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Recent trends in Seri-bioscience: Its prospects in modern sericulture

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Abstract

The recent advances in the various spheres of bioscience viz., seri-biotechnology, bio-nanotechnology, crop protection, etc., have revolutionized research in sericultural science at the world level. However, India is yet to reap the benefits of seri-bioscience to bring out products in the form of technology as the same is still in infant stage in spite of making sincere but compartmentalized attempts. In view of this status, we have to consolidate our focus on developing technologies targeted at improvement of silkworm and host plants. Significant seasonal variations occur in the nutritional value and composition of host plant depending on factors such as weather, pests and diseases and agricultural practices which in turn have a great impact on growth and development of silkworm. Lower animals do not have well developed humoral immunity and under such circumstances vaccine development may not be of much use and in these lower animals and factors that achieve immune-stimulation could be very useful for disease resistance. Promoting the establishment of silkworm models in scientific studies will provide new solutions and new insights into traditional views of problem-solving and greatly benefit science as well as the society. The present article is an overview on the status of work conducted on seri-bioscience, future approaches and the road ahead.

Keywords: Seri bioscience, silkworm, molecular biology, bio-fungicide, silk, transgenics

Introduction

Sericulture is the art and science of rearing silkworms by feeding their host plants in order to obtain the cocoons spun by them from which silk is obtained. It is an agro-based cottage industry which deals with the mass-scale rearing of silkworms (Ganga and Chetty, 2017) [7]. The caterpillars of the domesticated silkworm *Bombyx mori* are the most commonly used silkworm species in sericulture. Other types of silkworms (such as eri, muga and tasar) are also cultivated for the production of 'wild silks' or 'Vanya Silk'. Assam holds a unique position in muga and seri culture in the world. The golden muga silk is the gift of nature to the state. It is the "Pride of Assam" and the extreme monopoly of northeast India (Gogoi *et al.*, 2017) [8]. Sericulture is a very important domestic industry in many countries of the world (Anon., 2021a) [1]. India and China are the world's leading producers of silk. The silk output of these two countries combined accounts for over 60% of the global production.

Sericulture comprises of the following activities:

- Cultivation of host plants to produce leaf
- Rearing of silkworm on the host plants to get the cocoon
- Reeling of the cocoon to obtain silk yarn

Bioscience is a diverse field of study. It deals with the science of applying technology to develop biological solutions that sustain, restore and improve the quality of life for humans, plants and animals on earth. It consists of various branches like: biotechnology, nanotechnology, botany, zoology, genetics, microbiology, molecular biology, biophysics and biochemistry, etc. Seri-bioscience is the field of study which deals with the science of applying knowledge to develop biological solutions to sustain, restore and improve the quality/quantity of silkworms, their host plants and ultimately the silk. In the host plant cultivation and silkworm rearing sector, silk farmers practising sericulture face several challenges that could potentially destroy their harvest and cause economic loss to them (Kovarova *et al.*, 2021) [10]. The host plants and silkworms are prone to many health hazards. The host plants such as mulberry, castor, Som, Sualo, Arjun, Asan, *Quercus* sp. *etc.* are prone to various diseases like

leaf spot, blight, wilt, die back, powdery mildew, stunted growth due to nematode infestation, infestation by pests like semi-looper, capsule borer, jassids, thrips and whiteflies, etc. Silkworms are vulnerable to several diseases such as pebrine, muscardine, grasserie and flacherie. The healthy growth of silkworms larvae is also threatened by several pests. The pebrine disease can infect the eggs, resulting in their death before the hatching of the larvae. Any larvae affected by this disease develop dark pepper-like black spots on the body and become lethargic. Viral infections in the larvae may result in inter-segmental swelling and shrinkage of silkworm body. They may also start giving off an unpleasant odour. Other viral infections such as cytoplasmic polyhedrosis can cause the larvae to lose their appetite. The muscardine disease caused by fungi can cause the larvae to become mummified, feeble and eventually die (Anon., 2021a) [1]. The larvae of dermestid beetle can bore into the silkworm cocoons and eat the pupae. Silk cannot be reeled from these defective cocoons. In the post cocoon or reeling sector, the farmers have to deal with the problems related to grainage and storage of cocoons.

