



# **Emerging Nutritional and Climate-resilient Strategies in Mulberry for Enhanced Cocoon Quality in Sericulture**

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## **Authors' contributions**

*This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.*

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## **ABSTRACT**

Sericulture relies fundamentally on the nutritional quality of mulberry (*Morus* spp.) leaves, the exclusive feed for the silkworm, *Bombyx mori* L. In recent years, efforts have intensified to improve cocoon yield and silk quality through strategic nutritional enrichment of mulberry foliage and the adoption of climate-resilient cultivation practices. This review outlines how integrating soil and foliar

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nutrient supplementation particularly with elements like nitrogen, zinc, and iron alongside organic amendments and microbial inoculants such as plant growth-promoting rhizobacteria (PGPR) and mycorrhizal fungi, can significantly enhance leaf quality. Improved leaf nutrition directly benefits key cocoon characteristics, including larval growth, cocoon weight, shell percentage, filament length, and feed utilization efficiency. Moreover, cultivation techniques tailored to mitigate climatic stresses such as the use of drought-tolerant mulberry genotypes, mulching, and efficient irrigation systems help maintain consistent leaf yield and quality. By combining targeted nutrient strategies with climate-smart agronomic practices, sericulture can achieve higher productivity and superior cocoon traits while reducing dependence on synthetic inputs, lowering production costs, conserving natural resources, and minimizing environmental impact thereby aligning silk production with long-term economic viability and ecological sustainability.

**Keywords:** *Sericulture; mulberry leaf quality; nutritional enrichment; climate-resilient cultivation; plant growth-promoting rhizobacteria (PGPR); mycorrhizal fungi; cocoon yield; sustainable silk production; micronutrient supplementation; climate-smart agriculture.*

## 1. INTRODUCTION

The rearing success of the silkworm (*Bombyx mori* L.) is fundamentally dependent on the quality of mulberry (*Morus* spp.) leaves, which serve as the exclusive nutritional source throughout its larval life cycle. The biochemical composition of mulberry foliage especially its protein, carbohydrate, moisture content, and micronutrient levels play a pivotal role in determining larval development, silk gland function, and ultimately cocoon yield and filament quality (Kusuma et al., 2020).

Traditionally, sericulture practices have focused on maximizing mulberry leaf biomass. However, recent research and field applications have shifted the emphasis toward leaf quality, recognizing that nutrient-rich and physiologically balanced foliage is essential for enhancing feed conversion efficiency, cocoon weight, and shell percentage. Field-level challenges including soil nutrient depletion, climatic variability, and imbalanced fertilization have made it increasingly difficult to sustain consistent leaf quality (Tilahun, 2020).

To address these limitations, innovative approaches focusing on nutritional enhancement and climate-resilient agronomic practices are gaining ground. Nutrient enrichment using both chemical and organic sources, along with microbial consortia like Plant Growth-Promoting Rhizobacteria (PGPR) and arbuscular mycorrhizal fungi (AMF), has been shown to improve mulberry physiology and increase the bioavailability of critical nutrients in the leaves (Rani et al., 2018). These interventions have demonstrated significant improvements in

silkworm health and cocoon traits under both laboratory and field conditions.

Parallely, climate-adaptive strategies, including the adoption of drought-tolerant mulberry genotypes, mulching, intercropping, and water-conserving irrigation systems, are being explored to stabilize leaf yield and quality in regions facing heat stress, erratic rainfall, and water scarcity (Rahmathulla, 2012). Such techniques not only sustain plant growth under adverse environmental conditions but also support year-round silkworm rearing with minimal ecological impact. Additionally, fortified feeding strategies such as the use of fermented leaves or supplements enriched with amino acids, vitamins, and microbial proteins have been reported to enhance silk gland development and filament synthesis, further contributing to cocoon productivity (Kavitha et al., 2014). This review synthesizes emerging knowledge on the integration of nutritional and climate-resilient strategies in mulberry cultivation. It highlights their role in enhancing the biological efficiency of *Bombyx mori*, improving cocoon parameters, and promoting sustainable sericulture. It also outlines practical insights for field-level adoption and future research priorities for eco-friendly cocoon yield enhancement.

## 2. NUTRITIONAL COMPOSITION AND QUALITY OF MULBERRY LEAVES

The quality and nutritive value of mulberry (*Morus* spp.) leaves are fundamental to the success of silkworm rearing. Various physiological, biochemical, and morphological characteristics of mulberry influence the feeding behavior, digestion efficiency, and silk-producing ability of *Bombyx mori*. The nutritional profile of

mulberry is not static; it varies with cultivar type, plant age, season, agronomic practices, and soil fertility (Chandrakala & Fatima Sadatulla, 2020).

