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Integrating Multi-Omics, Bioinformatics and Genome Editing for Sustainable 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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Review Article

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ABSTRACT

Sericulture, centred on the mulberry silkworm *Bombyx mori*, is a millennia-old agro-based industry now facing challenges from climate variability, pathogen outbreaks, and the limitations of conventional breeding. Recent advances in molecular biology have revolutionised silkworm research, offering precision tools for genetic improvement and sustainable silk production. This review synthesises progress across three interconnected domains: multi-omics, bioinformatics, and

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genome editing. Genomic, transcriptomic, proteomic, and metabolomic studies have provided system-level insights into silk gland biology, immune responses, stress tolerance, and dietary adaptation. Specialised bioinformatics platforms such as SilkDB, KAIKObase, and MorusDB enable integration of diverse datasets, facilitating gene discovery, trait prediction, and marker-assisted breeding. Concurrently, genome editing technologies—particularly CRISPR/Cas9—have transformed functional genomics and trait engineering, enabling targeted improvements in silk yield, fibre quality, disease resistance, and climate resilience. Emerging tools such as base and prime editors further expand the potential for precise genetic manipulation. By linking omics-driven target identification, bioinformatics-guided prioritisation, and genome editing–based validation, sericulture is entering a new era of precision breeding. This integrative framework accelerates the development of resilient, high-yielding silkworm strains but also broadens applications in biomaterials, pharmaceuticals, and sustainable agro-industrial systems. Together, these innovations position sericulture as a modern bioindustry capable of meeting global demands while preserving rural livelihoods and ecological balance.

Keywords: Bombyx mori; sericulture; multi-omics; genomics; transcriptomics; proteomics; precision breeding; sustainable sericulture.

1. INTRODUCTION

Silkworm rearing has been a cornerstone of sericulture, contributing significantly to the global textile industry. Mulberry silkworm (*Bombyx mori* L.) is an important economic insect in the commercial production of silk, a natural fiber prized for its luster, strength and breathability. India, being a tropical country. Improving silk yield and quality in silkworms holds tremendous promise for various industries, including textiles, medicine, and biotechnology. The traditional method of silk production involves labor-intensive processes and relies heavily on the natural behavior and traits of silkworms. However, genetic improvements offer the potential to revolutionize silk production by enhancing key attributes of the silkworm (Baci et al., 2021; Pavithra et al., 2024). Sericulture, the cultivation of silkworms for silk production, has been practised for more than 5,000 years and remains a vital agro-based industry with immense cultural, economic, and scientific significance. Among domesticated insects, the mulberry silkworm (*Bombyx mori*) holds a unique position as both a model organism for Lepidoptera genetics and the principal source of commercial silk worldwide. Beyond textiles, sericulture supports rural livelihoods, drives regional economies, and contributes to ecological sustainability by promoting agroforestry-based systems (Punyavathi, 2013; International Silkworm Genome Consortium, 2008; Xia et al., 2009; Nagaraju & Goldsmith, 2002). The historical success of silk cultivation demonstrates humanity's ability to harness the biology of insects, yet modern challenges necessitate transformative approaches for the future.

Traditional breeding techniques, such as hybridisation, mutation breeding, and selection, have successfully yielded productive silkworm strains and high-yielding mulberry varieties (Anusha & Vijayan, 2023). Although these strategies have been successful, they are time-consuming, less exact, and strongly reliant on phenotypic selection, rendering them ineffective for meeting the changing needs of modern sericulture (Moulidharshan et al., 2025). Traditional silkworm breeding strategies have focused on selective crossing of high-yield strains to improve cocoon traits such as filament length, tensile strength, and silk ratio. While effective to some degree, these classical methods are inherently constrained by several factors: long generation times, restricted genetic diversity within domesticated strains, and the unpredictability of polygenic traits that are influenced by environmental variables. As a result, trait improvement through conventional breeding is both time-intensive and of limited efficiency, leaving the industry vulnerable to biotic stresses (such as viral and bacterial diseases) and abiotic factors (including fluctuating temperature and humidity) (Ruiz & Almanza, 2018). Furthermore, the monoculture-like dependence on mulberry leaves as the sole feed source for *B. mori* adds another layer of risk, especially under changing climatic conditions that threaten mulberry cultivation.

In response to these limitations, the last two decades have witnessed an unprecedented acceleration in silkworm research driven by advances in genomics and multi-omics sciences. The publication of the *B. mori* draft genome marked a watershed moment, enabling genome-

wide investigations into silk gland physiology, immune responses, and metabolic adaptations (International Silkworm Genome Consortium, 2008). Follow-up studies, such as the resequencing of 40 diverse silkworm genomes, revealed insights into domestication events, natural variation, and key genes associated with traits of agronomic importance (Xia et al., 2009). Building on these resources, transcriptomic analyses have shed light on tissue-specific gene expression, particularly in the silk gland, identifying regulators of fibroin and sericin synthesis (Wang et al., 2016). Similarly, proteomic and metabolomic studies have begun to map the dynamic molecular landscape underlying silk production, cocoon quality, and dietary responses. Together, these datasets form the foundation of a multi-omics framework that connects genotype to phenotype at a systems level.

