Application of Nanotechnology in Sericulture: A review

Abstract

One of the beneficial insects, silkworm (*Bombyx mori* L.): produces abundant silk in the form of a cocoon by ingesting mulberry leaves (*Morus alba*) in the course of the larval period. Factors such as food, humidity and temperature affect the physiology of insects. Vegetative part is the main component of mulberry, which is stimulated by application of fertilizer. Nanoparticles have received considerable attention in the field of agrochemicals due to their special properties, such as small particle size, surface structure, solubility and chemical composition. The application of nanotechnology in agrochemicals dramatically overcomes the defects of conventional agrochemicals, including low bioavailability, easy photolysis and organic solvent pollution, etc. In this review, we describe advances in the application of nano particle for mulberry to enhance its growth, development and also its impact on silkworm development and cocoon productivity.

Keywords: Sericulture, Mulberry silkworm (*Bombyx mori*), Nanotechnology, Nanoparticle.

Introduction

The mulberry silkworm economically important insect due to its silk secretion. The production of quality cocoons attributes to nutrition of the silkworm Mulberry leaves serves as ideal nutritional food for silkworm, which expands the quality and quantity of cocoon production. The essential trace elements *viz*., iron, nickel, cooper, manganese, potassium, zinc and iodine should be included in nutrition of silkworm growth. Mulberry leaves contains the vitamins which provides minimum requirement of silkworms and these vitamins varies on ecological conditions, fertilizer doses and mulberry varieties. The mulberry leaf quality is influenced by variety of spacing's, irrigation levels, nitrogen levels, seasons and the extra foliate that are supplied exogenously through mulberry leaves (Triped *et al*., 2009). Enrichment and fortification of mulberry leaves can be enhanced the quality and increase the quantity of cocoon ultimately silk productivity (Krishnaswami *et al*., 1978).

Nanotechnology is rapidly evolving field affords novel ways to enhance the growth and production in the field of nutrition and is expected to bring revolutionary changes in the field of agriculture. Nanotechnology has gained huge attention over time. The fundamental component of nanotechnology is the nanoparticles. Nanoparticles are particles between 1 and 100 nanometres in size and are made up of carbon, metal, metal oxides or organic matter [Hasan

2015]. Nanoparticles are generally defined as particulate matter with at least one dimension that is less than 100 nm. This definition puts them in a similar size domain as that of ultrafine particles (air borne particulates) and places them as a sub-set of colloidal particles (SCENIHR 2005). In 2008 the International Organization for Standardization (ISO) defined a nanoparticle as a discrete nano-object where all three Cartesian dimensions are less than 100 nm.

Nanoparticle components include silica, Fe, ZnO, titanium dioxide, cerium oxide, aluminium oxide, gold nanorods, ZnCdSe/ ZnS core-shell, P/ZnS core-shell and Mn/ZnSe quantum dots. The size, content, concentration and chemical properties of nanoparticle addition to type of the crop have a significant impact on their effectiveness as nano-fertilizers for plant growth. The release of nutrients into the soil occurs when nanoparticle (NP) containing nano-fertilizers react with water [Singh *et al*., 2021].

Nanoparticle such as Metal oxides, AgO, MgO, ZnO and TiO_2 are inorganic nanomaterials, whereas lipids, polymers and CNTs are organic nanomaterials. Biodegradable, natural and agriculturally safe carriers, such as chitosan, called polymeric NPs (Ghormade *et al*., 2011). Owing to the polymeric cationic properties and the ability to interact with negatively charged molecules or polymers, chitosan is a promising agrochemical carrier. Diverse types of nanomaterials like copper, zinc, titanium, magnesium, gold and silver nanoparticles have arisen with effective antimicrobial efficacy against viruses, bacteria and other eukaryotic micro-organisms. Some nanomaterials possess antiviral, antibacterial and antifungal properties and have an excellent capacity to deal with pathogen-related diseases (Castro *et al*., 2017, Makvandi *et al*., 2020).