Recent areas of advancement in seri-bioscience

Seri-biotechnology

Seri-biotechnology is the area of biology that uses living processes, organisms or systems to produce products like antibiotics, hormones, etc. or technology intended to improve the quality of silkworm and their host plants (Mahesha, 2017) [11].

The Seribiotech Research Laboratory [SBRL] was established during 1993 under the World Bank aided National Sericulture Project as per the advice of a high level committee headed by Prof. Lynn Riddiford, University of Washington, USA to carry out research in the frontier areas of biology for the development of the sericulture industry.

The following are the mandates of the laboratory

a) To conduct research in frontier areas of modern biology

and to seek potential applications of these work towards improving silk productivity.

- b) To interact with other institutions doing basic or applied research in areas related to sericulture and other allied areas.
- c) To disseminate technology developed to the target groups through the other R & D constituents of CSB.

Functions

Presently the laboratory is implementing research projects in four main areas as indicated below:

Silkworm Genomics

The focus is on identification of silkworm genes and their functions associated with resistance to viral pathogens, regulation of diapause, regulation of yolk proteins, characterization of RNA dependent RdRp (RNA-dependent RNA polymerase) gene etc.

Host Plant Genomics

They aim on molecular characterization and identification of various mulberry species and other host plants, development of microsatellites for mulberry etc.

Proteomics

The focus is on identification of immune response proteins and their interactions, and silkworm transcriptome analysis under stress from pests like uzi fly etc.

Molecular Pathology

Focus is on identification and molecular characterization of various pathogens like virus, bacteria, microsporidia etc. infecting silkworms, diagnosis of pathogens using molecular tools and development of diagnostic tools for the detection of virulent and non-virulent strains of microsporidia (Mahesha, 2017) [11].

Table 1: Mulberry cultivars developed through tissue culture technique

Species	Cultivar	Explants used	References
<i>Morus indica</i>	K2	Cotyledon, leaves, petioles, internodal segments, roots	Bhatnagar <i>et al.</i> , 2001
<i>Morus alba</i>	M5, S36, S13, Chinawhite	Leaves	Chitra and Padmaja, 2005
<i>Morus alba</i>	Sujanpur	Nodal segments	Kashyap and Sharma, 2006

Plant tissue culture

Plant tissue culture is a technique used to design, maintain and grow plant cells, tissues or organs *in vitro* under sterile conditions on a nutrient culture medium of known composition. Tissue culture clones are 'true-to-type' as compared with seedlings which show greater variability and also it helps to culture virus-free or disease-free plants (Anon., 2021b) [2].

In mulberry, it is difficult to produce the desired hybrids due to several barriers such as environmental barriers, pollen incompatibility and unreceptive nature of stigma, maturity of female and male flower parts at different times and sometimes the uneven ploidy status of the two parents (Khan *et al.*, 2016) [9]. To overcome this problem, the CSR&TI, Mysore has developed several cultivars of mulberry using tissue culture technique using various explants. Apart from this, the CMER&TI, Lahdoigarh has been successful in performing clonal propagation of Som plant, the primary host plant for muga silkworm through single leaf bud cutting.

Table 2: Mulberry varieties developed suitable for different conditions

(1)	For Irrigated Conditions
	K ₂ (M ₅), S-54, S-36, V-1, G-2, G-4
(2)	For Rainfed Conditions
	S-13, S-34, RFS-135, RFS-175, AR-11, RC-1, RC-2
(3)	For Alkaline Conditions
	AR-12
(4)	In Shade Conditions
	Sahana (Kanva-2 x Kosen)

Genetically modified organisms (GMOs)/transgenics

Genetically modified organisms (GMOs) are organisms in which the genetic material (DNA) has been altered in a way that does not occur naturally by mating or natural recombination. Transgenic organisms contain a transgene *i.e.* foreign DNA from another species, or else a recombinant DNA from the same species that has been manipulated in the laboratory then reintroduced.