Mulberry leaves are rich in essential nutrients such as proteins, carbohydrates, minerals, vitamins, and secondary metabolites. These nutrients influence several biological traits in silkworms, including larval weight, cocoon shell percentage, and silk filament length (Aramani et al., 2025; Shanthi Sree et al., 2024). Among the different mulberry varieties, genotypes such as V1, S36, Thailand Male, and M5 are known for their superior nutritional traits, especially in terms of high protein and moisture content (Rani et al., 2018).

### **2.1 Macronutrient Profile: Proteins, Carbohydrates, and Moisture**

Soluble proteins and reducing sugars are the most critical leaf constituents influencing larval development. High protein content in leaves supports silk gland growth, while sugars provide the metabolic energy required for feeding and spinning (Kumar et al., 2019). The protein-to-carbohydrate ratio plays a crucial role in determining food conversion efficiency and silk output.

### **2.2 Micronutrients and Minerals in Leaf Physiology**

Moisture content contributes significantly to the palatability and digestibility of leaves. Young and tender leaves generally have higher moisture, making them more suitable for early instar larvae. High crude fiber in mature or coarse leaves reduces digestibility, adversely affecting cocoon production (Bu et al., 2022).

### **2.3 Influence of Leaf Maturity and Seasonal Variability**

Mulberry leaves supply essential minerals such as calcium (Ca), potassium (K), magnesium (Mg), and trace elements like zinc (Zn) and iron (Fe), which are important for various enzymatic and metabolic activities in silkworms. These nutrients play a vital role in silk protein synthesis, immune functions, and overall larval vigor (Lakshmi & Ramesh, 2016; Mdpi, 2024; Zhang et al., 2023).

### **2.4 Genotypic Variation in Nutritional Value**

The stage of leaf maturity also affects nutrient content. Generally, medium-matured leaves (5th or 6th leaf from the apex) are nutritionally optimal, containing a balanced level of protein, sugars, and minerals (Ramesha et al., 2010). Additionally, biofortification aids in restoring degraded soils and promotes carbon sequestration through increased root biomass (Ramesha et al., 2010). Over-aged leaves may accumulate more lignin and fiber, which can hinder digestion and reduce cocoon weight.

### **2.5 Seasonal Influence**

Leaf quality is significantly affected by seasonal changes. During the monsoon and cooler months, mulberry tends to produce nutrient-rich foliage due to better soil moisture and lower evapotranspiration. In contrast, summer leaves often show reduced chlorophyll, moisture, and protein content due to heat stress (Rahmathulla, 2012).

Overall, the nutritional composition of mulberry leaves is a key determinant of silk productivity. Selecting high-yielding and nutritionally rich genotypes, along with appropriate cultivation and harvesting practices, can significantly improve cocoon yield and silk quality.

## **3. NUTRITIONAL STRATEGIES TO ENHANCE COCOON TRAITS IN *Bombyx mori***

Improving the nutritional quality of mulberry (*Morus* spp.) leaves is a critical frontier in sustainable sericulture. Since silkworms (*Bombyx mori* L.) derive their entire diet from mulberry foliage, the nutritional profile of the leaves directly impacts larval development, cocoon quality, and silk yield. Biofortification refers to the enhancement of the nutritional content of crops via agronomic practices, selective breeding, organic enrichment, or biological interventions such as microbial inoculation. In mulberry, biofortification strategies are increasingly recognized as a sustainable means to improve leaf protein, micronutrient density, and digestibility without relying on synthetic chemical inputs.

### 3.1 Conventional Breeding for Nutrient-Rich Mulberry Genotypes

Traditional mulberry breeding programs have successfully produced cultivars with superior nutritional quality. Improved varieties such as V1, S36, and G4 consistently exhibit higher crude protein, moisture content, and soluble sugar levels compared to traditional types like Mysore Local or Berhampore, leading to enhanced leaf quality (Lakshmi & Ramesh, 2016; Mdpi, 2024) and improved silkworm rearing performance (Aramani et al., 2025; Shanthi Sree et al., 2024). These high-yielding genotypes not only support better larval weight gain and survival but also contribute to an improved cocoon shell ratio and filament length (Dandin et al., 2003). Breeding efforts now increasingly focus on integrating multi-trait selection, including disease resistance, leaf yield, and nutrient bioavailability, to meet the dual goals of sustainability and productivity (Naik et al., 2018).