The nanoparticles exhibit a unique physical, chemical and biological properties at nanoscale compared to their respective particles at higher scales. This phenomenon is due to a relatively larger surface area to the volume, increased reactivity or stability in a chemical process, enhanced mechanical strength, etc. [Assessment R 2007 cited by Elia 2017]. These properties of nanoparticles have led to use various applications including medicine, engineering, catalysis and environmental remediation. The inclusion of nanomaterial in sericulture are novel; therefore, it is imperative to know and exploit their effects on mulberry silkworms and silk productivity. This review attempts to provide information about the nanoparticles effect on the mulberry and silkworm growth and development via covering the related information and discussing its influence on larval growth, cocoon productivity and resistance to disease in silkworm and possible opportunities relating to the broad and fascinating area of nanomaterials in sericulture.

UPTAKE MECHANISM OF NANOPARTICLES

The process of uptake typically entails the passage of nutrients from the soil toward the root surface, the transport of ions via the membranes of root surface cells, the radial transport of ions into the root xylem vessels, the transport in the xylem and the distribution of ions in the aboveground parts of the plant [Bowling, 1976]. Recent studies have focused on determining the total nutrient uptake over time, as well as the nutrient uptake of a particular root and its growth rate.

After penetrating into the plant nanoparticles can follow two different paths to move through tissues *i.e*., apoplast or symplast. Apoplastic transport involve movement of water and substances through the extracellular spaces, xylem vessels and cell walls of adjacent cells. This process takes place outside the plasma membrane (Sattelmacher, 2001). In symplastic movement transportation takes place through the specialized structure called plasmodesmata (Roberts and Oparka 2003). Radial movement of nanoparticles within plant tissues was mainly triggered by apoplastic pathway which assists nanomaterials to reach the root central cylinder and the vascular tissues (Larue *et al*. 2012; Zhao *et al*. 2012; Sun *et al*. 2014). After reaching the central cylinder, nanoparticles move upward to the aerial part through xylem (Cifuentes *et al*., 2010; Wang *et al*., 2013; Sun *et al*., 2014).

In case of foliar applications, nanoparticles cross cuticular barrier by lipophilic or hydrophilic pathway (Schonherr 2002). In lipophilic pathway diffusion takes place through cuticular waxes, whereas in hydrophilic pathway dispersion through polar aqueous pores of cuticle or stomata takes place (Eichert *et al*., 2008; Eichert and Goldbach, 2008).

