Annals of Phytomedicine 15(1): 411-419, 2026

DOI: http://dx.doi.org/10.54085/ap.2026.15.1.36

Annals of Phytomedicine: An International Journal
http://www.ukaazpublications.com/publications/index.php

Print ISSN : 2278-9839 Online ISSN : 2393-9885

Reveiw Article : Open Access

Growth regulation and disease management in silkworms using *Psoralea corylifolia* L.: A review

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Article Info

Article history
Received 10 January 2026
Revised 14 February 2026
Accepted 15 February 2026
Published Online 30 June 2026

Keywords
Bakuchiol
Disease management
Economic traits
Juvenile hormone analogue
Phytojuvenoid
Psoralea corylifolia L.
Silkworm

Abstract

Juvenile hormone (JH) analogs are required for enhancing silk production in *Bombyx mori* (*Bm*), but the developmentally toxic and environmentally damaging chemical juvenoids methoprene and fenoxycarb are not suitable. This review highlights the distinctiveness of bakuchiol from *Psoralea corylifolia* L. as a non-toxic phytojuvenoid, which possesses the potential to prolong larval life, delay metamorphosis, and induce silk gland function at 1.25 ppm in the fifth instar stage, leading to maximum cocoon weight, shell weight, and filament length. In addition, the phenolic compounds and tannins in *P. corylifolia* prevent viral *Bm*NPV grasserie disease in larvae by reducing larval mortality. Therefore, *P. corylifolia* is a practical and eco-friendly method for achieving sustainable sericulture productivity.

1. Introduction

The production of juvenile hormone (JH) by an endocrine gland promotes the regulation of larval growth, moulting, transformation and silk production in the mulberry silkworm, *B. mori.* The JH is transported by the hemolymph and stabilized by a special 30 kDa juvenile hormone binding protein (JHBP), which protects the JH against enzyme action and ensures that it reaches its target organs or tissues as the fat body and silk gland. This regulatory system plays an important role in the regulation of the larval stage, food consumption, and the activity of the silk glands on the *B. mori* (Suzuki *et al.,* 2011).

Juvenoids are substances that imitate the juvenile hormone and thus are very commonly used to control insects. The 50-year-old history of insect endocrinology defined two central hormones: brain neuropeptides (*e.g*., activation hormone) that control the cycle of development and corpora allata-derived JH that suppresses morphogenesis to permit the development of larvae and adult reproduction. This prompted the identification of more than 4000 synthetic juvenoids such as terpenoid, aryl, and polycyclic mimics as eco-friendly third-generation pesticides to disrupt pests with the help of the juvenile features and stimulation of metabolism to boost silk production (Sláma *et al.,* 2013). In sericulture, R394, methoprene, and fenoxycarb are some of the synthetic juvenoids that have been discovered to delay the larval stage and disrupt the pests by preventing metamorphosis. These juvenoids affect the enzyme activities involved in oxidative metabolism that affected the growth of cocoon weight positively (Nair *et al.,* 2006). Synthetic juvenoids will stimulate the production of proteins in larval tissues resulting in a higher silk production (Mamatha *et al.,* 2008). Even though, synthetic juvenoids are unsafe for longer term use because of their toxicity and developmental malformations in silkworms.

Currently, attention of researchers has changed to the impact of phytojuvenoids of plants as they are regarded to be safe and environmental friendly compounds as substitutes of the synthetic juvenoids. It was discovered that the phytojuvenoids applied to the silkworms through plants as medicine enhances the cocoon shell thickness, shell ratio, and protein content in the larval growth without causing any adverse effects to the health of the silkworms and the natural environment (Srivastava and Upadhyay, 2013). Of the phytochemical-rich vegetation, *P. corylifolia* has been studied especially due to its bakuchiol bearing juvenile hormone analog that has tremendous potential in sericulture. *P. corylifolia* showed a noteworthy enhancement of silkworm grainage characteristics and lesser infection rates of *Bm*NPV when provided to either larvae or moths (Shubha *et al.,* 2010). *P. corylifolia* was found to have high bioefficacy against polyphagous pests that pose a threat to sericulture, and also served as phagostimulants to stimulate feeding and growth in *B. mori*. Leaf/seed extracts deterred uzi fly and other predatory insects, and increased larval appetite in the presence of mulberry, and therefore enhanced weight gain and cocoon statistics (Bhatt, 2013).