Transgenic/genetically modified silkworm

Japanese scientist Tetsuya Iizuka, 2014 from the National Institute of Agro-biological Sciences created silkworms that can successfully spin silk which glow in the dark. For this, DNA sequences that produce foreign fluorescent proteins were inserted into the silkworm genome which gets expressed in the silk spun by the transgenic silkworms. The fluorescent genes were initially derived from fluorescence genes cloned from coral and jellyfish. Red fluorescence genes originated from *Discosoma* corals, orange from *Fungia concinna* coral and the green fluorescence protein from jellyfish. While the transgenic silk looks like regular silk in daylight, fluorescence can be observed under UV light. The silk fluorescence proteins stay vibrant and continue glowing for more than two years. Despite the transgenic silk being slightly weaker in tensile strength than conventional silk, fluorescent silk has been popular for garments including wedding dresses, ties and suits.

Spider silk producing transgenic silk moth (monster silkworm) Due to the cannibalistic and territorial nature of spider, it is very difficult to rear spiders in order to obtain the spider silk which is famous for its mechanical properties. A massive spider silk production system in *B. mori* was developed in 2018 by using transcription activator-like effector nuclease-mediated homology-directed repair to replace the silkworm fibroin heavy chain gene (*FibH*) with the major ampullate spidroin-1 gene (*MaSp1*) in the spider *Nephila clavipes*. The transgenic produced was called monster silkworm, which had red eyes for their identification from the normal mulberry silkworms. The ~ 16-kb endogenous *FibH* gene was successfully replaced with a 1.6-kb *MaSp1* gene fused with a 1.1-kb partial *FibH* sequence and achieved up to 35.2% chimeric MaSp1 protein amount in transformed cocoon shells (Xu *et al.*, 2018) [16]. The presence of the MaSp1 peptide significantly changed the mechanical characteristics of the silk fiber, especially the extensibility. This study provides a native promoter-driven, highly efficient system for expressing the heterologous spider silk gene instead of the transposon-based, random insertion of the spider gene into the silkworm genome. Targeted *MaSp1* integration into silkworm silk glands provides a paradigm for the large-scale production of spider silk protein with genetically modified silkworms and this approach will shed light on developing new biomaterials.

RNAi technology

RNA interference (RNAi)

It is a regulatory system that controls the activity of genes (Smith, 2021). RNAi functions specifically to silence or deactivate genes. The Juvenile Hormone Epoxide Hydrolase (JHEH) of *Bombyx mori* is a hydrolytic enzyme responsible for degradation of Juvenile Hormone (JH). Since rapid JH degradation signals onset of pupation in silkworm, gene silencing of JHEH by RNAi technology might extend the larval stage and leads to the formation of super cocoons, with increased silk productivity.

Achievements in silk biotechnology

The DBT has established a Centre of Excellence in Genetics and Genomics of Silkworm at Centre for DNA Fingerprinting and Diagnosis, CDFD, Hyderabad. Transgenic lines of silkworm resistant to baculovirus (BmNPV) have been generated through RNAi technology (Ponnuvel *et al.*, 2013) [14]. The transgenic lines are being transferred to sericulture centres for controlled trials after necessary clearance from

RCGM. Analysis of immune transcriptome of bacteria-challenged wild silkworm, *Antheraea mylitta* led to the identification of a number of potential immune-related genes. Eleven potential silkworm (*Bombyx mori*) lines tolerant to BmNPV varying from 27 per cent to 67 per cent have been developed through DNA-marker assisted breeding jointly at Seribiotech Research Laboratory (SBRL), Bangalore and Andhra Pradesh State Sericulture Research and Development Institute (APSSR&DI), Hindupur. The selected lines are at BC-5 F-9 generation and are being further taken up for field trials. A programme on comparative genetic analysis of sex chromosomes and sex determining genes in silkworms has been continued at CDFD, Hyderabad. Two novel genes predicted to be involved in the silkworm sex determination, were identified using various biochemical and molecular procedures. Their validation is in progress. W-derived gene *Bmz1* has been analysed for its involvement in sex determination. Full-length cDNA of vitellogenin receptors from *B. mori* and *Samia ricini* and lipophorin receptor from *S. ricini* have been determined. Functional characterization of the receptors for their possible role in fecundity using RNAi and binding assays is under progress. Work has been continued under a Consortium Project on Mulberry Genomics (CPMG) involving four institutions – CSR&TI, Mysore, CSGRC, Hosur, UAS, Bangalore and UDSC, New Delhi. A total of 188 genomic and genic SSR markers have been developed in *Morus alba*, which can be utilized in assessing molecular diversity as well as in QTL mapping towards crop improvement through marker assisted selection at UAS, Bangalore. Transcriptome analysis has been carried out to identify SSRs, SNPs and novel drought responsive genes using mulberry leaf tissues exposed to different levels of drought stress jointly at UAS, Bangalore and UDSC, New Delhi. Over 10,000 ESTs have been identified and annotated. Ten drought tolerance genes have been cloned from mulberry and are being functionally validated in Arabidopsis. Work has been recently initiated to identify QTLs conferring resistance to root rot disease and trait introgression in mulberry at CSR&TI, Mysore. Application of sericin (a silk protein) on alkali modified polyester using a cross linking agent has been developed at IIT, New Delhi. Improved smoothness, moisture retention and wicking were observed which made the fabric more suitable for medical and sport garments. Silk based super macro porous cryogel matrices have been synthesised successfully for exploring their potential in cell-material interactions at IIT, Kanpur. Composite silk based biomaterials were synthesized which can be applied for soft and hard tissue engineering applications. The synthesized materials were thoroughly characterized by scanning electron microscopy, micro-computer tomography, TF-IR, mechanical properties, *in vitro* and *in vivo* biocompatibility. Nano-silk sericin based hydrogels prepared from silk industry waste have been found to have application in sanitary napkins and baby diapers at RV College of Engineering, Bangalore. The scaffold of nano-fibroin was prepared as an ideal biopolymer for enzyme immobilization.