APPLICATION OF NANOPARTICLES

Fig 1. Application of nanoparticles in different fields

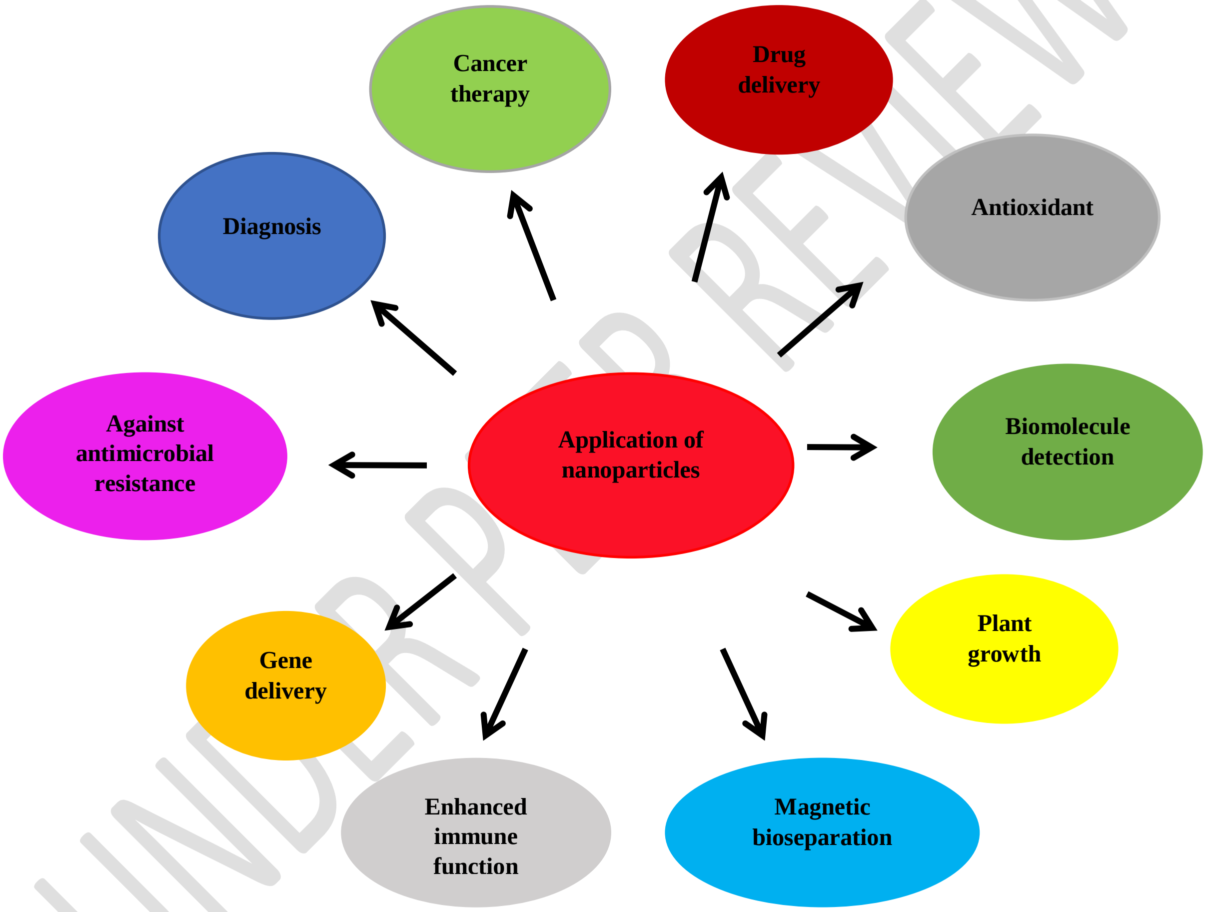
Cancer therapy- Nanoparticles specifically target cancer cells, where they undergo either endocytosis or phagocytosis to eliminate the diseased cells.

Drug delivery -Nanoparticles enclosing drugs enhance the stability and solubility of the medication, facilitating targeted delivery to particular tissues or cells.

Antioxidant - Certain oxide nanoparticles exhibit properties similar to antioxidant molecules as a result of their inherent physicochemical characteristics.

Biomolecule detection -Nanoparticles adhere to biomolecules on their surface, enabling the detection of these biomolecules through bio-tagging or labeling techniques.

Plant growth -Nanoparticles can serve as nano-fertilizers by encapsulating fertilizers, leading to improved growth, yield and quality of crops due to positive morphological and biological effects.



Magnetic bio-seperation - Magnetic nanoparticles selectively absorb the desired product onto their surface, facilitating its separation from the solution.

Enhanced immune function -Nanoparticles have the capability to trigger an immune response by functioning as adjuvants.

Gene delivery-Nanoparticles offer an alternative to vectors in the transfer of genes within the field of genetic engineering.

Against antimicrobial resistance- In combating antimicrobial resistance, nanoparticles surpass barriers due to their unique physio-chemical properties. These properties empower nanoparticles to employ diverse bactericidal pathways, thus achieving antimicrobial activity through novel means.

Diagnosis -Combining diagnostic and therapeutic agents within a single nanoparticle formulation.