The earliest experiment that established the corpora allata as the juvenile hormone (JH)-producing gland in the insects was done in

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Rhodnius wherein JH has been shown to suppress premature metamorphosis actively. Its activity is based on the developmental stage, whereby it is high during the nymphal development, reduces during the pre-metamorphic stage and re-emerges in the adult stage (Wigglesworth, 1934). These trends were subsequently confirmed in hemi and holometabolous insects such as silkworms (Wigglesworth, 2015) and formed the basis of the role of JH in growth regulation. This was further improved by study of *Cecropia* silkworms, which verified JH dynamics and permitted its extraction, concepts still used in contemporary sericulture design such as phytojuvenoid use of *P. corylifolia* to achieve a safe larval growth and increase silk production without interfering with natural endocrine regulators. The comparison between synthetic juvenoids and phytojuvenoids is given in Table 1.

Table 1: Illustrates the comparison of synthetic juvenoids with phytojuvenoids

Parameter	Synthetic juvenoids	Bakuchiol (*P. corylifolia*)	References
Source	Chemical synthesis	Plant-derived (seeds)	Suzuki *et al.,* 2011; Nair *et al.,* 2006; Nair *et al.,* 2003; Nizam *et al.,* 2023
Efficacy	Cocoon/shell weight ↑ 15-25%	Comparable ↑ 18-24% at 1.25 ppm	Nair *et al.,* 2006; Mamatha *et al.,* 2008; Nair *et al.,* 2003; Nair *et al.,* 2010
Optimal dose	5-10 ppm	1.25 ppm (48 h V instar)	Mamatha *et al.,* 2008; Nair *et al.,* 2010
Toxicity	Developmental defects	Non-toxic, biodegradable	Suzuki *et al.,* 2011; Nizam *et al.,* 2023; Chopra *et al.,* 2023
Environment	Persistent pollutants	Eco-friendly degradation	Suzuki *et al.,* 2011; Alam *et al.,* 2018
Application	Topical, regulatory limits	Mulberry leaf treatment	Mamatha *et al.,* 2008; Srivastava and Upadhyay, 2015
Cost	High production cost	Low (local sourcing)	Alam *et al.,* 2018
Disease control	None	*Bm*NPV ↓ 60-80%	Mahalingam *et al.,* 2010; Manimegalai *et al.,* 2010
Stability	Weeks persistence	Fresh extraction preferred	Khuranna *et al.,* 2020
Regulation	Country restrictions	Botanical approval	Alam *et al.,* 2018

2. *Psoralea corylifolia* L.

Psoralea corylifolia L. is an ancient medicinal herb within the Fabaceae family that is highly growing in areas like India, China, Pakistan and some parts of Africa. It is a straight herb, which is between 30-180 cm in height and grows better in warm areas and well-drained soils. The plant has broader, elliptic, dented margin leaves and small purple blue flowers in form of raceme. It is a small, ovoid, oblong pod which is usually black or chocolate in colour with only one long and dark brown seed. Its seeds are oily, not starchy, and smooth and compressed on the surface. The plant has several vernacular names in different parts including babchi, bawachi, bavanchi, and bakuchi (Alam *et al.,* 2018).

The *P. corylifolia* (babchi), which has been widely used in ayurveda medicine and chinese medicine as a curative of the skin such as vitiligo and psoriasis, has antimicrobial, antioxidant and anti-inflammatory effects and side effects that are minimal and far better than the synthetics (Chopra *et al.,* 2023). The seeds and fruits of it are high in coumarins such as bakuchiol, which are used traditionally as blood purifiers and wound healers, and new preparations are under investigation. This safety profile makes *P. corylifolia* extracts a potentially promising ecological effective juvenoids to modulate silkworm growth and prevent diseases in silkworm rearing (Khushboo *et al.,* 2010). *P. corylifolia* boasts of a wide range of medicinal activities including antibacterial, antiviral, antioxidant, and anti-inflammatory activities mainly due to its coumarins (psoralen, isopsoralen) and flavonoids present in the seeds and fruits. Such bioactivities are less larval mortality (19.23% grasserie at 1000 ppm chloroform extract) and improved cocoon shell ratios (18.75%) have direct application in silkworm rearing (Munir Anwar *et al.,* 2011). Bioactive isoflavones such as novel corylinin (7,4'-dihydroxy-3'-[(E)-3,7-dimethyl-2,6-octadienyl]isoflavone), isopsoralen, psoralen and neobavaisoflavone are present in dried fruits of *P. corylifolia* and this has been confirmed using spectroscopy. These products, particularly, prenylated coumarins, form the basis of the juvenoid-like action of the plant (Ruan *et al.,* 2007).