The dressing membrane made up of fibroin has been found to heal wound much faster. Under post harvest technology, degumming of silk with fungal protease enzyme, a valuable alternative to the existing methods of degumming has been developed at IIT, New Delhi. In India, molecular markers for high cocoon shell weight as well as virus resistance are developed using divergent parents like Sarupat and CSR2 (Meng *et al.*, 2017) [12].

Nanotechnology

Nanotechnology refers to the field of applied science which works on the atomic or molecular level in scales smaller than 1 micrometer, normally 0.1 to 100 nanometers. It is a rapidly evolving field with the potential to forward the sericulture and silk industry with new tools which promise to increase silk production in a sustainable manner and protect crops from pests and diseases.

Nanoparticles

Bombyx mori cytoplasmic polyhedrosis virus (BmCPV) is a serious disease harmful to silk industry. So far, there is still no good way to prevent or treat this disease. In a recent study, titanium dioxide nanoparticles (TiO₂ NPs) were used to pre-treat silkworm larvae, and good results were achieved in improving silkworm immunity and alleviating the damage of cytoplasmic polyhedrosis virus. The results showed that nanotitanium dioxide pre-treatment could inhibit the proliferation of BmCPV in the midgut of silkworm, activate JAK/STAT and PI3K-AKT immune signaling pathways, and up regulate the expression of key immune genes, so as to improve the immunity of silkworm and enhance the resistance of silkworm to BmCPV (Zhao *et al.*, 2020)^[19].

Silk sericin facial mask

Sericin is reported to have diverse biological activities, such as anti-oxidation, anti-bacterium, anti-coagulation and promoting cell growth and differentiation. In the field of regenerative medicine, owing to its biodegradability, easy availability, and hydrophilicity with many polar side groups, sericin is mostly copolymerized, crosslinked, or blended with other polymers to form various scaffolds in order to help obtain improved properties for relevant biomedical applications, such as skin regeneration. Besides being jointly fabricated with other biomaterials, the possibility of using only pure sericin to generate scaffolds has just begun to be explored. Silk sericin face mask provides superior quality over the conventional serum-soaked cloth masks. Compared with the traditional standard essence masks, silk sericin facial mask provides extraordinary moisturizing quality that has been proven to be superior to the traditional essence masks by up to 18%. The new concept of Thermo-Sensible Water-soluble gel mask gradually melts its active ingredients into skin, reacting to the skin temperature. It also utilizes the bio-matrix method, which is safe for the human body (Anon., 2021c). This gel mask has a great cooling effect and it keeps skin moisturized as well. The enhanced moistness and adhesive ability guarantees effective delivery of active ingredients into the deeper skin without damage, while resolving the problem of other sheet-type masks' losing active ingredients.