### 3.2 Micronutrient Supplementation and Fertilizer Management

Agronomic biofortification through foliar and soil application of micronutrients particularly zinc (Zn), iron (Fe), boron (B), and magnesium (Mg) has proven effective in enhancing mulberry leaf quality. Zinc application, for instance, has been reported to enhance photosynthetic efficiency, protein synthesis, and antioxidative enzyme activities in mulberry (These effects are attributed to enhanced enzymatic activities, higher photosynthate availability, and better nitrogen metabolism in the plant, translating into improved nutrient transfer to the silkworm (Sarkar et al., 2017). Similarly, iron is essential for chlorophyll formation and energy metabolism, and its supplementation has been linked with increased leaf biomass and nitrogen assimilation (Lakshmi & Ramesh, 2016; Harish Reddy et al., 2024). The synergistic application of Zn, Fe, and Mn significantly improves silk gland growth in silkworms and enhances cocoon parameters (Chandrakala & Fatima Sadatulla, 2020; Lokanath & Shivashankar, 1986). Recent studies emphasize that optimal micronutrient application schedules can improve leaf protein content, moisture retention, and palatability, ultimately boosting larval development and silk quality (Harish Reddy et al., 2024).

### 3.3 Organic Amendments and Integrated Nutrient Management (INM)

Organic matter amendments such as farmyard manure (FYM), green manure, vermicompost, and enriched composts provide essential macro- and micronutrients, improve soil microbial diversity, and enhance the bioavailability of nutrients in mulberry foliage. Rani et al. (2018) demonstrated that long-term application of compost improved leaf nitrogen and phosphorus levels while reducing the need for chemical fertilizers. Moreover, organic inputs contribute to improved soil structure, water-holding capacity, and cation exchange, all of which indirectly benefit leaf nutritional quality (Manjunatha et al., 2019; Reddy et al., 2020). Integrated nutrient management (INM) a combination of organic and inorganic fertilizers offers a balanced approach for sustained productivity and nutrient-rich foliage under varying agro-climatic conditions.

### 3.4 Plant Growth-Promoting Rhizobacteria (PGPR)

Biofertilization using beneficial microbes like *Azospirillum*, *Azotobacter*, *Bacillus subtilis*, and *Pseudomonas fluorescens* is an emerging trend in mulberry biofortification. PGPR enhances nitrogen fixation, phosphate solubilization, and phytohormone production, thereby increasing plant vigor and leaf nutritional content (Rani et al., 2018). PGPR-treated mulberry plots have shown significant increases in leaf protein and chlorophyll content, leading to better cocoon quality and larval survival rates (Rani et al., 2018). Co-inoculation with arbuscular mycorrhizal fungi (AMF) further improves root nutrient uptake, drought resistance, and overall plant health (Bhadra et al., 2019). This biological approach is especially effective in low-input farming systems and degraded soils.

### 3.5 Fermented and Fortified Leaf Supplements

Innovative strategies such as leaf fermentation and post-harvest nutrient fortification are gaining attention for their ability to directly enhance silkworm nutrition. Fermented mulberry leaves using microbial cultures, yeast extracts, or amino acid solutions increase digestibility, improve gut health in larvae, and stimulate silk gland development (Kavitha et al., 2014). Studies have shown that *Spirulina*- and *Chlorella*-based

mulberry leaf enrichments enhance larval metabolism and silk protein synthesis, especially during late instar stages (Rao et al., 2016). These approaches offer flexibility for targeted nutritional interventions, particularly in controlled rearing conditions or commercial sericulture units.

#### 4. IMPACT OF BIOFORTIFIED MULBERRY ON SILKWORM PERFORMANCE

The nutritional status of mulberry leaves plays a decisive role in shaping the biological efficiency of *Bombyx mori* L. Silkworms are highly responsive to changes in their diet, and even slight improvements in leaf quality can bring about significant enhancements in growth, survival, and silk production. Biofortified mulberry leaves, enriched through agronomic, microbial, or post-harvest methods, have shown promising results in improving several larval and cocoon traits.

The link between leaf nutrition and silkworm physiological response is depicted in Fig. 1, which summarizes how improved nutrient content supports silk gland development and cocoon formation through enhanced metabolic activity, nutrient absorption, and enzyme regulation.

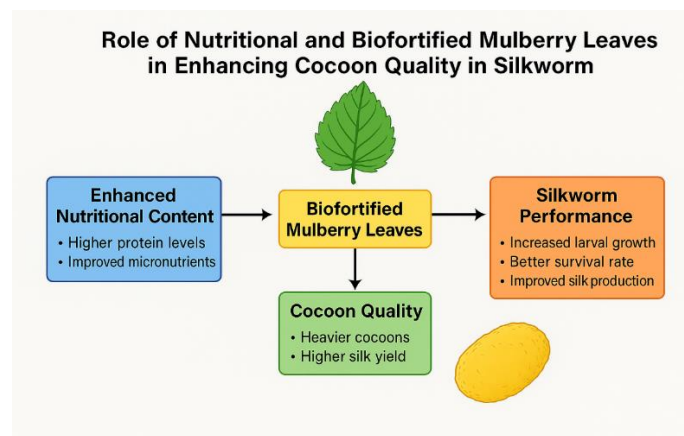
##### 4.1 Larval Growth and Survival

Improved nutritional quality of mulberry leaves characterized by elevated crude protein, essential amino acids (particularly alanine, glycine, and serine), and readily digestible carbohydrates is critical for enhancing larval growth, body weight gain, and developmental