P. corylifolia is a useful plant that is known to contain a high phytochemical profile which comprises of coumarins, flavonoids, monoterpene phenols, benzofurans, glycosides, lipids, fatty acids, and volatile oils. The compounds do not only serve its medicinal purposes but also have important ecology and agricultural applications especially in pest control and growth regulation in insects. *P. corylifolia* seeds are also particularly highly concentrated with bioactive compounds including psoralen, isopsoralen, bakuchiol, and other flavonoids that have been examined as having a plant defense function or potential growth regulator or biopesticide (Chen *et al.,* 2023).

The seeds and fruits are the main sources of coumarins, flavonoids, and meroterpenes, and they aid the various biological functions. These bioactive compounds are indicative of their possible application in agriculture, including natural pest control and growth regulation (Zhang *et al.,* 2016). This botanical and chemical diversity positions *P. corylifolia* as a promising resource for both sustainable agriculture and further scientific exploration.

3. Extraction of bioactive compounds from *P. corylifolia*

3.1 Primary ethanol extraction of phytojuvenoid trials feeding

The most applied procedure of bakuchiol extraction that can be used in the examination of silkworm growth entails starting the procedure by washing dried *P. corylifolia* seeds with 80% ethanol before boiling at 95°C to acquire PCE extract. This extract is filtered, concentrated through rotary evaporation and stored to be used in mulberry leaf dipping procedures applied on fifth-instar larva in the treatment (Lim *et al.,* 2008).

To achieve full bioactive recovery in growth regulation as well as antiviral applications, exhaustive and stepwise extraction of dried seeds are done with the use of petroleum ether (defatting), benzene, ethyl acetate, acetone and also with methanol. This is a polarity gradient method that maximises coumarins, flavonoids, and meroterpenes recovery. Following the extraction step, purification is done using HPLC, LC/MS, FT-IR, and NMR spectroscopy to meet the criteria of analysis needed in conducting determinate sericultural dose-response experiments (Ahandani *et al.,* 2013).

3.2 Psoralen isolation protocol on *Bm*NPV control

Psoralen that has been defatted with 1 kg of petroleum ether at room temperature and then subjected to soxhlet extraction with methanol. Crude extract is subjected to silica gel column chromatography, benzene:chloroform (20:80 v/v) and the yellow needle-shaped crystals of psoralen were obtained (11.2 mg). UV spectroscopy, FT-IR and HPTLC densitometry at 254 nm against standards confirm identity which is necessary when 800 ppm antiviral leaf treatments are needed (Rajput *et al.,* 2008).

3.3 Bakuchiol growth regulation

Topical applications at 1.25 ppm were investigated on four extraction methods:

Maceration: Solvent (1:7 w/v) Seed powder (10 g) 7 days x 2 cycles.

Reflux: Boiling 40 min x 3 cycles

Soxhlet : Coarse powder, hot solvent (1:12) 6 h

Ultrasonic: 220 V/550 W for 45 min

The solvents to be used were ethanol, methanol, acetone, petroleum ether, and dichloromethane. Rotary evaporated extracts were determined using HPLC (C18 column, acetonitrile: water gradient, 262 nm detection) with ultrasonic ethanol extraction being the best way to extract bakuchiol. The efficiency of extraction of sericultural formulations is influenced by the granularity of seed powder, solvent:sample ratio, temperature (high increases solubility), extraction time and the addition of acetic acid. These variables are directly verified in single-factor optimization, and they are crucial in determining phytojuvenoid and coumarin recovery rates that are essential when using the field (Khuranna *et al.,* 2020).

3.4 Ultrasonic ethanol superiority

Ultrasonic-assisted ethanol extraction has the highest recovery of bakuchiol, psoralen and bavachin, than both cold maceration and decoction, therefore, it is the most optimal technique to use in large-scale sericulture formulations because of its speed and coumarin selectivity (Qin *et al.,* 2023).

3.5 Comparative species validation

Ethanol extraction (same family) *P. drupacea*: Soxhlet extraction results are 5.80% dry extract with 72.34% furocoumarins (1.25 mm grind optimum) and heated maceration results 5.66% yield with 57.23% furocoumarins. Both of them are effective in isolating bakuchiol and psoralen and isopsoralen, which have reported antiviral effects applicable in the treatment of grasserie diseases (Baisalova *et al.,* 2024). Extraction methods of *P. corylifolia* for sericulture application are given in Table 2.