EFFECT OF NANOPARTICLES ON MULBERRY

Nanoparticles as fertilizer for mulberry propagation

Mulberry is a high biomass producing, fast-growing, perennial, woody plant belonging to the genus Morus under the family Moraceae. Lu *et al*. (2003) suggested that, a proper nutrient management is required for appropriate root establishment, growth and leaf production. Hansch and Mendel (2009) stated that micronutrients like iron, copper, zinc, manganese are required in very low amount, but presence of correct balance of these elements is essential for growth and quality leaf production. Geetha *et al*. (2016) suggested that in case of multi micronutrient deficiency in Mulberry, yield can be reduced even up to fifty percent. To address the deficiency, application of chemical and chelated fertilizers needs to be adopted. Chelated fertilizers are quite costly and often applied for high value crops. Nanoparticles helps in seed germination, degradation of pesticide residue and improves soil quality (Ghrair *et al*., 2010; El-Temsah *et al*., 2014).

Nithya *et al*. (2018) recorded significantly highest shoot height (96.63 cm), number of branches per plant (8.47), number of leaves per shoot (18.60), number of leaves per plant (157.15), leaf area (96.90 cm^2) and leaf yield (0.46 kg /plant) in the foliar application of nano Zinc Oxide at 20 ppm in V-1 mulberry variety. They also reported that nano zinc fertilizer treatment to be cost effective with higher B:C ratio (2.93) and highest net returns per hectare as compared to $ZnSO_4$ fertilizer. Das and Mandal (2020) reported that nano-silver solution act as effective preservatives and enhance the activity of enzymatic and non-enzymatic antioxidants thereby reducing the harmful effect of accumulated free radicals and reactive oxygen species (ROS). Prevention of ROS generation helps in preventing plastid membrane peroxidation and thus maintaining chlorophyll content, extending the shelf life. They also reported that, the total chlorophyll, total protein, total sugar, reducing sugar, proline, total phenol, ascorbic acid, carotenoid and flavonoid contents were maximum after 7 days of leaf preservation in biosynthesized silver nanoparticles.

Soil and foliar application of iron oxide nanoparticles and EDTA functionalized iron oxide nanoparticles on mulberry improves the overall growth parameters. The application of iron oxide nanoparticles @ 10 mg/kg in soil significantly improved sprouting percentage

(82%), number of leaves (52.73% improved over control), plant biomass (37.20% and 90.24% increase of shoot and root biomass over control, respectively), root attributes (34% increment for root length) and also shortened the first leaf appearance period in mulberry (Haydar *et al*., 2021). Similarly, growth parameters, including shoot height, number of branches per plant, number of leaves per plant and total leaf area recorded in mulberry plant raised with foliar application of nano nitrogen fertilizer (Pooja *et al*., 2022). Also, noted that increased maximum leaf yield and improved quality parameters, such as total carbohydrates, crude protein, crude fibre, chlorophyll content and leaf elemental compositions, with foliar application of nano nitrogen fertilizer.

EFFECT OF NANOPARTICLES ON LARVAE

Effect on larval Feed efficacy

The feeding efficiency of silkworm larvae is important as it accounts for their growth rate and development. Low concentrations of nanoparticles (NPs) enhance larval body growth and feeding efficiency. Silkworm larvae fed with TiO_2 NPs (5 or 10 mg/L) improved the ingestion and digestibility of mulberry leaves, which significantly accelerated their body weight gain [Li *et al*., 2016].

Prabhu *et al*., (2012) recorded the superior feed efficacy *viz*, food consumption (gm) food utilization (gm) approximate digestibility (%) food consumption index (%) and Co-efficient of food utilization (%) in silkworm fed with MR2 mulberry leaves treated with 25 % silver nanoparticles solution. Similarly feeding low concentration of TiO_2 NPs showed significantly improved the amount of ingested food (g/larvae), the amount of digestion (g/larvae) and percentage of ingested food (%) in silkworms [Zhang *et al*. 2014; Li *et al*. 2016]. Riboflavin NPs fortified mulberry leaves enhanced the feed efficacy specially with higher production rate (mg/day) and metabolism rate (mg/day) in silkworm (Kamala and Karthikeyan 2019).