Crucial to the interpretation of this vast information are the bioinformatics resources specifically tailored for silkworm biology. Databases such as KAIKObase, SilkDB, and MorusDB integrate genomic, transcriptomic, and proteomic data, providing researchers with tools for functional annotation, evolutionary comparisons, and marker-assisted breeding strategies (Shimomura et al., 2009). These platforms also serve as repositories of curated datasets that enable cross-strain comparisons and gene discovery for traits such as artificial diet adaptability and pathogen resistance. By consolidating heterogeneous omics data, bioinformatics transforms raw sequence information into actionable knowledge that can guide breeding and genetic engineering.

The most transformative breakthrough, however, has been the advent of genome editing technologies. Early programmable nucleases such as zinc-finger nucleases (ZFNs) and transcription activator-like effector nucleases (TALENs) established proof-of-concept for targeted mutagenesis in *B. mori*, but their use was limited by technical complexity and cost. The introduction of the CRISPR/Cas9 system revolutionised insect functional genomics, providing a simple, precise, and efficient tool for gene disruption, modification, and functional validation (Pickar-Oliver & Gersbach, 2019; Cong et al., 2013). In silkworms, CRISPR-based editing has already been applied to silk gland–specific genes to enhance cocoon characteristics, investigate immunity against pathogens, and explore metabolic pathways crucial for growth and development. Beyond basic science, such approaches offer the prospect of creating silkworm strains with improved disease resistance, higher silk yield, and adaptability to artificial diets—traits that are central to ensuring the sustainability of sericulture.

At the same time, challenges remain. The silkworm's complex life cycle, reliance on environmental cues, and susceptibility to off-target editing present hurdles to the broad deployment of genome engineering. Moreover, socio-economic considerations—such as acceptance of genetically modified silkworms, intellectual property issues, and the cost of advanced technologies—must be addressed for successful translation into sericultural practice. Nevertheless, the convergence of multi-omics, bioinformatics, and genome editing represents a powerful integrated framework that can overcome the limitations of conventional breeding and usher in a new era of precision sericulture.

This review aims to provide a comprehensive synthesis of recent progress in this field. Specifically, we highlight (i) the role of genomics, transcriptomics, proteomics, and metabolomics in decoding silkworm biology, (ii) the emergence and application of bioinformatics tools that facilitate large-scale data integration, (iii) key insights from genetic studies on artificial diet utilization, (iv) advances in genome editing technologies including CRISPR/Cas9 and beyond, and (v) the potential of combining these innovations for sustainable sericulture. By integrating knowledge across these domains, we outline a roadmap for the rational design of resilient, high-yielding silkworm strains capable of supporting both traditional sericulture and modern biotechnological applications.

2. MULTI-OMICS APPROACHES IN SILKWORM RESEARCH

The evolution of omics technologies has transformed sericulture research by providing system-level insights into silkworm biology. Multi-omics strategies, which include genomics, transcriptomics, proteomics, and metabolomics, allow the dissection of complex biological processes that underlie silk production, stress tolerance, and artificial diet adaptation. Unlike traditional approaches, which are limited to single-gene or phenotype-level analysis, multi-omics integrates diverse datasets to reveal how

molecular changes propagate across cellular networks, shaping economically important traits.

2.1 Genomics: Building the Foundation of Silkworm Biology

The completion of high-quality *Bombyx mori* genome assemblies enabled the identification of gene families associated with silk protein synthesis, immunity, and development (Xia et al., 2014). Population genomic studies further highlighted selective sweeps in genes linked to domestication, including those affecting cocoon color, size, and disease resistance (Goldsmith et al., 2005).

Comparative genomics between domesticated and wild silkworms has provided insight into evolutionary divergence and adaptive traits. For example, resequencing studies uncovered polymorphisms in detoxification and stress-responsive genes that help certain strains thrive on non-mulberry diets (Xiang et al., 2018). Advances in long-read sequencing technologies have also facilitated the discovery of structural variants and repeat expansions, improving annotation of silk gland–specific genes (Kawamoto et al., 2019).

2.2 Transcriptomics: Functional Gene Expression Profiling

Transcriptome sequencing (RNA-seq) has emerged as a powerful tool for identifying stage- and tissue-specific expression in silkworms. Genome-wide expression profiling revealed that silk gland subregions (anterior, middle, and posterior) express distinct sets of genes, many encoding transcription factors and secretory pathway components critical for fibroin and sericin synthesis (Xia et al., 2007).

