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Investigations on the reproductive efficiency and progeny fitness of *Tetrastichus howardi* as influenced by Multivoltine races of *Bombyx mori L.*

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***Abstract*— *Sericulture is a vital agro-based industry in India, contributing significantly to the economy and rural livelihoods, with approximately 9.5 million individuals, including a large number of women, employed in the sector. The cultivation of mulberry (*Morus alba*) is integral to silkworm rearing, which directly influences silk productivity and farmer income. However, the industry faces challenges from many pests.* Diaphania pulverulentalis *(leaf roller), one of the major pest which causes substantial crop loss. This study investigates the potential of Integrated Pest Management (IPM) strategies, focusing on biological control through parasitoids such as* Tetrastichus howardi*, to mitigate pest damage while safeguarding silkworms for pesticides. The reproductive performance of* T. howardi *was evaluated on different multivoltine silkworm races, including Pure Mysore, Nistari, HB6, BL6, MO6, and MCon. The findings indicate that certain silkworm races, such as Pure Mysore and Nistari, are more conducive to the mass multiplication of* T. howardi*, with* MO6 *exhibiting the highest progeny production. The results underscore the potential of utilizing multivoltine silkworms as alternate host for mass multiplication of T. howardi.*

***Keywords*— Sericulture, Integrated Pest Management (IPM), Biological Control, Tetrastichus howardi, Multivoltine Silkworms.**

I. INTRODUCTION

Sericulture, an agro-based industry deeply rooted in India, plays a significant role in the nation's economy and rural livelihoods. The industry provides employment to a substantial workforce, with 9.5 million people in rural & semi-urban areas, with a significant portion of these workers coming from economically background, also huge number of women work force engagement. India holds the position of the world's largest silk consumer and ranks as the second-largest silk producer. During the 2023-24 period, India's raw silk production reached 38,913 metric tons (MT), with the mulberry silk sector contributing 77% (29,892 MT) of this total, according to the Central Silk Board (CSB) Annual Report for 2023-24. The Central Silk Board aims to achieve sustainable growth in the silk industry, driven by rising domestic demand.

Mulberry (*Morus alba*), the sole food source for silkworms (*Bombyx mori* L.), is cultivated across approximately 2.6 lakh hectares to support silkworm rearing and silk production. As a perennial crop with a lifespan of 15-20 years, the yield and quality of mulberry leaves directly influence silk productivity and farmer profits. The quantity of leaves determines the silkworm rearing capacity, while leaf quality significantly impacts silkworm growth, development, and silk yield.

The production of quality mulberry leaves is influenced by several factors, including plant variety, spacing, and the management of water, nutrients, pests, and diseases. Both biotic and abiotic factors can affect the growth, yield, and quality of mulberry leaves by around 20-25%. As a perennial plant that produces high biomass, mulberry is susceptible to pest infestations that can diminish both the quantity and quality of its leaves. Key pests include the leaf roller (*Diaphania pulverulentalis* Hampson), Bihar hairy caterpillar (*Spilosoma oblique* Walker), pink mealybug (*Maconellicoccus hirsutus* Green), papaya mealybug (*Paracoccus marginatus*), and cutworm (*Spodoptera litura*). In Karnataka, studies have documented crop losses due to these pests, with leaf rollers causing 12–15% loss, pink mealybugs leading to 10-15% loss, and Bihar hairy caterpillars resulting in 25–30% loss. Additionally, losses of 17-18% have been attributed to thrips, 12.62% to whiteflies, 9.08% to jassids, and 8.24% to scale insects affecting mulberry crops (Manjunath et al, 2000, Govindaiah.et.al, 2005). Beyond causing qualitative damage, pest infestations can disrupt the biochemical composition of mulberry leaves, negatively impacting the growth and development of silkworms. The quality of mulberry leaves is essential for cocoon production since silkworms are particularly sensitive to variations in feed quality. Leaves with higher moisture levels, total sugars, and soluble carbohydrates are more attractive to silkworms. The nutritional profile of mulberry leaves significantly influences the growth and development of silkworm larvae, which subsequently affects both the quality and quantity of silk produced.