Surface modification

Super hydrophobic silk fabric surfaces with high-aspect-ratio nanostructures were fabricated using ion beam treatment. The ion beam irradiated silk fabrics were characterized for wettability, as well as other physical properties unique to silk. The nanostructures were produced in various configurations, ranging from columnar to hairy shapes, on the silk fibres through anisotropic etching using oxygen ion beam treatment (Oh *et al.*, 2014)^[13]. Subsequent hydrophobic coating on the nano structured, super hydrophobic silk fiber surfaces was achieved with an increase of the static contact angle from 0° for the pristine hydrophilic silk fabric to 170° for the super

hydrophobic silk fabric and with a decrease of shedding angle by less than 5°, which is sufficient roll-off a water droplet to the silk fabric surface. Because the ion beam-treated side of silk fabric becomes super hydrophobic, while the opposite side, or body contacting side, remains pristine or super hydrophilic, an extreme asymmetric wettability can be achieved in the silk fabric, which improves its breathability by improving moisture transmittance through the fabric from the body to the outer surface. The luster and color of silk fabric before and after ion beam irradiation were found to exhibit no significant degradation. The breaking load of the silk fabric after the ion beam treatment was assessed to be mechanically durable in comparison to that of the pristine fabric. Therefore, after introducing the super hydrophobic property *via* the ion beam treatment, the silk fabric maintained its primary advantages, as a result, the range of its applications can expand into breathable self-cleaning clothing textiles, such as neckties, blouses and dresses.

Crop protection

Crop protection is the science and practice of managing diseases, weeds and other pests that damage sericultural host plant plantations or silkworm crop.

It has 2 basic divisions:

1. Host plant protection
2. Silkworm crop protection

For host plant protection, several formulations have been developed by the CSR&TI, Mysore.

Bio-fertilizers/ micronutrients/ mulberry protection

Azotobacter: It is a free-living nitrogen-fixing bacterium, which is used as a biofertilizer in the cultivation of most crops. Utilization of this specific strain of *Azotobacter chroococcum* isolated from the rhizosphere of mulberry plant assures a substantial saving of nitrogen fertilizer and thus indirectly reduces the costs involved in the silkworm rearing.

Morizyme-B

Use of plant growth hormones in crop production helps in increasing the leaf area index which leads to an overall increase in the yield of crops. A new formulation based on naphthalene acetic acid (NAA) has been developed by the Central Sericultural Research and Training Institute (CSB), Berhampore. The formulation in addition to NAA contains other micronutrients which are essential increasing the leaf yields and overall yield of crops.

Navinya

It is a formulation mainly consisting of plant based anti-fungal biological compounds that are eco-friendly; non-toxic and non-corrosive for the management of the root rot fungus, *Rhizoctonia bataticola* (Taub.) in the mulberry. In addition to the biological compounds, few selected organic and inorganic chemicals in small proportions are also used for the preparation of the formulation that function as a synergistic to biological compounds and also these enhance the basic metabolism of the mulberry plant. Trials carried out in the field using this formulation have been very positive in the recovery and rejuvenation of the infected mulberry plants and further spread of the disease has been checked. There has been no residual effect any of the chemicals and the sericulture farmers was able to harvest and use the mulberry leaves for the silkworm rearing without any effects on the silkworms (Bhat and Choure, 2014)^[5].

Nemahari

It is a plant based eco-friendly product formulation with 75% plant components and 25% chemical ingredients has been developed for the management of root knot disease of Mulberry. Tests conducted under laboratory level revealed reduction of disease severity by almost 82% and prevented loss of leaf yield by 22%. Trials conducted at Farmers fields resulted in reduction of disease incidence by 82 to 84% and reduction in leaf yield loss to the tune of 22-24%. Adoption of this technology has resulted with a cost benefit ratio of 1:2.2 when compared with the traditional practices of management of root knot disease.

Poshan

With introduction of high yield mulberry varieties and improved agronomical practices, the demand for nutrients for achieving higher production has increased leading to depletion of nutrients in soil and consequently leading to nutritional deficiencies in mulberry crop. Poshan, a well-balanced multi-nutrient foliar formulation can be sprayed on mulberry crop for correcting the nutrient deficiencies and to improve the leaf quality. Successful trials were conducted at the Institute and farmers' field for correcting the nutrient deficiencies and improving the leaf yield and quality. Poshan has shown to be better than the existing formulations available in the market with higher cost - benefit ratio. The formulation can be manufactured by using simple organic and inorganic chemicals available indigenously.