uniformity in *Bombyx mori* (Mdpi, 2024; Ponnusamy et al., 2023). Such nutrient-rich foliage significantly promotes silk gland tissue development and larval growth during the fourth and fifth instars, ultimately improving cocoon quality and silk production (Mdpi, 2024; Ponnusamy et al., 2023; Zhang et al., 2023). Nutritionally superior leaves result in better feed intake and higher assimilation efficiency, leading to shorter larval durations and increased biomass accumulation (Tilahun, 2020).

Several studies have demonstrated the significant impact of micronutrient supplementation and biological interventions on larval performance. For instance, foliar application of zinc and iron has been shown to enhance protein synthesis and promote overall silkworm vigor (Lakshmi & Ramesh, 2016; Srivastava et al., 2020). Similarly, inoculation of mulberry roots with plant growth-promoting rhizobacteria (PGPR) such as *Azospirillum* and *Pseudomonas fluorescens* improves nitrogen uptake and phytohormone production, resulting in nutritionally enriched leaves that support up to 25% greater larval weight and higher survival percentages (Rani et al., 2018).

Additionally, the use of organic soil amendments like vermicompost and enriched farmyard manure (FYM) has been reported to improve leaf nitrogen and phosphorus content, translating into accelerated larval growth and reduced mortality (Kumar et al., 2018; Ramesha et al., 2023). Under controlled experiments, silkworms reared on fermented or amino acid-fortified mulberry leaves exhibited superior metabolic performance and better synchronization of instars compared to those reared on conventional leaves (Kavitha et al., 2014).



**Fig.1. Schematic illustration showing the impact of enhanced nutritional content in biofortified mulberry leaves on silkworm performance and cocoon quality**

Such improvements not only contribute to the uniformity of silkworm development and batch-wise cocoon harvest but also form the basis for standardized cocoon quality parameters essential for commercial silk production. Therefore, optimized mulberry leaf nutrition stands as a critical factor in achieving enhanced silkworm productivity and economic sustainability of sericulture systems.

#### 4.2 Cocoon Weight and Shell Ratio

Cocoon weight and shell ratio are two of the most critical commercial parameters in sericulture as they directly determine silk yield and quality. These traits are highly responsive to the nutritional composition of the mulberry leaves consumed by *Bombyx mori*. Elevated levels of crude protein, total sugars, and essential micronutrients in the foliage have been shown to significantly improve cocoon formation by enhancing silk gland function and protein synthesis (Rahmathulla, 2012).

Silkworms enter their peak nutritional demand during the fifth instar, during which silk proteins like fibroin and sericin are synthesized in large quantities. Feeding with leaves fortified with zinc, iron, and magnesium either through foliar application or PGPR-based biofertilizers has been reported to improve cocoon weight by 10–20% and shell weight by 15% or more compared to untreated controls (Patil et al., 2014).

These effects are attributed to enhanced enzymatic activities, higher photosynthate availability, and better nitrogen metabolism in the plant, translating into improved nutrient transfer to the silkworm (Sarkar et al., 2017). Improved nutritional quality of mulberry leaves plays a pivotal role in enhancing larval growth, body weight gain, and developmental uniformity in *Bombyx mori*. High levels of crude protein, amino acids, and digestible carbohydrates in the foliage facilitate rapid cell division and tissue development, especially in the silk glands during the fourth and fifth instars (Mdpi, 2024; Ponnusamy et al., 2023; Zhang et al., 2023).

Varieties such as V1 and S36, when cultivated under integrated nutrient management regimes, consistently produce leaves that result in heavier cocoons and superior shell ratios (Datta, 2000; Mdpi, 2024).

#### 4.3 Silk Filament Length and Quality

The length, strength, and fineness of the silk filament are essential characteristics that

determine the commercial value of the cocoon. These parameters are influenced not only by genetic factors but also by the nutritional quality of the larval diet. Silkworms fed with mineral-enriched, fermented, or amino acid-fortified mulberry leaves have been found to produce significantly longer and more uniform filaments with superior reelability and denier consistency (Kavitha et al., 2014).