Table 2: Illustrates the various extraction methods of *P. corylifolia* for sericulture application

Purpose	Method	Target compound	Citation
Growth regulation	Ethanol (80%, 95°C)	Bakuchiol (1.25 ppm)	Lim *et al.,* 2008
Growth regulation	Ultrasonic ethanol	Bakuchiol	Khuranna *et al.,* 2020; Qin *et al.,* 2023
Disease control	Soxhlet methanol	Psoralen (800 ppm)	Rajput *et al.,* 2008
Disease control	Soxhlet ethanol	Furocoumarins	Baisalova *et al.,* 2024

4. Chemical compounds in *P. corylifolia*

The preliminary screening of *P. corylifolia* demonstrates the important physicochemical characteristics, including moisture content and ash values, extractive yield, and pH and qualitative analysis of alkaloids, flavonoids, coumarins, and terpenoids in the seed/fruits by using the standard tests (*e.g.*, Dragendorff, Shinoda). These indicators validate high loading of bioactive, especially, JH-mimetic bakuchiol, to maintain the same level of consistency in the extract during silkworm feeding experiments. Making use of standardized profiles is helpful within the context of reliable application in growth prolongation and disease reduction, improving cocoon quality in sericulture (Pandey *et al.,* 2013). The chemical compounds found in *P. corylifolia* are given in Table 3.

Table 3: Chemical compounds in *P. corylifolia*

Compound name	Chemical nature	Plant part	Relevance
Bakuchiol	Meroterpene	Seeds/fruit	Potent juvenile hormone analogue; enhances larval growth, silk gland activity, cocoon weight, and filament length
Psoralen	Furanocoumarin	Whole plant/root	Antiviral activity against *Bm*NPV
Isopsoralen	Furanocoumarin	Whole plant	Antiviral; supports larval survival
Bakuchicin	Coumarin	Seeds	Antiviral; inhibits viral replication
Bavacoumestan C	Flavonoid	Fruit	Antiviral properties; enhances host defense

Bavachin	Flavonoid	Seeds/fruit	Supports silk gland function; antioxidant effects
Corylifol A/B/C/D/E	Flavonoid	Seeds/fruit	Supports protein metabolism; potential growth-promoting effect
Psoralidin	Coumarin	Whole plant/seeds	Antiviral against *Bm*NPV; improves larval health
Hydroxy bakuchiol	Meroterpene	Seeds	Related to bakuchiol; potential juvenile hormone activity
Bakuisoflavone	Flavone	Fruit	Antioxidant; contributes to general larval health
Bakuflavanone	Flavone	Fruit	Potential metabolic support
Bavachinin	Flavone	Seeds	Antioxidant and general phytochemical activity
Bavachalcone	Chalcone	Seeds	Antioxidant; some evidence for metabolic modulation
Corylifolinin	Chalcone	Seeds	General bioactive flavonoid
Corylifols	Prenyl flavonoid	Seeds	Antioxidant properties
Coryfolin	Flavonoid	Whole plant	General bioactive support
Corylin	Flavonoid	Whole plant	General bioactive support
Corylinin	Isoflavonoid	Fruit	Antioxidant effects
Dadzin	Isoflavonoid	Fruit	Antioxidant; general health support
Dadzein	Isoflavone	Fruit	Phytoestrogen; antioxidant
Dihydroxy coumestan	Essential oil	Seeds	Limited data; possible antioxidant effects
Genistein	Isoflavone	Fruit	Antioxidant; potential metabolic support
Hydroxypsoralenol A/B	Flavonoid	Fruit	Antiviral potential; limited data
Isobavachalcone	Chalcone	Seeds	General phytochemical activity
Isobavachin	Flavonoid	Seeds/fruit	Antioxidant and growth-supportive potential
Psoracorylifol D	Flavonoid	Seeds	Antioxidant and general bioactivity
Psoracoumestan	Coumestans	Seeds	Antiviral and antioxidant activity
Xanthoangelol	Chalcone	Seeds	General antioxidant and metabolic support

Source: Alam *et al*., 2018

5. Bakuchiol

Bakuchiol is a meroterpene derivative that is mainly produced in the seeds and leaves of *P. corylifolia* and has the structure of a chiral tetraalkylated quaternary center. It is also found in other flora like *Ulmus davidiana* var. *japonica* and *Piper longum* and this highlights its extensive distribution and importance. It is known that bakuchiol exhibits a wide range of biological effects, such as antimicrobial, anti-inflammatory and antioxidant, which is a promising compound to be used in pharmacological, ecological and agricultural environments (Nizam *et al.,* 2023). Bakuchiol has a broad spectrum of bioactivity, making it an attractive target in the study of sustainable farming and a natural product with potential to adopt environmentally-friendly usages and further scientific study (Chopra *et al.,* 2023). It is very important in the defence of plants and has been promising in agricultural conversion, where it affects insect development and growth. Bakuchiol is also included in the list of constituents that helps the plant to survive in the ecologically unstable environment and gives the plant multiple applications in both traditional and modern settings (Jige, 2022).