Transcriptomics has also been applied to explore how environmental conditions and diet shape gene expression. For instance, larvae reared on artificial diets show differential expression of genes encoding digestive enzymes, heat shock proteins, and detoxification factors compared with mulberry-fed controls (Dong et al., 2017). Moreover, infection-triggered transcriptome shifts highlight the silkworm's immune system plasticity, offering candidate genes for breeding disease-resistant strains (Cheng et al., 2017).

2.3 Proteomics: From Silk Gland to Stress Biology

Proteomic analyses complement transcriptomic studies by quantifying protein abundance and post-translational modifications. High-resolution proteomics has identified hundreds of silk gland proteins, including fibroins, sericins, and structural glycoproteins (Zhang et al., 2006). Proteomic comparisons across strains with varying silk yield show differential abundance of folding chaperones, redox enzymes, and transport proteins that contribute to cocoon quality.

Proteomics has also been central in studying stress responses. Under conditions of temperature stress or altered diets, significant upregulation of heat shock proteins and metabolic enzymes has been reported (Li et al., 2012). These findings support the hypothesis that silk gland productivity is tightly linked to proteome-level resilience, suggesting potential molecular markers for selecting robust strains.

2.4 Metabolomics: Linking Diet, Environment, and Silk Production

Metabolomics provides the biochemical layer of omics, directly connecting nutrition and metabolism with silk yield. Studies have shown that mulberry-fed larvae accumulate high levels of amino acids such as glycine, alanine, and serine, all critical for fibroin biosynthesis (Dong et al., 2017). By contrast, larvae fed artificial diets display altered energy metabolism, elevated detoxification intermediates, and modified fatty acid profiles, reflecting adaptation challenges.

Environmental stresses also leave detectable metabolic signatures. For instance, pesticide exposure or elevated rearing temperatures disrupt carbohydrate and lipid metabolism, leading to reduced cocoon shell weight (Li et al., 2012). Such metabolic biomarkers are increasingly recognised as indicators of strain resilience and dietary efficiency.

2.5 Toward Integrated Omics for Sericulture

The power of multi-omics lies not in isolated datasets but in their integration. Systems biology frameworks that combine genomics, transcriptomics, proteomics, and metabolomics are beginning to reconstruct regulatory networks of silk gland function. Instead of treating each dataset separately, a consolidated view highlights how molecular changes across different layers converge to shape silk yield, stress resilience, and disease resistance. Representative applications of each omics approach in silkworm research are summarised in Table 1.

Table 1. Applications of Multi-Omics Approaches in Silkworm (*Bombyx mori*) Research

Omics Approach	Focus Area	Key Findings / Applications	Example References
Genomics	Genome assembly, domestication genes, structural variants	Identification of loci controlling cocoon color, size, and disease resistance; discovery of adaptive genes for artificial diets	(Goldsmith et al., 2005; Xiang et al., 2018; Kawamoto et al., 2019)
Transcriptomics	Tissue- and stage-specific gene expression	Revealed distinct gene expression in anterior, middle, and posterior silk gland regions; insights into diet- and stress-responsive expression; immune pathway regulators identified	(Xia et al., 2007; Dong et al., 2017; Cheng et al., 2017)
Proteomics	Protein abundance, post-translational modifications, stress markers	Catalogued fibroins, sericins, and chaperones; linked protein abundance to cocoon quality; identified heat shock proteins and redox enzymes under stress	(Zhang et al., 2006; Li et al., 2012)
Metabolomics	Nutrition, energy metabolism, environmental stress	Demonstrated amino acid profiles essential for fibroin synthesis; metabolic shifts under artificial diets, pesticides, and heat stress linked to reduced cocoon weight	(Dong et al., 2017; Li et al., 2012)
Integrated Omics	Systems biology of silk production	Multi-layer integration of transcriptome, proteome, and metabolome revealed regulatory networks for fibroin secretion and candidate genes for CRISPR editing	(Xia et al., 2007; Zhang et al., 2006; Li et al., 2012)

This holistic approach enables precision breeding and genome editing: omics identifies candidate targets, bioinformatics tools refine predictions, and CRISPR-based editing validates their functions. Together, multi-omics integration provides a roadmap for rational strain improvement, ultimately enhancing silk yield, disease resistance, and environmental adaptability.

3. BIOINFORMATICS TOOLS IN SERICULTURE

The large-scale generation of genomic and transcriptomic data in *Bombyx mori* and related silk moths has driven the development of bioinformatics platforms that enable systematic analysis, integration, and application of omics data. Unlike traditional breeding tools, bioinformatics resources allow researchers to mine, compare, and visualize complex datasets, thereby facilitating molecular breeding, functional genomics, and disease resistance research in sericulture.