Effect on larval growth parameters

During the larval stages, silkworms feed on mulberry leaves which have all the required nutrients needed for their growth and development. The fat body of the silkworm is responsible for the storage, utilization and transfer of the nutrients required for the growth and development of the silkworm larvae. Mulberry leaves treated with silver nanoparticles or along with spirulina led to significant increase in length and weight of fifth-instar silkworm larvae (Thangapandiyan and Dharanipriya, 2019; Meng *et al*., 2017). Similarly, Prabhu *et al.* (2011) reported the increased length, width and weight of 3rd, 4th and 5th instar larvae when fed with silver nanoparticles. Pandiarajan *et al*. (2016) reported that the exposure of silkworm larvae to Ag NPs (1 ppm) improved the larval growth rate and the cocoon weight. Tian *et al*. (2016) focused on studying the impact of TiO_2 NPs (5 mg/L) on the nutrient metabolism of the silkworm fat body. The treating mulberry leaves with TiO_2 NPs activated the insulin signalling pathway of the silkworm by enhancing the metabolism of carbohydrates, proteins and fat when compared to the control group.

Nithya (2018) found that larval weight and effective rate of rearing (ERR) increased after feeding with nano zinc oxide treated mulberry leaves. Also found that reduced Moulting duration (h) and 5th instar larval duration (days) with the same. Further, Pramila *et al*., (2019) reported that full grown larval weight (g/10), 5th instar larval duration (h), total larval duration (h) and ERR (%) significantly increased in silkworm fed with nano zinc along with nano copper. Pooja *et al*. (2022) reported that significant improvement in the larval traits of silkworm fed with mulberry leaves with foliar application of nitrogen nano-fertilizer.

Influence of nanoparticles on cocoon traits

Patil *et al*. (2016) stated that the silkworms fed with mulberry leaves treated with gold nanoparticles (300 ppm) were significantly superior in cocoon and reeling traits. Similarly, silkworms fed with nanoparticles of riboflavin treated mulberry leaves showed significantly highest cocoon weight, shell weight, pupa weight and shell ratio (Kamala and Karthikeyan, (2019). They also found that nanoparticles of riboflavin enhanced the denier, silk filament weight (g) and filament length(m). Further, silver nanoparticles or along with spirulina improved the cocoon weight (g), silk filament weight (g), sericin content (%) and fibroin content (%) (Thangapandiyan and Dharanipriya, 2019). Similarly, Ag NPs, TiO_2 and Nano Zn along with CuNPs enhanced the economic traits of cocoons [Prabhu *et al*., 2011; Zhang *et al*., 2014; Li *et al*., 2016; Charya *et al*. 2019; Pramila *et al*., 2019].

According to Nithya (2018) adequate supply of zinc nanoparticles which accelerates the activity of enzymes and auxin metabolism in the plants that increased the larval parameters, thereby cocoon parameters of silkworms. Nano micronutrients might have stimulated the metabolic activities in silkworm resulting in better growth and development, resulting in production of good quality cocoons (Pramila *et al*., 2019).

By feeding silkworm with the carbon nanotube (CNT) obtained high strength silk fibre (SF) from silkworm. It proved that the stress, strain, conductivity and thermal stability of SF have been visibly enhanced, with the mechanical properties being comparable with those of super SF and even the spider silk fibre (Wang *et al*. 2014). The mechanical properties of the resulted silk were enhanced after feeding silkworms with MoO_2 nanoparticles (Liang *et al*., 2020).

