulberry's climate resilient features and to elucidate the molecular underpinnings of abiotic stress tolerance (Sami *et al*., 2021) [22]. An integrated strategy that incorporates conventional breeding, VIGS and RNA interference-based reverse genetics, genomic tools, *in vitro* plant regeneration, and genetic transformation protocols, along with adequate funding, capacity building, scientific networking and concerted efforts on biosafety clearance, would be necessary milestones to achieve sustained development and commercialization of climate resilient mulberry cultivars (Xiangyun, 2010) [36].

9. Conclusion

The development of high yielding mulberry varieties with vigorous vegetative growth and early sprouting through controlled pollination is important for the success of the sericulture industry in tropical agroclimatic regions. Various methods can be used to achieve this goal, including the selection of desirable traits, selection of parent plants, controlled pollination techniques and evaluation of progenies. The resulting varieties will have a significant impact on the sericulture industry, leading to increased profits for farmers and higher quality silk for consumers.

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11. Contribution of the authors

Writing of manuscript: HD, SB, AS, and SD; Curation and arrangement of tables: HD, AP, RBR, KKP, and SD; Manuscript correction: SD and KB.

12. Conflicts of interest

Authors declare that no conflict of interest exists.

13. References

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