3.1 Databases for Genomic and Functional Data

The creation of SilkDB marked the first major milestone in silkworm bioinformatics. Originally developed as a genomic repository, SilkDB evolved into an integrative platform incorporating expression profiles, QTL markers, and comparative tools for functional annotation of genes (Duan et al., 2010). Complementary to SilkDB, KAIKObase was launched with expanded gene models, microsatellite markers, and data-mining functions, allowing identification of loci involved in silk quality and domestication-related traits (Wang et al., 2005).

Beyond *B. mori*, bioinformatics has extended to related resources. MorusDB, which catalogues mulberry genomes and transcriptomes, links host plant genetics to silkworm physiology (He et al., 2013). Similarly, WildSilkbase provides transcriptomic datasets of wild silk-producing moths such as tasar (*Antheraeamylitta*), eri (*Samiaricini*), and muga (*Antheraeaassamensis*), supporting comparative analyses across diverse silk species (Arunkumar et al., 2006). Together, these platforms expand the bioinformatics landscape of sericulture, enabling cross-species insights into silk protein evolution and stress adaptation.

3.2 Bioinformatics for Silk Protein Annotation

Silk proteins such as fibroins and sericins are the primary focus of many molecular studies due to their direct economic importance. Bioinformatics pipelines are used to predict signal peptides, post-translational modifications, and protein–protein interactions (Li et al., 2020). Such analyses provide insights into the secretion pathways that maintain silk gland productivity. Comparative annotation using multi-species databases has further clarified the conserved motifs in silk fibroin heavy and light chains, helping identify candidate sites for genome editing to improve fiber strength and elasticity.

3.3 Pathogen Databases and Immune Response Mining

Diseases remain a major threat to sericulture productivity. Bioinformatics platforms such as SilkPathDB integrate genomic data of *Bombyx mori* pathogens, including microsporidia, viruses, and fungi, with host gene expression datasets (Li et al., 2017). These resources allow researchers to track co-evolution between silkworms and pathogens, predict virulence factors, and identify host defense genes. For example, analysis of host–virus interactions has pinpointed antiviral RNA interference pathways, providing molecular targets for genetic improvement (Cheng et al., 2016).

3.4 Computational Tools for Genome Editing Applications

As CRISPR/Cas9 and related editing technologies expand in silkworm research, computational tools are increasingly employed to predict off-target sites, design sgRNAs, and evaluate target gene networks. Integrated platforms provide algorithms for on-target scoring and off-target minimization, enabling more efficient editing strategies. These bioinformatics frameworks accelerate functional validation of candidate genes identified from multi-omics datasets, closing the loop between data discovery and experimental application.

3.5 Integration, AI, and the Future of Sericulture Bioinformatics

The future of sericulture bioinformatics lies in integration and artificial intelligence (AI). Linking SilkDB, MorusDB, WildSilkbase, and SilkPathDB into interoperable platforms would allow true systems biology approaches, reconstructing gene–protein–metabolite interaction networks that govern silk production and stress responses. Moreover, AI-driven predictive models can mine multi-omics datasets to forecast breeding outcomes, identify novel silk protein variants, and prioritize targets for CRISPR-mediated editing. Cloud-based bioinformatics platforms could democratize access to these tools, benefiting sericulture industries in resource-limited regions.

4. ADVANCES IN GENOME EDITING TECHNOLOGIES IN SERICULTURE

The application of genome editing technologies in *Bombyx mori* has transformed functional genomics and opened unprecedented opportunities for molecular breeding in sericulture. Unlike traditional mutagenesis and selection methods, which are slow and often imprecise, genome editing tools enable targeted manipulation of specific loci, allowing researchers to probe gene function and directly engineer desirable traits such as higher silk yield, improved fiber quality, and disease resistance.

4.1 Early Genome Editing Tools: Zinc-Finger Nucleases (ZFNs) and TALENs

The first attempts at targeted mutagenesis in *B. mori* employed zinc-finger nucleases (ZFNs), which function by fusing engineered zinc-finger proteins to the FokI nuclease domain (Takasu et al., 2010). These programmable nucleases generated double-strand breaks (DSBs) at specific genomic loci, leading to indels via the non-homologous end joining (NHEJ) pathway. However, their application in silkworm was limited by difficulties in engineering zinc-finger arrays, off-target cleavage, and relatively low efficiency.

Subsequently, transcription activator-like effector nucleases (TALENs) improved precision and versatility. TALENs rely on customizable DNA-binding domains derived from *Xanthomonas* effectors, offering easier design compared to ZFNs (Ma et al., 2012). In *B. mori*, TALENs enabled knockouts of pigmentation and developmental genes, serving as proof-of-concept for precise mutagenesis. Yet, despite their promise, TALENs were hampered by labor-intensive cloning, variable activity, and low scalability for large-scale functional genomics.