Raksha

Trichoderma harzianum is a naturally occurring fungus, which is used as a biofungicide to protect mulberry (*Morus alba* L.) from root-rot and wilt diseases caused by other harmful fungi. A talc-based biofungicide (Raksha) has been developed using *T. harzianum* for the control of root rot diseases of mulberry. Raksha is applied after mixing with farm-yard manure (FYM) (1:50) @ 500 g mixture/plant in the root zone followed by irrigation. There are no expected health risks in human and residual toxicity in mulberry as well as silkworm from the use of product containing *T. harzianum* as active ingredient. The mass production and quality control methods for production of the biofungicide have been standardized.

Disinfectants

Amruth

Amruth is an eco-friendly botanical based, non-toxic, non-polluting and biodegradable powder formulation meant to be fed to silkworm to control both the grasserie and flacherie diseases caused by bacteria, viruses or their combined infection. Amruth has antiviral and antibacterial properties. This suppresses the multiplication of the pathogen in the host and also improves the immunity level of the host thereby lowering the mortality. The product is prepared by mixing indigenously available eco-friendly chemicals and botanicals in definite proportions, pulverized in to 250 mesh size powder, packed in polythene packets and sealed. For application, the powder formulation is mixed in clean water @2.0g/100 ml.

This liquid is sprayed uniformly on mulberry @70.0 ml/kg leaf, air-dried and then fed to silkworms. This is done immediately after the detection of the disease and repeated once after 24 hour in the same instar and is continued once in ever instar.

Ankush

It is an eco-friendly silkworm body and rearing seat disinfectant aimed at avoiding environmental deterioration and user friendly by eliminating the hazardous chemicals in the product. The product is also biodegradable, economical and as effective as other popular bed disinfectants. The product is prepared by mixing indigenously available eco-friendly chemicals and botanicals in definite proportions (all in dry state) and pulverized in to 250 mesh size powders. This mixture is then packed in polythene packets, sealed, stored in dry and cool place before marketing The shelf life is one year. The cost of production for a kg of product is Rs.12.00.

Silkworm growth promoters/ silkworm pest control

Nutrid

Nutrid is an artificial diet for replacing mulberry leaves during the rearing young age silkworms (first and second instar). The artificial diet is prepared hygienically using mulberry leaf powder, plant products and other chemical ingredients. This product saves young age leaves and provides more hygiene during rearing the fragile young age silkworms. Use of this diet results in saving Rs.150.00 in comparison to feeding on mulberry leaves for 100 disease free layings.

Phytoecdysone (Sampoorna)

Phytoecdysone (20-hydroxy ecdysone) is a molting hormone extracted from a locally available weed. This hormone is used in the early and uniform maturation of silkworms in the fifth instar so as to synchronize the spinning of cocoons. This results in saving of precious mulberry leaves for further rearing and also saving in manpower. Suitably diluted concentration of this hormone is sprayed on the silkworms directly and within 24 hours all the worms are ripened for spinning of cocoons.

Microbiology

Disease management

LAHDOI: Muga silkworm (*Antheraea assamensis*) is one of the economically important wild silk reared in tropical North Eastern region of India. It is a polyphagous insect, which feeds on leaves of Som, Soalu and other plants. The muga silkworm rearing is semi-domesticated and multivoltine in nature and reared by about 35,000 farmers' family. Precious golden yellow muga yarn is produced from the muga cocoons. Muga silkworm rearing being outdoors is exposed to various rigors of changing environment and is prone to number of bacterial, viral and fungal diseases causing crop loss and cocoon production. Among the fungal diseases, muscardine disease caused by *Beauveria bassiana* is predominant and causes upto 60-90% losses in muga silkworm crop. The LAHDOI formulation for the management of muscardine is in powder form and is non-toxic to silkworm and non-phytotoxic to the host plant. It is used for foliar application on the host plant of muga silkworm during any/all rearing stages after suitable dilution. By using LAHDOI there was improvement in the effective rearing rate of muga silkworm and reduction in disease incidence. The cost benefit ratio worked out was 1:75.24.