Leaf supplementation with Spirulina, Chlorella, or protein hydrolysates enhances the amino acid composition of the larval hemolymph, thereby facilitating more efficient fibroin and sericin biosynthesis in the silk glands (Joshi et al., 2002). This leads to silk threads that are finer, stronger, and less prone to breakage. For instance, Kavitha et al., (2014) reported up to an 18% increase in filament length and notable improvement in uniformity and tenacity when larvae were reared on yeast-fermented leaves.

Micronutrients such as zinc, manganese, and iron also play essential roles in enzymatic activation and oxidative metabolism within the silk gland. Their application through foliar sprays or microbial mediation (PGPR/AMF) enhances the physiological condition of the larvae and stimulates the sericigenic activity of the posterior silk gland (Chandrakala & Fatima Sadatulla, 2020). Furthermore, feeding on organically enriched leaves has been shown to yield cocoons with a higher raw silk percentage and reduced waste silk (Terefe et al., 2018).

Field and lab-based studies support the view that optimizing mulberry leaf nutrient profiles either through agronomic practices, microbial inoculation, or post-harvest supplementation can significantly enhance filament characteristics, meeting the needs of both traditional and commercial silk markets.

#### 4.4 Immunity and Disease Resistance

Nutritional quality of the diet significantly influences the immune competence of silkworms. Leaves enriched with micronutrients like zinc and magnesium support enzymatic and immunomodulatory functions, including antioxidant defense and stress response mechanisms (Marin et al., 2022). Silkworms fed on such fortified diets demonstrate higher resistance to microbial infections such as flacherie, grasserie, and muscardine.

Antioxidants and secondary metabolites in organically or biologically enriched mulberry

leaves such as flavonoids, phenolics, and alkaloids can significantly reduce oxidative stress during critical growth stages in *Bombyx mori*, thereby supporting larval health and silk gland activity (Choudhury et al., 2023; Saha et al., 2024). Experimental studies have shown that feeding with PGPR-treated leaves can reduce larval mortality by up to 30%, enhance hemocyte activity, and stimulate enzyme systems such as catalase and peroxidase (Rani et al., 2018). These traits are crucial for maintaining the health and biosecurity of silkworm populations under both controlled and open-rearing systems.

To support the above findings, Table 1 compiles experimental results from various studies

evaluating the impact of different mulberry biofortification methods on larval performance and cocoon traits under controlled conditions and Table 2 further illustrates the mode of action of various biofortification approaches and their observed impact on silkworm biology, highlighting the practical relevance of these interventions.

The use of biofortified mulberry leaves has a multifaceted impact on silkworm biology. Enhanced leaf nutrition improves larval performance, cocoon productivity, silk quality, and disease resistance. These benefits demonstrate the potential of nutritional enhancement as a low-cost, high-impact intervention in sustainable sericulture practices.

**Table 1. Compiled results from previous studies on the impact of different biofortification methods in mulberry cultivation on silkworm (*Bombyx mori*) growth, cocoon quality, and survival under laboratory conditions**

Treatment Type	Larval Weight (g)	Cocoon Weight (g)	Shell Ratio (%)	Filament Length (m)	Survival Rate (%)	Reference
Control (non-treated leaves)	3.6 ± 0.1	1.35 ± 0.04	18.2 ± 0.6	850 ± 15	85.4 ± 1.2	Kavitha et al. (2014)
ZnSO <sub>4</sub> Foliar Spray (0.5%)	4.2 ± 0.2	1.52 ± 0.05	20.6 ± 0.8	945 ± 20	91.3 ± 1.5	Naveen et al., 2019; Lakshmi & Ramesh, 2016
Vermicompost Enriched Leaves	4.0 ± 0.1	1.48 ± 0.03	20.1 ± 0.7	920 ± 18	89.5 ± 1.0	Kumari & Raj (2021)
PGPR Treated Leaves	4.3 ± 0.2	1.56 ± 0.05	21.0 ± 0.5	980 ± 22	92.1 ± 1.3	Rani et al. (2018)
Fermented Mulberry Leaf Extract	4.1 ± 0.1	1.50 ± 0.04	20.4 ± 0.6	930 ± 17	90.6 ± 1.1	Tilahun (2020)

**Table 2. Summary of nutrient-based and microbial biofortification techniques in mulberry and their effects on *Bombyx mori* growth and silk yield**