4.2 CRISPR/Cas9 Emergence and Adoption in *B. mori*

The introduction of the CRISPR/Cas9 system revolutionized genome editing by offering a simple, programmable, and efficient platform. In *B. mori*, CRISPR/Cas9 was first demonstrated through microinjection of Cas9 mRNA and single-guide RNAs (sgRNAs) into early embryos, yielding high-efficiency germline mutations (Wang et al., 2013). Compared to ZFNs and TALENs, CRISPR required minimal design effort, as the guide sequence is easily customized to target almost any genomic region adjacent to a protospacer adjacent motif (PAM).

CRISPR enabled multiplexed editing, allowing simultaneous knockout of multiple genes involved in silk production, pigmentation, and development (Liu et al., 2014). It has since become the standard tool for silkworm functional genomics, accelerating trait dissection and providing new avenues for molecular breeding.

4.3 Refinements: Transgenic Cas9 Lines and Tissue-Specific Editing

To improve efficiency and reduce mosaicism, researchers developed transgenic Cas9-expressing silkworm lines, enabling stable germline and tissue-specific editing. For example, posterior silk gland (PSG)-Cas9 and middle silk gland (MSG)-Cas9 strains express Cas9 under tissue-specific promoters, which can be crossed with sgRNA-expressing lines to achieve targeted knockouts within silk gland subregions (Liu et al., 2017).

This approach allows functional dissection of gland-specific genes, minimises off-target editing in non-silk tissues, and produces clear phenotypes in cocoon yield and fibre composition. Moreover, inducible Cas9 systems responsive to temperature or drug treatments provide temporal control, adding further flexibility to silkworm genetic engineering.

4.4 Beyond Knockouts: HDR, Base Editors, and Prime Editing

While NHEJ-mediated knockouts dominate silkworm CRISPR applications, increasing attention is directed toward homology-directed repair (HDR) for precise gene insertions or replacements. Although HDR efficiency remains low in silkworm embryos, optimised donor template delivery, synchronised embryo injections, and inhibition of competing repair pathways have improved success rates (Long et al., 2016). HDR has been applied to introduce reporter constructs, enabling visualisation of gene expression patterns in silk glands.

More recently, base editors and prime editors have emerged as next-generation CRISPR derivatives. Base editors combine a catalytically impaired Cas9 with deaminases, enabling targeted single-nucleotide substitutions without DSBs (Komor et al., 2016). Prime editing expands versatility by using a Cas9 nickase fused to reverse transcriptase, capable of introducing small insertions, deletions, or precise substitutions guided by prime editing gRNAs. While their application in *B. mori* is still in early stages, these tools promise to enhance trait engineering precision without relying heavily on HDR.

4.5 Applications in Functional Genomics and Trait Improvement

CRISPR-based editing has already yielded significant biological insights and practical outcomes in sericulture. For instance, knockout of *BmEcKL1* demonstrated its role in silk gland growth and silk protein synthesis, with mutants displaying reduced cocoon shell weight (Li et al., 2024). Similarly, targeted disruption of chitin synthase genes clarified their contribution to larval cuticle formation and molting, while editing of pigmentation loci created novel cocoon color morphs.

In terms of breeding, genome editing has facilitated the development of silkworm strains with enhanced cocoon size, altered silk protein ratios, and improved pathogen resistance. Such targeted improvements hold potential to shorten breeding cycles dramatically compared with traditional crossbreeding.

Table 2. Genome Editing Tools and Applications in *Bombyx mori*

Tool	Period of Use	Advantages	Limitations	Applications in *B. mori*	Example References
Zinc-Finger Nucleases (ZFNs)	~2010	First proof-of-concept for targeted mutagenesis	Complex design, low efficiency, off-target cleavage	Knockout of pigmentation and developmental genes	(Takasu et al., 2010)
TALENs	2012 onward	More precise, customizable than ZFNs	Labor-intensive cloning, variable activity	Gene knockouts for pigmentation, cuticle formation	(Ma et al., 2012)
CRISPR/Cas9	2013 onward	Simple, efficient, multiplex gene editing	Mosaicism, off-target effects	Trait dissection, silk yield enhancement, disease resistance	(Wang et al., 2013; Liu et al., 2014; Liu et al., 2017)
Base Editors	2016 onward	Precise single-nucleotide changes without double-strand breaks	Still experimental in silkworms	Point mutation corrections, fine-tuning silk protein genes	(Komor et al., 2016)
Prime Editors	2019 onward	Small insertions/deletions, versatile edits	Limited efficiency in insects	Potential for precise trait engineering	(Komor et al., 2016)

4.6 Future Prospects and Challenges

Despite rapid progress, challenges remain in applying genome editing to sericulture. Issues such as off-target effects, mosaicism, and low homology-directed repair (HDR) efficiency limit large-scale applications. Moreover, ethical concerns and regulatory frameworks surrounding genetically modified insects need to be addressed before commercialisation. Nevertheless, the evolution of genome editing technologies has already transformed *Bombyx mori* research, providing tools that range from early mutagenesis to next-generation precision editing. A comparative overview of these tools is presented in Table 2.