Phytochemical analyses of leaf extracts of *P. corylifolia* identified important bioactives such as flavonoids, alkaloids, tannins and coumarins using GC-MS and initial tests involving preliminary assays in relation to phytochemical analysis. Leaves of *P. corylifolia* exhibited strong bakuchiol precursors and phenolics, which were in favor of JH-like activity (Kumar *et al.,* 2022). Bakuchiol is also an analogue of juvenile hormone that modulates moulting and metamorphosis in insects. On silkworm, it improves the production of silk by boosting the cocoon weight, cocoon shell weight and filament length. It is dose and time-dependent and bivoltine hybrids respond more than the multivoltine hybrids (Nair *et al.,* 2003). The chemical structure of bakuchiol is given in Figure 1.

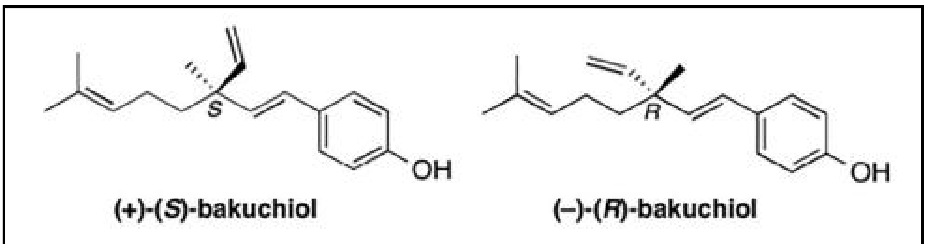

Figure 1: Chemical structure of bakuchiol (Source: Nizam *et al*., 2023).

6. Role of *P. corylifolia* in juvenile hormone activity

P. corylifolia is a useful botanical source of phytojuvenoid compounds that exhibit strong juvenile hormone (JH)-like activities in *B. mori* larvae (Nair *et al.,* 2003). Its major active ingredient, bakuchiol, is a potent natural JH analogue that mimics endogenous hormonal actions, which simultaneously maintains juvenile phenotypes, delays metamorphosis and prolongs the feeding period to achieve optimal development. The main mechanism by which bakuchiol promotes growth is endocrine interference, inhibition of ecdysone biosynthesis to prolong silk-productive larval stages where increased feeding, the accumulation of proteins, and further growth of silk glands occur. The larval instar extension by bakuchiol increases the level of feed intake and nutrient uptake which directly increases silk protein production. It is proven that the phytojuvenoid, *P. corylifolia* introduced orally or topically causes significant changes in protein metabolism and hypertrophy of silk glands providing better cocoon parameters and changes in the developmental patterns (Srivastava and Upadhyay, 2015).

The optimum 30% phytojuvenoids concentrations maximize fat body and silk gland protein concentrations, and 40% concentrations induce metabolic stress and decreased synthesis. Bakuchiol, a powerful JH analog of *P. corylifolia* greatly increases the weight of larvae, the yield of cocoons (maximum of 18.75 shell ratio at 1000 ppm), and lowers the death rate of grasseries to 19.23% (Mar *et al.,* 2001). Probiotics *P. corylifolia* extracts significantly improved the economic parameters of *B. mori* larvae, especially in PM silkworm races, the larval weight of 2.98 g, cocoon weight of 1.45 g, and shell weight are improved when compared to the control group (Mohanraj *et al.,* 2016). Such fortification through dipping of mulberry leaves enhanced digestion, immunity and silk production.

P. corylifolia extracts as mulberry leaf supplements are recently used in silkworm rearing to enhance the output of cocoon in unfavorable climatic conditions, which makes them become sustainable growth modulators (Ghorpade *et al.,* 2022). This makes *P. corylifolia* a competitive biotech resource in terms of the production of hormonally-optimized silk. The effect of *P.corylifolia* treatment on larval parameters of silkworm, *B. mori* is given in Table 4a and the effect of *P. corylifolia* treatment on cocoon parameters of silkworm, *B. mori* is given in Table 4b.