Nutrient / Fortification	Mode of Action	Observed Impact on Silkworm	Reference
Zinc (Zn) biofortification	Enhances silk gland growth and enzyme activity	Increased cocoon weight and shell ratio	(Sarkar et al., 2017)
Fermented leaf feed (microbial)	Improves digestibility and nutrient availability	Better larval weight, survival rate	(Kavitha et al., 2014)
PGPR and Mycorrhiza inoculation	Enhances root nutrient uptake in mulberry	Enriched leaves improve cocoon traits	(Rani et al., 2018)
Foliar micronutrient spray	Boosts leaf protein and sugar content	Improved silk filament length	(Marin et al., 2023)
High-nitrogen mulberry varieties	Increases crude protein in leaves	Accelerated larval growth	Dandin et al. (2003)
Iron (Fe) supplementation	Improves leaf quality and hemolymph composition	Enhanced metabolic activity in larvae	(Lokanath & Shivashankar, 1986)
Balanced macronutrient fertilization	Increases carbohydrate and moisture in leaves	Boosts silk gland development	(Lakshmi & Ramesh, 2016; Kumar et al., 2018)

## 5. SUSTAINABILITY AND FUTURE PROSPECTS OF MULBERRY BIOFORTIFICATION

Incorporating biofortification practices in mulberry cultivation holds significant promise for sustainable sericulture. As global agriculture moves toward eco-friendly and resource-efficient systems, the sericulture sector must align its practices to reduce dependency on chemical inputs while maintaining high productivity. Biofortification especially through biological and agronomic means offers a viable solution to this challenge by enhancing leaf nutritional quality without compromising environmental health.

### 5.1 Environmental Sustainability

Biofortification through microbial inoculants, organic compost, and integrated nutrient management reduces the overuse of synthetic fertilizers and pesticides. These practices contribute to improved soil fertility, enhanced microbial activity, and reduced greenhouse gas emissions (Rani et al., 2018). The application of plant growth-promoting rhizobacteria (PGPR), such as *Azospirillum*, *Bacillus*, and *Pseudomonas* spp., enhances nitrogen fixation, phosphate solubilization, and nutrient uptake efficiency, which collectively reduce nitrate leaching and chemical residue build-up (Rani et al., 2018). Additionally, biofortification aids in restoring degraded soils and promotes carbon sequestration through increased root biomass (White et al., 2013).

### 5.2 Economic Viability for Farmers

Smallholder farmers, who constitute the majority of the sericulture community, can benefit economically from biofortification techniques. Improved cocoon yield, better-quality silk, and reduced disease incidence translate into higher returns on investment. For example, fortified mulberry leaves have been shown to increase silk filament length and cocoon shell weight, directly improving market value (Kavitha et al., 2014). Since many biofortification strategies utilize locally available resources such as compost, farmyard manure (FYM), or indigenous microbial strains, adoption is both cost-effective and scalable (Manjunatha et al., 2019; Reddy et al., 2020). Moreover, the reduction in chemical inputs leads to additional cost savings and improves long-term farm sustainability.

## 5.3 Integration with Climate-Resilient Practices

Climate variability and soil degradation are emerging challenges in sericulture regions. Biofortification helps develop more resilient cropping systems by improving plant vigor and resistance to abiotic stressors such as drought, salinity, and heat (Das & Sarkar, 2024). Fortified mulberry leaves support sustained leaf biomass production under marginal environmental conditions, ensuring consistent silkworm feeding and cocoon development. PGPRs and mycorrhizal fungi also contribute to better water retention in soil and hormone regulation (such as increased indole-acetic acid and cytokinin levels), enhancing drought tolerance in mulberry plants (Nithya et al., 2018).

### 5.4 Future Research Directions

Despite promising results, biofortification in mulberry is still in its early stages compared to food crops. Future research should focus on identifying nutrient-dense mulberry genotypes using omics-based tools, including transcriptomics and metabolomics, to study the expression of nutrient transporters and storage proteins (Tilahun, 2020). Moreover, field-scale studies under different agro-climatic conditions are essential to validate lab and pot-level findings. Research into nanonutrient delivery systems, such as nano-zinc and nano-iron, also holds potential for targeted and efficient leaf enrichment (Nithya, 2018; Ding et al., 2023). Interdisciplinary collaboration between soil scientists, entomologists, and breeders will be critical to advance this field.

### 5.5 Policy and Extension Implications

For the widespread adoption of biofortification, policy-level support is crucial. Training programs, subsidies for bio-inputs, and demonstration trials can accelerate the uptake of these techniques among sericulture farmers. Inclusion of biofortification modules in sericulture extension manuals and farmer field schools will bridge the knowledge gap and promote sustainable practices (Dandin et al., 2003). Government-led certification programs recognizing biofortified leaf production may also incentivize adoption. Strengthening public-private partnerships and research-extension linkages will be instrumental in transforming mulberry biofortification from a niche innovation to a mainstream practice.

## **6. EMERGING FRONTIERS: NANO-BIOFORTIFICATION AND MOLECULAR INSIGHTS IN MULBERRY NUTRITION**

Recent advances in agricultural biotechnology have paved the way for more efficient and sustainable approaches to enhance the nutritional profile of mulberry leaves. Among these, nano-biofortification and molecular-based nutritional understanding represent significant innovations in the field of sericulture. These emerging tools not only improve the nutritional quality of mulberry foliage but also provide insights into the biological responses of silkworms to enriched diets.