Jeevan Sudha

Jeevan Sudha is a botanical formulation developed by Central Tasar Research and Training Institute (CTR&TI) Ranchi, Jharkhand from the medicinal plants for containment of virosis in tasar silkworm, *Antheraea mylitta*. Application of

1% aqueous extract of Jeevan Sudha during rearing protects tasar silkworm from virosis. Aqueous extract is given in three doses as foliar spray, once each in 1st, 2nd and 3rd instars during feeding stage. Jeevan Sudha reduces virosis by 37%, resulting in an improvement of minimum 10-12 cocoons/dfl at laboratory and farmers field. Cost benefit ratio is 1:6.

Sericillin

It is a synergistic composition for disinfecting silkworm body and silkworm bed. Sericillin is a cost-effective bed disinfectant. It has been formulated to prevent all common silkworm diseases thus ensures crop stability. This powder formulation is found effective against muscardine as well as it is equally effective against all common silkworm diseases such as grasserie, flacherie, gattine and pebrine. It inactivates all types of pathogens of silkworm existing on the rearing bed and silkworm integument and thus prevents secondary contamination.

The formulation contains mixture of three locally available chemicals. The formulation maintains 1% chlorine that inactivates all fungal, bacterial, viral and protozoan pathogens of silkworm. Powder is to be dusted on the silkworm bed @ 3-4g /sq. ft of bed area.

About 4 kg Sericillin is required for the whole rearing of 100 dfls i.e., 40,000 larvae. Sericillin is now being popularized at farmers' field in Eastern and North Eastern region for control of major silkworm diseases.

Bioformulations for management of diseases of mulberry

Bionema

A bionematicide for control of root knot disease. The *Verticillium chlamydospreum* based bio-formulation developed by CSRTI, Mysore is very effective against root knot (*Meloidogyne incognita*) disease of mulberry. It reduces the diseases up to 80-85%.

Raksha

A biofungicide for the control of root rot disease. A bio formulation of *Trichoderma harzianum* is very effective for management of root rot (*Fusarium solani* and *Fusarium oxysporum*) diseases of mulberry. The disease is reduction is up to 75-8%.

Nursery Guard

It is a very effective for control and management of nursery diseases like stem canker & die back (*Botryodiplodia theobromae*), collar rot (*Phoma sorghina* and *Phoma mororum*) and cutting rot (*Fusarium solani*), a bio-formulation consisting of the bio agent *Trichoderma pseudokoningii*.

It suppresses the incidence of nursery diseases by 80% & increases sapling survivability by 40%.

Chetak

Chetak controls all major foliar diseases of mulberry such as leaf spot, powdery mildew, leaf rust and blights and major soil borne diseases viz., stem canker, cutting rot, die back, root knot, root rot and diseases complex (nematode + root rot pathogens). The disease reduction is upto 85-90%.

Bio-mix

Bio-mix is a consortium of *Trichoderma harzianum* and *T. viride* developed for management of root disease complex. The disease reduction is upto 75-80%.

Tri-mix

A bio-consortium for management of root rot disease. Tri-mix prepared from region specific (Karnataka, Tamil Nadu and Andhra Pradesh) isolates of *Trichoderma harzianum* was developed for management of root rot disease caused due to pathogen complexity (*F. solani*, *F. oxysporum*, *B. theobromae* and *M. phaseolina*). The disease reduction is upto 75-80%.

Compo-mix

Compo-mix is a *Trichoderma* based bio-inoculants used for hastening both aerobic and anaerobic composting process to produce compost with disease suppressive quality. One kg of Compo-mix is required for treating 1,000 kg of raw material.

Probiotics

Probiotics are live microorganisms which provide health benefits when consumed, generally by improving or restoring the gut flora. Enriching mulberry leaves with vitamins, minerals, hormones, antibiotics, probiotics, sugars and botanicals influence the growth, development and disease resistance in silkworm (Yeruva *et al.*, 2020)^[17].