Table 4a: Effect of treatment on larval parameters of silkworm, *B. mori*

Treatment hour (V instar)	Concentration (ppm)	Larval weight (g)	Survival (%)
24	0.625	4.912	87.66
	1.25	4.915	86.77
	2.5	4.823	88.33
48	0.625	4.993	87.11
	1.25	5.131	89.44
	2.5	4.860	88.33
72	0.625	4.945	87.55
	1.25	4.781	87.22
	2.5	4.784	87.55
96	0.625	4.829	86.11
	1.25	4.719	87.00
	2.5	4.722	85.66
Control	–	4.64	86.44

Source: Nair *et al*., 2010

Table 4b: Effect of treatment on cocoon parameters of silkworm, *B. mori*

Treatment hour (V instar)	Cocoon yield/ 1,000 larvae (kg)	Cocoon weight (g)	Shell weight (g)	Shell percentage	Filament length (m)
24	1.645	2.028	0.418	20.63	941.09
	1.648	2.007	0.414	20.62	941.17
	1.616	2.008	0.416	20.71	908.96
48	1.671	2.049	0.422	20.59	959.13
	1.67	2.119	0.441	20.81	975.65
	1.591	2.012	0.413	20.55	949.73
72	1.601	2.021	0.416	20.59	924.49
	1.591	1.971	0.412	20.89	935.77
	1.593	1.947	0.400	20.57	885.31
96	1.578	1.973	0.405	20.57	904.70
	1.556	1.928	0.412	21.40	892.25
	1.587	1.933	0.399	20.66	850.34
Control	1.585	1.895	0.385	20.32	860.92

Source: Nair *et al*., 2010

7. Role of *P. corylifolia* in disease management

Besides its growth control effect, *P. corylifolia* contributes also great independent effect in the control of grasserie disease in the mulberry silkworm, *B. mori*. Grasserie is a virus disease brought about by the *B. mori* nuclear polyhedrosis virus (*Bm*NPV) and is among the most devastating viral infections in the survival and the yield of the cocoons. *P. corylifolia* botanical extracts have been widely tested in terms of antiviral activity against *Bm*NPV. The bioactive compounds such as coumarins (psoralen, isopsoralen), flavonoids, and meroterpenes (bakuchiol) which are responsible for the properties like antimicrobial, antioxidant, anti-inflammatory, and antipigmentary are present in *P. corylifolia* fruits and seeds (Yang *et al.*, 2025). They are also applicable for 80% reduction of the grasserie disease in silkworms at 800 ppm feeding.

Oral application of *P. corylifolia* extract or with TNAU seri dust reduced substantially the mortality caused by grasserie as percentages of 3.28 in the control group and 1.35 in treated batches (Mahalingam *et al.,* 2010). It was proposed that the antiviral activity was due to the presence of hydrolysable tannins and phenolic compounds, which caused the disaggregation of polyhedral bodies, and interference with the host cell recognition sites necessary to enable viral infection and replication. Field-level experiments also indicated that *P. corylifolia* extract when administered at the third instar stage was effective in reducing disease as well as enhancing the economic characteristics of larval weight, cocoon weight, shell weight, shell ratio, and overall cocoon yield.

The chemical profiling of *P. corylifolia* using GC-MS and found that the plant contained a number of bioactive compounds that are related to antiviral activity, including psoralen, phenol, eicosane, and undecane (Manimegalai *et al.,* 2009). Further research indicated that the hexane extract of *P. corylifolia* had a specific effect against grasserie disease. Almost 80% of the incidence of grasserie was reduced with hexane leaf extract at 800 ppm (Manimegalai *et al.,* 2010). This antiviral activity was highly explained by flavonoid like bakuchicin and bavacoumestan, which inhibit the viral multiplication and bind viral DNA, viral proteins and effect viral adhesion and enzyme activity.

The bioactive compounds like flavonoids and phenolics of *P. corylifolia* suppress the release of infective virions and promote the host defense responses of silkworm larvae (Thakur *et al.,* 2025). Taken together, the results demonstrate that *P. corylifolia* is a powerful, natural and economical botanical compound to manage grasserie disease with the aim of boosting larval survival and superior cocoon yielding without the use of artificial antiviral agents. The economic parameters of *P. corylifolia* treated cocoons is given in Table 5.

Table 5: Economic parameters of *P. corylifolia* treated cocoons

Treatments	Larval weight (g)		Cocoon weight (g)		Shell weight (g)		Shell ratio (%)	
	PM × CSR2	CSR2 × CSR4	PM × CSR2	CSR2 × CSR4	PM × CSR2	CSR2 × CSR4	PM × CSR2	CSR2 × CSR4
P. corylifolia	3.47	2.93	1.72	1.74	0.32	0.43	18.60	24.71
Treated control	2.49	2.40	1.43	1.51	0.26	0.32	18.18	21.20
Untreated control	3.30	3.20	1.70	1.72	0.29	0.40	17.06	23.20

Source: Manimegalai *et al*., 2010

8. Application

8.1 Juvenile activity

The discovery was that topical JH-I application to fifth-instar larvae enhances hemolymph prothoracicotropic hormone (PTTH) titers during early stages (through time-resolved fluoroimmunoassay), but decays by wandering stage; delays pigmentation of the spineret, an indicator of larval-pupal transition, by inhibition of tissue ecdysteroid response, not by blocking PTTH or increases in ecdysteroid (Mizoguchi *et al.,* 2001). The same PTTH stimulation is witnessed in the fourth-instar larvae and JH plays a role in late development as a major PTTH secretagogue. These interactions impart phytojuvenoid activity of *P. corylifolia* to control growth and prevent early moulting in sericulture.