### **6.1 Nano-Biofortification for Targeted Nutrient Delivery**

Nano-biofortification refers to applying essential micronutrients (e.g., Zn, Fe, B) as nanoparticles to enhance their absorption, translocation, and utilization within plant tissues. In mulberry (*Morus alba*), such nanoscale micronutrients have been shown to increase leaf nutrient concentration, photosynthetic activity, and overall vigor (Nithya, 2018). This improvement in leaf physiology directly translates into better cocoon characteristics in *Bombyx mori*.

For example, foliar spraying of nano-zinc oxide at a concentration of 25 ppm has been shown to improve leaf protein content and larval growth rate, ultimately enhancing cocoon weight and shell percentage. The slow-release behavior of nano-fertilizers ensures consistent nutrient availability, thereby reducing nutrient leaching, minimizing environmental degradation, and improving input use efficiency (Nithya et al., 2018).

### **6.2 Molecular Responses of Silkworms to Enriched Mulberry Diets**

The ingestion of nutrient-rich or biofortified mulberry leaves induces physiological and molecular changes in silkworms. Notably, enriched diets lead to upregulation of genes involved in silk gland development, fibroin and sericin synthesis, and metabolic enzyme regulation (Nithya et al., 2018). Enhanced nutrient availability, particularly proteins and micronutrients like Zn and Fe, influences hemolymph composition, antioxidant defense mechanisms, and stress tolerance in larvae.

Gene expression studies have indicated that biofortified diets increase the transcription levels of fibroin heavy chain (*Fib-H*) and sericin 1 (*Ser1*) genes, which are crucial for silk filament formation. In addition, improved enzyme activities such as superoxide dismutase (SOD) and catalase (CAT) contribute to better oxidative stress management in larvae fed with fortified diets (Marin et al., 2022). These molecular responses establish a strong link between mulberry leaf enrichment and enhanced silk biosynthesis.

### **6.3 Integrated Molecular–Agronomic Approaches for Future Sericulture**

Combining molecular biology with agronomic management offers a holistic strategy for enhancing cocoon yield. The integration of transcriptomic, proteomic, and metabolomic data can aid in the identification of nutrient-efficient mulberry varieties and tailor-made feeding regimens. By aligning plant and insect nutrition, it becomes feasible to develop site-specific management practices that improve both productivity and sustainability.

Future studies should explore the role of nutrient transporters, stress-responsive genes, and silk-specific regulatory proteins in response to fortified diets. Such insights can guide the development of targeted biofortification strategies, enhancing silk output while maintaining ecological balance (Tilahun, 2020).

## **7. CLIMATE RESILIENT MULBERRY CULTIVATION FOR SUSTAINABLE SILK PRODUCTION**

Climate change poses serious challenges for sericulture, especially in sustaining the quality and quantity of mulberry leaf production. Rising temperatures, erratic rainfall, prolonged droughts, and progressive soil degradation weaken mulberry physiology, leading to reduced biomass and diminished nutrient content in the leaves. As a result, silkworm health, growth, and cocoon productivity also suffer. In India, an estimated 29–33% of total agricultural land nearly 96 million hectares is degraded, with over 37 million hectares of this being rainfed farmland often found in sericulture belts. Such conditions heighten the vulnerability of mulberry cultivation, making the adoption of climate-resilient practices a priority in both research and field applications.

### 7.1 Development of Drought- and Heat-Tolerant Mulberry Genotypes

Breeding efforts have resulted in the development of mulberry varieties such as RC2, S1635, and VI-1 that exhibit higher tolerance to drought and temperature extremes. These varieties possess traits such as deeper root systems, high leaf moisture retention, better osmolyte accumulation (e.g., proline), and efficient stomatal regulation, which enable them to maintain leaf quality under stress conditions (Rahmathulla, 2012).

Field evaluations have shown that these genotypes maintain relatively stable levels of crude protein and soluble sugars during heat waves, making them suitable for rearing silkworms even in off-seasons. Such climate-adapted varieties are key to sustaining year-round cocoon production.

### 7.2 Microbial Interventions for Stress Mitigation

The application of plant growth-promoting rhizobacteria (PGPR) and arbuscular mycorrhizal fungi (AMF) has been shown to enhance stress tolerance in mulberry plants. These beneficial microbes improve nutrient acquisition, root architecture, and production of stress-alleviating phytohormones such as indole-3-acetic acid (IAA) and abscisic acid (ABA) (Abdalla et al., 2023). Additionally, they increase antioxidant enzyme activities in mulberry, mitigating the effects of drought and salinity.