The progression from ZFNs and TALENs to CRISPR/Cas9 and its derivatives illustrates the increasing precision and versatility of genome editing in sericulture. As these tools mature, their integration with multi-omics and bioinformatics will accelerate the development of high-yielding, resilient silkworm strains tailored for sustainable silk production.

5. APPLICATIONS OF GENOME EDITING IN SERICULTURE

The rapid adoption of genome editing tools, particularly CRISPR/Cas9, has revolutionised functional genomics in *Bombyx mori*, enabling precise dissection of gene function and accelerating trait improvement in sericulture. Whereas conventional breeding required multiple generations to introduce or combine traits, targeted editing allows direct manipulation of loci underlying silk production, disease resistance, and developmental processes. Applications now extend beyond traditional sericulture improvement to novel areas such as biomaterial engineering, pharmaceutical production, and climate resilience.

5.1 Functional Genomics of Silk Production and Gland Biology

Silk yield and quality remain central objectives in sericulture. Genome editing has been applied to probe genes controlling silk gland growth and protein secretion. For example, knockout of the *BmEcKL1* gene using CRISPR disrupted posterior silk gland development, reducing gland size and silk protein accumulation (Sun et al.,

2024). Conversely, targeted enhancement of growth-regulating genes in transgenic posterior silk gland Cas9 lines produced larger glands, increased fibroin synthesis, and heavier cocoons (Zhou et al., 2025).

CRISPR has also revealed insights into sericin and fibroin synthesis pathways. Editing genes involved in sericin glycosylation modified silk viscosity and tensile strength, while fibroin promoter engineering has been used to modulate heavy chain–light chain expression ratios, improving fibre uniformity (Inoue et al., 2000; Zhou et al., 2001). These applications show how functional genomics is directly linked to practical improvements in silk fibre traits.

5.2 Engineering Disease Resistance

Pathogens such as *Bombyx mori* nucleopolyhedrovirus (BmNPV), microsporidia, and fungal invaders cause major losses in sericulture. Genome editing has facilitated the identification and manipulation of antiviral and immune pathway genes. For instance, CRISPR knockout of *BmNPC1*, a cholesterol transporter required for baculovirus entry, rendered silkworms resistant to BmNPV infection (Chen et al., 2017).

Similarly, editing of RNA interference (RNAi) pathway components has clarified their role in antiviral defense. Mutants lacking *Dicer* or *Argonaute* genes show heightened susceptibility to viral infection, confirming RNAi as a key antiviral mechanism (Sienski et al., 2012). Transgenic approaches coupling CRISPR editing with overexpression of antimicrobial peptides have further demonstrated improved survival under pathogen challenge (Dubey & Mostafavi, 2023). These advances pave the way for developing silkworm strains with stable resistance, reducing reliance on antibiotics or chemical disinfectants.

5.3 Modifying Developmental and Morphological Traits

Developmental processes in *B. mori* are also amenable to genome editing, providing both fundamental insights and breeding applications. Knockout of *chitin synthase* genes has clarified the molecular basis of molting and cuticle formation, whereas editing of hormone receptor loci has elucidated the regulation of metamorphosis (Daimon et al., 2014).

Importantly, these studies have translational relevance. Modifications in developmental pathways can alter growth rates, larval robustness, and rearing duration—traits directly influencing sericulture economics. Shorter larval stages, for instance, could reduce feeding costs, while stronger cuticles might enhance tolerance to crowding and environmental stress.

5.4 Cocoon Color and Novel Phenotypes

Cocoon coloration is a valuable trait with economic and aesthetic significance. Traditionally, color mutations arose through spontaneous variation or selection, but genome editing now allows customized cocoon colors. CRISPR-mediated knockout of *yellow* and *ebony* loci has produced distinct pigmentation phenotypes, ranging from pale cocoons to darker shades (Nie et al., 2021).

These traits can increase market diversity, particularly in niche silk industries that value natural coloration to reduce dyeing costs. Moreover, coloration genes serve as visible markers for validating editing efficiency in functional genomics experiments, making them practical tools in laboratory settings.

5.5 Silkworm as a Bioreactor

Beyond traditional sericulture, genome editing has expanded the role of *B. mori* as a bioreactor for recombinant protein production. By editing silk fibroin promoters and inserting exogenous coding sequences, silkworms have been engineered to produce human collagen, growth factors, and therapeutic proteins within their cocoons (Kuwana et al., 2003).