The presence of juvenile-like hormonal and biochemical effects of the *P. corylifolia*-derived bakuchiol has been established with great effort. Topical application of phytojuvenoid treatments of *P. corylifolia* during fifth instar were effective to extend the larval instar, delay pupation, and induce soft-bodied gland functionality (Nair *et al.,* 2003). Subsequent studies confirmed bakuchiol as an efficient JH mimic that regulates developmental stages and improves silkworm properties that have commercial importance. It is vital to appearance because bakuchiol depends on the time and concentration of use. Optimal results were obtained repeatedly when applications of 1.25 ppm were done 48 h after the fifth instar stage resulting in significant gains in cocoon weight, shell weight, and filament length in the treated larvae (Nair *et al.,* 2010). This prolonged larval stage allows more protein synthesis and growth of cells in the silk glands producing better quality silk. Median phytojuvenoid dosage increases haemolymph concentrations of free amino acids essential substrates in fibroin and sericin synthesis during cocoon spinning (Srivastava and Upadhyay, 2015). On the contrary, excessive dose or repeated doses compromise metabolic performance, which is typical of hormesis.

8.2 Disease management

P. corylifolia has become one of the potential botanicals in terms of disease management in silkworm rearing especially in controlling of grasserie disease caused by *Bombyx mori* Nuclear Polyhedrosis Virus (*Bm*NPV). Petroleum ether extracts of *P. corylifolia* that was applied as mulberry leaves in the third, fourth, and fifth instar stages not only improved larval growth, but also increased considerably cocoon weight, shell weight, and shell ratio, but also decreased the presence and severity of grasserie disease (Rajasekharagouda, 1991). Such discoveries gave *P. corylifolia* a dual status of a disease suppressive and productiveness-enhancing plant.

The aqueous extracts of leaf by dipping and shade drying mulberry leaves and feeding the larvae on the third to fifth instar stages led to a significant reduction in mortality of the larvae due to grasserie disease (Samuel Manohar Raj, 1994). This study indicated the appropriateness of aqueous extracts to be used in the field and verified the effectiveness of *P. corylifolia* in enhancing the larval survival in rearing environments prone to diseases. An application protocol using aqueous extracts of *P. corylifolia* at 800 ppm was standardized for fourth instar larvae via treated mulberry leaves. This led to strong reduction in grasserie-induced larval mortality and positive gains in larval weight, cocoon weight, shell weight, and filament weight, underscoring the economic benefits of the botanical treatment (Padma *et al.,* 2007).

The use of *P. corylifolia* extract with other common hygienic interventions like disinfection of rearing houses using TNAU seri dust (Mahalingam *et al.,* 2010). This experiment revealed that when *P. corylifolia* extract was orally administered after disinfection, larval deaths due to *Bm*NPV were reduced drastically and that there was significant improvement in cocoon productivity in both laboratory and field experiments. This synthesized methodology stressed on the strong applicability of *P. corylifolia* based remedies as a cost-efficient and environmentally friendly disease control method in sericulture. Simultaneously, cumulatively, these sequential studies clearly reveal that Mulberry leaf treatment in terms of concentration and instar stage of *P. corylifolia* will not only suppress the grasserie disease, but also enhance growth, survival and cocoon related economic characteristics of *B. mori*. The similarity of findings in various extraction procedures, concentrations and silkworm species demonstrate the possibility of using *P. corylifolia* as a stable, natural and sustainable botanical resource in the disease control in contemporary sericulture.

9. Future directions

The role of *P. corylifolia* and its bioactive compound bakuchiol in silkworm rearing in future studies should focus on optimization, mechanistic insights, field level testing, *etc.,* to ensure maximum yield of silk and resistance to diseases. Though, research has discovered 1.25 ppm used at 48 h fifth instar as an effective dosage, the dosage needs to be refined on other silkworm breeds and agro climatic conditions. Individual versus repeated application and graduated concentrations need to be evaluated in a detailed fashion to determine their impact on the larval development, gland metabolism, protein synthesis and amino acid consumption without causing negative physiological effects on them in high doses.