Silkworms fed on leaves from PGPR-treated mulberry plants have shown better weight gain, reduced larval mortality, and improved cocoon traits, even under suboptimal environmental conditions (Rani et al., 2018).

### 7.3 Climate-Smart Agronomic Practices

The implementation of climate-smart cultivation practices such as organic mulching, drip irrigation, and intercropping with legumes plays a critical role in enhancing soil moisture conservation and reducing evapotranspiration losses. The adoption of organic mulching significantly conserves soil moisture, enhances microbial health, and boosts mulberry leaf yield and quality (Verma & Pradhan, 2024). Intercropping with legumes such as soybean and greengram in paired-row systems further enhances mulberry leaf yield, improves nutrient

status and boosts cocoon performance in *Bombyx mori* (Banu Priya et al., 2022). Drip irrigation systems, when paired with fertigation techniques, ensure uniform nutrient distribution while minimizing water wastage. These water-saving methods are especially important in drought-prone sericulture regions such as Kolar district in Karnataka, Dharmapuri district in Tamil Nadu, and parts of Anantapur district in Andhra Pradesh, where recurrent water scarcity severely limits mulberry cultivation and silkworm rearing during summer months.

### 7.4 Long-Term Prospects

Integrating stress-resilient mulberry varieties, microbial inoculants, and adaptive agronomic techniques holds significant promise for climate-resilient sericulture. These approaches will not only help stabilize mulberry productivity under adverse conditions but also improve the sustainability and profitability of silk farming. Future research should aim to develop decision-support tools and weather-adaptive nutrient schedules tailored for different agro-climatic zones.

## 8. CONCLUSION

The quality and nutritional composition of mulberry leaves are critical factors that directly influence the physiological performance, growth dynamics, and silk-producing capabilities of the silkworm, *Bombyx mori*. As the exclusive food source for silkworms, the biochemical makeup of mulberry foliage particularly its protein, carbohydrate, moisture, and micronutrient content play a decisive role in determining larval health, metabolic efficiency, and ultimately, cocoon yield and silk quality. Given the intimate relationship between mulberry nutrition and silkworm development, enhancing the nutritive value of these leaves presents a strategic and sustainable opportunity to improve sericultural outcomes.

Recent advancements have highlighted the potential of various interventions including agronomic practices, microbial applications, and post-harvest treatments to significantly boost the nutritional quality of mulberry leaves. Agronomic improvements, such as optimized fertilization, soil management, and cultivar selection, have been shown to enhance leaf biomass and nutrient density. Microbial enrichment, particularly through the application of plant growth-promoting rhizobacteria (PGPR) and

beneficial fungi, improves nutrient uptake, plant vigor, and resilience to environmental stressors. Post-harvest fortification techniques, such as nutrient dipping or foliar application of micronutrients, offer further avenues for improving leaf quality just before silkworm feeding.

These strategies have demonstrated tangible benefits in sericulture. Improved leaf quality contributes to faster larval development, higher cocoon weights, better shell ratios (the proportion of cocoon shell to total cocoon weight), longer silk filaments, and increased larval survival. These improvements not only enhance raw silk yield and quality but also contribute to reduced production cycles and more consistent cocoon output.

As global demand for natural and high-quality silk continues to rise—driven by the textile, medical, and cosmetic industries—modernizing sericulture through optimized mulberry nutrition is becoming increasingly important. A nutrition-centric approach to mulberry cultivation aligns with global goals for sustainable agriculture, as it reduces reliance on chemical inputs, improves land-use efficiency, and supports climate-resilient practices.

Looking ahead, future research should focus on developing region-specific biofortification protocols that consider local soil conditions, climate variability, and farmer practices. The identification and integration of effective PGPR strains that enhance nutrient uptake and plant growth under diverse field conditions is another key research priority. Furthermore, the development and dissemination of eco-friendly, affordable field technologies—such as compost-based biofertilizers, nano nutrient solutions, or microbial consortia—can make nutritional interventions accessible to smallholder farmers.

A systems-level understanding of the nutritional interactions between mulberry plants and silkworms is essential for unlocking the full potential of this sustainable approach. By addressing both plant and insect nutritional needs, such integrated strategies can drive improvements in silk production, enhance farm incomes, and reinforce the socio-economic foundations of sericulture, particularly in developing countries such as India, China, Thailand, Vietnam, and Bangladesh, where it serves as a major livelihood source.

## DISCLAIMER (ARTIFICIAL INTELLIGENCE)

Author(s) hereby declares that NO generative AI technologies such as Large Language Models (ChatGPT, manuscript).

## COMPETING INTERESTS

Authors have declared that no competing interests exist.

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