These engineered silks combine natural fibroin strength with bioactive properties, creating hybrid biomaterials for medical sutures, scaffolds, and drug delivery systems. CRISPR has streamlined this process, enabling precise insertion of transgenes into fibroin loci, ensuring stable expression and inheritance. This establishes silkworms as cost-effective alternatives to mammalian cell cultures for producing high-value biomaterials.

5.6 Climate and Environmental Resilience

As climate change intensifies, genome editing offers solutions for enhancing silkworm resilience. Genes regulating thermal tolerance, oxidative stress response, and detoxification

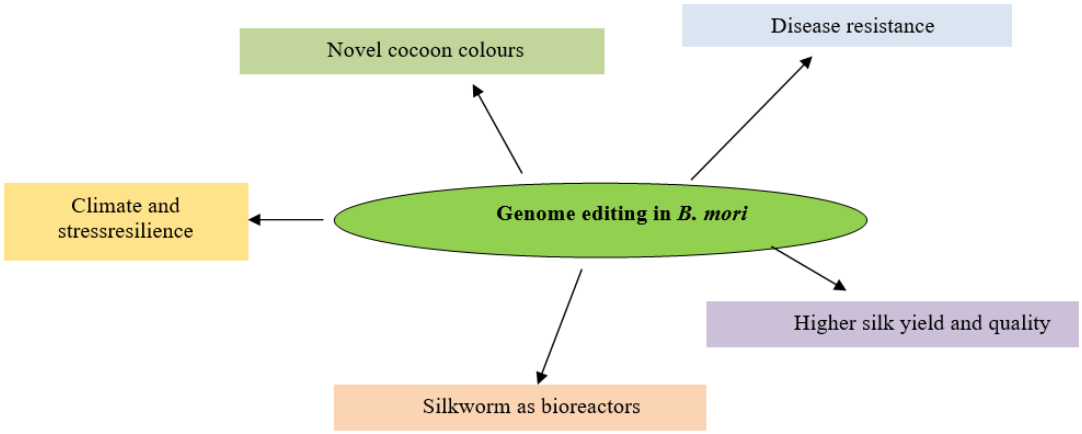

Fig. 1. Application of genome editing in Sericulture

have been identified through transcriptomics and validated with CRISPR knockouts or overexpression. For example, silkworms edited for heat shock protein (HSP) regulators displayed improved survival at elevated rearing temperatures (Li et al., 2019).

Similarly, editing of detoxification enzymes has been linked to enhanced tolerance of pesticide residues and alternative artificial diets. These traits will be crucial for sustaining sericulture in regions where environmental instability threatens productivity.

5.7 Toward Precision Breeding in Sericulture

Collectively, these applications demonstrate a decisive shift from conventional crossbreeding toward precision breeding in silkworms. Genome editing now enables the rational design of strains tailored for industrial needs—whether for enhanced silk yield and quality, stable disease resistance, novel cocoon colors, or tolerance to environmental stresses. Moreover, *B. mori* is increasingly positioned as a bioreactor for producing high-value biomaterials and pharmaceuticals. These diverse applications are summarized in Fig. 1, which highlights the expanding role of genome editing in shaping the future of sericulture.

6. INTEGRATION OF MULTI-OMICS, BIOINFORMATICS, AND GENOME EDITING FOR SUSTAINABLE SERICULTURE

The sustainability of sericulture in the 21st century depends on addressing critical challenges such as climate variability, pathogen outbreaks, declining mulberry cultivation, and the limitations of conventional breeding. A systems biology framework that integrates multi-omics, bioinformatics, and genome editing has emerged as a transformative strategy for improving *Bombyx mori* and other silk-producing species. This integrative approach enables the identification of candidate genes, validation of their functions, and translation of molecular knowledge into practical breeding outcomes.

6.1 Multi-Omics as the Foundation for Target Discovery

Multi-omics provides a comprehensive view of silkworm biology by combining genomic, transcriptomic, proteomic, and metabolomic data:

- **Genomics:** The sequencing of the *B. mori* genome provided the foundation for large-scale trait dissection (Biology Analysis Group et al., 2004). Comparative genomics with wild silk moths and mulberry host plants has revealed loci linked to domestication, silk quality, and stress tolerance (Sun et al., 2012).
- **Transcriptomics:** High-resolution RNA-seq studies have uncovered tissue-specific and developmental stage–specific gene expression in silk glands, midgut, and immune tissues, clarifying fibroin synthesis, cocoon colouration, and immune regulation (Niu et al., 2025).
- **Proteomics:** Quantitative proteomic mapping has identified fibroin and sericin secretion pathways, along with post-translational modifiers that influence silk

elasticity and tensile strength (Li et al., 2015).

- **Metabolomics:** Profiling of metabolic pathways has linked energy metabolism to cocoon quality, showing how diet and environmental stress alter metabolite pools in silk gland cells (Xu et al., 2019).