Increased focus should also be on the timing and delivery methods of its application such as using the mulberry leaf as fortification and as an ingredient of artificial diets to enhance its bioavailability and the practical viability of the field application. The molecular level future research needs to utilize molecular docking, transcriptomic and proteomic methods to understand how bakuchiol interacts with juvenile hormone-binding proteins and the subsequent effects on genes related to silk protein production, metabolism, and immunity. Combining the juvenile activity driven by bakuchiol with antiviral activity of *P. corylifolia* extracts in *Bm*NPV may result in more detailed management strategies that may increase both productivity and disease resistance. Lastly, standardization, quality control as well as sustainable implementation of *P. corylifolia* -based preparations in commercial sericulture requires large-scale field trials and the use of advanced analytical procedures like HPLC and LC-MS.

10. Conclusion

P. corylifolia, also known as babchi or bakuchi has become an important medicinal herb with considerable attention in the sericulture industry due to its varied phytochemical contents and established properties of enhancing silkworm growth, improving silk production and providing protection against diseases. The seeds of the plant contain a variety of bioactive compounds such as bakuchiol, psoralen, isopsoralen, a variety of flavonoids, phenolics, tannins, and coumestans. Despite their established popularity in historical and modern pharmacology and traditional medicine, recent studies have found these phytochemicals have an extraordinary level of success in enhancing both viability and productivity of the mulberry silkworm, *B. mori*.

Bakuchiol stands out of these active constituents due to its strong phytojuvenoid effect, which is believed to mimic the functions of natural juvenile hormone to control the larval development and growth. Topically applied or added to feed at the fifth instar stage with the most successful administration at about 48 h after the ecdysis delaying metamorphic change, an effective dosage is only about 1.25 ppm of bakuchiol. This growth extension enables increased periods of feeding and higher levels of nutrient absorption promoting high levels of protein incorporation in silk glands and increments in key commercial variables such as cocoon mass, shell mass, shell ratio percentage, and filament length indices. It is important to note that bivoltine strains have a higher response rate than multivoltine varieties, and it is evident that genotypic differentiation in sensitivity to treatment is pronounced.

Moderate concentrations of the *P. corylifolia* extracts (10-30% concentration) have been shown to raise the total protein levels in fat bodies and silk glands and at the same time improve haemolymph free amino acid profiles and promote the production of silk proteins and glandular performance. Conversely, high dosage near 40% concentration is dangerous since it can cause physiological strain, thus the need to carefully correct dosage during field testing is necessary.

P. corylifolia possesses significant antiviral effects against grasserie disease caused by the *Bm*NPV nuclear polyhedrosis virus besides its growth-boosting attributes. Its extracts, both orally and topically applied, significantly reduce the number of larvae that die owing to interruption of the process of viral growth. This cellular defense is mainly due to phenolic products and hydrolysable tannins which lead to aggregation of viral polyhedral structures and at the same time prevent the viral attachment to silkworm cellular receptors. Interestingly, leaf extracts with hexane as the solvent at the approximate concentration level of 800 ppm yield up to 80% reduction in the incidence of grasserie thus, demonstrating a significant potential to feasible disease management measures in silkworm culture in commercial settings.

However, a few limitations were found in these positive results even though the research was carried out. Most of the current studies are only limited to laboratory studies or small scale field tests without extensive large scale commercial studies in different agroclimatic conditions. In addition, there remains a case of data gaps on chronic effects of repeated phytojuvenoid regimens, future interactions of the treatment regimens on various mulberry strains, and the means of smooth integration into the usual silkworm rearing procedures.

Secondly, the attainment of reproducible extraction protocols and dependable quantification of pharmacologically active constituents has remained a major impediment to large scale commercial application.

To conclude, *P. corylifolia* is a valuable, eco-friendly and economically viable botanical resource to modern sericulture activity. By virtue of its distinct dual capability in enhancing growth parameters and silk yields at the same time reducing viral pathogens it offers some attractive possibilities of the more radical, sustainable production systems of silk, although it depends on dedicated future research to systematically address identified gaps in knowledge and enable large-scale commercial upholding.

Acknowledgements

The authors acknowledge the Department of Sericulture, Forest College and Research Institute, Tamil Nadu Agricultural University, Tamil Nadu, India.

Conflict of interest

The authors declare no conflicts of interest relevant to this article.

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Citation **T. Utchimahali, K.A. Murugesh, K. Senguttuvan, R. Shanmugam and S. Vellaikumar (2026). Growth regulation and disease management in silkworms using *Psoralea corylifolia* L.: A review. Ann. Phytomed., 15(1):411-419, http://dx.doi.org/10.54085/ap.2026.15.1.36.**