Probiotics in sericulture

Micro-organisms play an important role in the growth and development of numerous insect species. The mulberry silkworm, *Bombyx mori* (Lepidoptera), harbors several bacteria in its midgut aiding the metabolic processes; however, the variability of bacterial spp. present in the midgut and their role(s) in the growth and development of the silkworm are poorly understood. In a recent work, the diversity of midgut bacterial communities in silkworms of variable voltinism (Pure Mysore, PM: multivoltine; CSR2: bivoltine and PM × CSR2: crossbreed) was compared through metagenomics (Yeruva *et al.*, 2020)^[17]. The predominance of *Enterococcus* (30.30%) followed by *Bacillus* (16.96%) was observed in PM, whereas *Lactobacillus* (56.56%) followed by *Enterococcus* (10.58%) was seen only in CSR2. Interestingly, crossbreed harbored diverse bacterial communities (36.21% *Lactobacillus*, 25.94% *Bacillus*, 8.1% *Enterococcus*, and 18.37% uncultured bacteria). Meta genomic profiles indicate variability in the gut bacterial population in different kinds of silkworms influencing the physiological activities accordingly.

The dominant bacteria, particularly *Lactobacilli*, *Bacilli*, and *Enterococci* could be further explored for identifying the potential probiotic consortia based on a literature survey and potential involvement in nutrient absorption, disease/stress tolerance, and improved economic traits (Zafar *et al.*, 2013)^[18].

An experiment reported that oral immunization of probiotics containing *Lactobacillus acidophilus* controlled bacterial disease in mulberry by 88.62% and other diseases by 55.25%. The experiment was done in multivoltine breeds like Nistari (M), M12 and M6DP (Yeruva *et al.*, 2020)^[17].

Glucosamine HCl from silkworm Glucosamine (2-amino-2-deoxy glucose) is cellular glue naturally present in the body. It is primary building block for proteoglycans and offers support for building and maintenance of collagen between joints. It also helps to maintain healthy connective tissues. The function of proteoglycans is to attract water into the joint space of cartilage for lubrication during movement (Anon., 2021d)^[4].

Glucosamine HCl is for people who are experiencing limited flexibility related to their joints. It is intended for active

people who are aging and can also be used by those who participate in sports.

Tasar Rakshak

Tasar silkworm (*Antheraea mylitta D.*) is reared in tropical India for production of tasar silk in commercial scale. Rearing of tasar silkworm causes major loss (around 40%) to the rearers due to different silkworm diseases caused by pathogenic organisms. Certain leaf surface microbes (LSM) of tasar food plants (*Terminalia arjuna*) are capable of inhibiting the growth of other micro organisms including the pathogens of tasar silkworm (Anon., 2021d) [4]. The potent antagonistic bacteria having a strong antimicrobial action are cultured in the laboratory and the formulation prepared is sprayed as foliar application during rearing of tasar silkworm which prevents and controls bacterial and viral diseases of the silkworm larvae reared on its food plants in outdoor condition. The trials resulted in reduction of silkworm diseases and increase in cocoon production to the extend of 12-14 cocoons per dfl (disease free layings). A net gain of Rs.1000 per 100 dfls has been obtained at the farmers' level by investing Rs.40.00 towards the cost of this formulation. The newly evolved formulation is economic, eco-friendly, cost-effective and can be easily be affordable by the tasar growers.

Challenges

No doubt, the recent advancements made in different areas of seri-bioscience will certainly help sericulture to achieve a platform from where this sector can be seen flourishing in all specters. But to reach that goal, it has to face certain challenges like:

The extent of knowledge and level of adoption of improved technology by the farmers. Hazards of chemical dyes to silkworm health needs further study. Rate of effectiveness and compatibility of bioagents with changing soil microflora. Requirement of huge initial investment for tissue culture techniques may generate reluctance among farmers.

Conclusion

The various recent trends in the field of seri-bioscience discussed above have immensely modified sericulture from a cottage industry to a designation of fashion, luxury and biological model at the international level. But India is still yet to reap the fruit of such modern technology.

Future prospects

Bio-science has been successfully used in various aspects of sericultural research and has greatly promoted scientific development in this field. However, there are still many challenges ahead and the application of bioscience in many areas is just at the initial stage such as product-diversification through use of nano-technology, advanced study on probable adverse effects of microbial bio-pesticides used in agriculture and biotechnological intervention for solving problems related to sericulture like host plant/ silkworm diseases and pests, particularly in the non-mulberry sector. Promoting utilization of silkworm as model organism in scientific studies will provide new solutions and new insights to traditional views of problem solving and greatly benefit not only the sericulture sector but also science as well as the society as a whole.

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