The integration of these datasets provides robust targets for genome editing, ensuring precise interventions rather than random breeding trials.

6.2 Role of Bioinformatics in Multi-Omics Integration

The complexity of multi-omics datasets requires advanced bioinformaticspipelines to integrate information across molecular layers. Databases such as SilkDB and KAIKObase now host genomic, transcriptomic, proteomic, and epigenomic data for *B. mori* (Lu et al., 2020). Bioinformatics algorithms enable cross-layer mapping of gene–protein–metabolite interactions, predicting causal links between genetic variants and traits such as silk yield or pathogen resistance.

Moreover, machine learning models are increasingly being applied to predict silk protein secondary structures and regulatory motifs in fibroin promoters (Zhao et al., 2016). By training AI models on omics datasets, researchers can prioritize novel candidate genes for editing that may not be apparent from single-layer analyses.

These computational insights directly guide CRISPR-Cas9 applications, allowing targeted knockouts, knock-ins, or promoter modifications.

6.3 Linking Omics to Genome Editing Applications

Once high-confidence targets are identified through omics and bioinformatics, genome editing validates their function. Examples include:

- **Structural traits:** Transcriptomic studies identified the *BmCPG10* gene, encoding a cuticular protein that influences cocoon architecture. CRISPR knockout confirmed its role, producing cocoons with altered texture (Trochez-Solarte et al., 2019).
- **Silk productivity:** Proteomic studies highlighted silk gland chaperones critical for fibroin folding. Gene editing of these factors reduced secretion, validating their functional role (Zhang et al., 2018).
- **Stress tolerance:** Metabolomic profiling under thermal stress pointed to HSP-related genes. Editing these loci enhanced larval survival under heat stress (Sun et al., 2024).

The iterative cycle of omics-driven discovery, bioinformatics-based target prioritization, and genome editing–mediated validation represents a fundamental transition from conventional breeding toward precision sericulture.

6.4 Toward Next-Generation Silkworm Breeding

Integrating omics and genome editing opens direct industrial applications:

- **High-yield strains:** Editing fibroin regulatory genes enhances cocoon weight and shell ratio.
- **Disease resistance:** Omics-guided identification of immune regulators combined with CRISPR knockout of viral entry genes (e.g., *BmNPC1*) has produced virus-resistant strains (Dong et al., 2020).
- **Diet adaptation:** Genomic studies on strains such as *Guican No. 5* revealed adaptive loci for non-mulberry diets. Editing these loci may accelerate breeding of silkworms suitable for artificial diets (Xin et al., 2024).
- **Climate resilience:** Omics-informed stress markers, when combined with CRISPR editing, allow development of heat- and oxidative stress–tolerant strains.

These advances reduce dependence on conventional crossbreeding and accelerate the development of commercially viable silkworm lines.

6.5 Sustainability and Global Implications

Beyond productivity, sustainable sericulture must balance environmental stewardship and rural livelihoods. Omics-guided genome editing can:

- minimize pesticide/chemical use by breeding pathogen-resistant strains,
- reduce rearing costs via artificial diet adaptation,
- expand silk product diversity through biomaterial innovation.

Importantly, this approach is not limited to *B. mori*. Wild silkmoths such as *Antheraeaassamensis* (muga) and *Samiaricini* (eri) also benefit from cross-species omics

resources. Comparative studies have revealed conserved silk protein motifs across species, offering opportunities for pan-sericulture improvement.

By uniting multi-omics, bioinformatics, and genome editing, sericulture can evolve into a high-tech, sustainable bioindustry aligned with future global demands.

7. CONCLUSION

Sericulture is entering a transformative phase where traditional breeding approaches are being complemented—and in many cases surpassed—by the convergence of multi-omics, bioinformatics, and genome editing. Multi-omics technologies provide system-level insights into silk gland biology, immune responses, and environmental adaptability, while bioinformatics platforms translate these complex datasets into practical targets for trait improvement. Genome editing tools, particularly CRISPR/Cas9 and its emerging derivatives, offer unprecedented precision in validating candidate genes and engineering silkworm strains with enhanced silk yield, fiber quality, pathogen resistance, and climate resilience. Together, these advances establish a framework for precision sericulture that is faster, more efficient, and more sustainable than conventional methods. Importantly, the implications extend beyond silk production to biomaterial innovation, pharmaceutical applications, and rural livelihood security. Moving forward, the integration of omics-driven discovery, AI-enabled bioinformatics, and next-generation editing technologies will be central to building a resilient, eco-friendly sericulture industry that aligns with global demands for sustainable materials and biotechnology solutions.

DISCLAIMER (ARTIFICIAL INTELLIGENCE)

Author(s) hereby declare that NO generative AI technologies such as Large Language Models (ChatGPT, COPILOT, etc) and text-to-image generators have been used during writing or editing of this manuscript.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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