Influence of nanoparticles on reproduction and fecundity

Feeding the silkworm with TiO_2 NPs is found to increase the metabolism of proteins and carbohydrates to meet the energy demand for growth and development of gonads. Silkworms exhibited the denser oocytes differentiation and formation in ovaries resulting high density of eggs, indicates that TiO_2 NPs not only increase the nutrient accumulation and transformation during the reproductive development but also improve the oviposition ability in *B. mori* (Ni *et al*., 2015).

Influence of nanoparticles on resistance to silkworm diseases

Feeding TiO_2 NPs inhibits the proliferation of *Bm*NPV in silkworm larvae and improves larval survival rate and cocoon traits after *Bm*NPV infection (Xu *et al*., 2015; Zhao *et al*., 2020; Fometu *et al*. 2022). Das *et al*. (2013) found that silica nanoparticles (NP)-induced morphological transformation of *Bm*NPV polyhedra could reduce the infectivity of *Bm*NPV in silkworm larvae. Silver nanoparticle showed maximum zone of inhibition and lowest gut bacterial (*Bacilli sp*.) growth of larvae. (Li *et al*., 2013; Prabhu *et al*., 2012). Similarly, Silver nanoparticle of *P. Hornemannii* (100 µl) showed maximum zone of inhibition against *B. bassiana* (22.6 mm) and *M. anisopliae* (21.0 mm) (Ramamoorthy *et al*., 2019). By inhibiting reactive oxygen species (ROS), the Ag NPs activates the Toll-pathway in silkworm to boost humoral and cellular immunity against *S. aureus* infection (Rajasekhar reddy *et al*., 2017).

Thymoquinone-Encapsulated Chitosan Nanoparticles (Tq-Chs NPs) showed an inhibitory impact against pathogenic bacteria infecting *Bombyx mori* larvae, proved their effects as antioxidant and anti-inflammatory agents which could be improved by loading Tq on Chs nanoparticles (Hasan and Fahamy 2020). The biosynthesized silver and chitosan nanoparticles at low concentration showed significance in the prevention of silkworm pathogens (1ppm, 10ppm, and 100 ppm) (Pandiarajan *et al*., 2016; El-Adly 2022). Chitosan at different concentrations showed antibacterial activity against the bacterial pathogens (*Bacillus spp*., *Micrococcus spp*. and *Serratia spp*.) of tropical tasar silkworm under in vitro conditions. The low concentration (0.2%) of chitosan from silkworm and chitosan NPs was identified as a minimum inhibitory concentration (MIC) against bacterial pathogens (Priyadarshini *et al*., 2018; Madhusudhan *et al*., 2023).

These NPs have a large surface area to volume ratio which enhances the binding activity or saturation capacity of NPs which is predicted to increase binding to the microbe cell membrane and also destroy the cell wall structure [Ahmad *et al*., 2013].

Most studies showed positive upshot NPs on silkworms at their lower concentration. In addition, the exposure of diseased silkworms to some nanomaterials also exhibited some therapeutic properties. Applied nanoparticles also exhibited greater impact on several biochemical and antioxidant enzymes attributes. These nanoparticles might be an ideal substitution for the traditional fertilizer and will be helpful in fortification of plants with nutritional value.

CONCLUSION: -

Sericulture implies rearing of silkworm for production of silk and ultimately its usage for textile and garment. Time has come to diversify it to make sericulture more sustainable, lucrative and remunerative one. Modern nano-technological advancement has assumed greater importance in the development of sericulture and its diversification. The science of nanotechnology and its application particularly in the area of enhancing mulberry leaf production, improvement in feed efficacy, development of diseases resistant breed and feed and synthesis of high-quality silk may take sericulture into a new height. The introduction of nanomaterials through diet has been reported to improve quality, tissue repair and the overall survival rate of the silkworm. The present paper, therefore, is a comprehensive document where an attempt to accumulate different reports and research findings on thrust areas of sericulture like spraying effect of NPs on phenotypic characteristics of mulberry, showing enhancement in biomass and different growth attributes and its influence on silkworm have been discussed which will prove to be useful for further development of the silk industry in coming years.

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