Diaphania pulverulentalis (Lepidoptera: Pyralidae), commonly known as the leaf roller, is a key pest affecting mulberry crops in South India (Manjunath et al, 2000). This infestation, most severe between September and November, leads to a 20% reduction in leaf yield. The larvae typically target the apical shoots of the mulberry plant, binding tender leaves together with a silk-like substance. This creates a protected environment where they can feed, initially consuming the tender apical leaves before progressing to more mature foliage.

Although chemical control is a common approach for managing leaf rollers on mulberry, the potential harm to silkworms from pesticide residues makes it undesirable. This highlights the need for alternative strategies. Biological control, a key component of Integrated Pest Management (IPM), offers a viable solution through the use of natural enemies like parasitoids, predators, and pathogens. Research suggests that parasitoids are the most effective, accounting for approximately 82% of pest suppression, followed by predators (17%), and pathogens (1%) Van Lenteren (1983).

Tetrastichus howardi stands out as a promising biological control agent due to its ability to parasitize a wide range of Lepidopteran insects (Baitha et al., 2004). Beyond pupal parasitism, it also exhibits host-feeding behavior, destroying Lepidopteran eggs and further suppressing pest populations. Its high reproductive capacity allows for rapid population increase in treated areas, enhancing its effectiveness. Recognizing these benefits, the Central Sericultural Research and Training Institute (CSRTI), Mysuru, has recommended the release of 50,000 *T. howardi* adults per acre for leaf roller control (Sakthivel N et al, 2019) & Dandin, S. B., & giridhar, k. (2014). A limiting factor, however, is the challenge of finding new suitable and efficient hosts for mass-rearing this parasitoid for commercial purposes. Current research is investigating the potential of Multivoltine silkworm Races of *Bombyx mori* L. pupae as an alternative host to facilitate the mass production of *T. howardi*.

II. MATERIAL & METHODS

Parasitoid description:

Tetrastichus howardi, a member of the Hymenoptera order, is a gregarious parasitoid that targets both larvae and pupae of various insect pests. Its broad host range makes it a valuable biological control agent against pests like sugarcane stem borers and mulberry leaf rollers (Gangadhar, B. et al, 2009). This parasitoid remains active throughout the year, with particularly high effectiveness during the winter. Adult *T. howardi* wasps are characterized by a 3mm-long, dark metallic blue thorax and a black abdomen with a slight metallic sheen. Completing its life cycle in approximately 17 days, *T. howardi* is considered an effective biological control option due to its high reproductive capacity and efficient parasitization of target pests.

Host & Parasitoid Culture

The study was carried out at the Department of Sericulture Science, University of Mysuru during May – August 2022. Silkworm *Bombyx mori* L. pupae of multivoltine pure races i.e., Pure Mysore, Nistari, HB6, BL6, MO6 and Mcon were procured from the CSR & TI, Mysuru and Department of Sericulture Science, Manasagangotri, Mysuru. The host culture was maintained at 25-28°C and 60-80% RH (Plate 1). The cultures of T. howardi were procured from the Pest Management Laboratory of the Central Sericultural Research and Training Institute (CSR & TI), Mysuru, and maintained on the pupae of Samia cynthia ricini (Eri silkworm pupae) at 25-28 °C and 60-80% RH in the laboratory. The adults of the parasitoid were fed on aqueous honey at 30%.

With regard to this experiment, on the reproductive performance of the parasitoid in relation to age of the host, gravid female parasitoid was allowed to parasitize on 4 day-old pupae of multivoltine pure races i.e., Pure Mysore, Nistari, HB6, BL6, MO6 & Mcon at a host – parasitoid ratio of 1: 1 in a test tube (15 x 1.5 cm), with ten replications for 2 days (48 hrs). under 25 ± 2 ºC, 60-80%, RH. Then *T.*

howardi females alone were removed from the test tube after the parasitism exposure period. Silkworm (*Bombyx mori* L.) pupae, once parasitized, were kept in test tubes until the emergence of the *Tetrastichus howardi* offspring. Subsequently, various data points were recorded, including the time taken for development, the number and sex ratio of the resulting parasitoid progeny, the lifespan of both parent and offspring females, and physical measurements (body length, head width, wingspan, abdominal dimensions) of the emerged adult parasitoids (Plate 2).

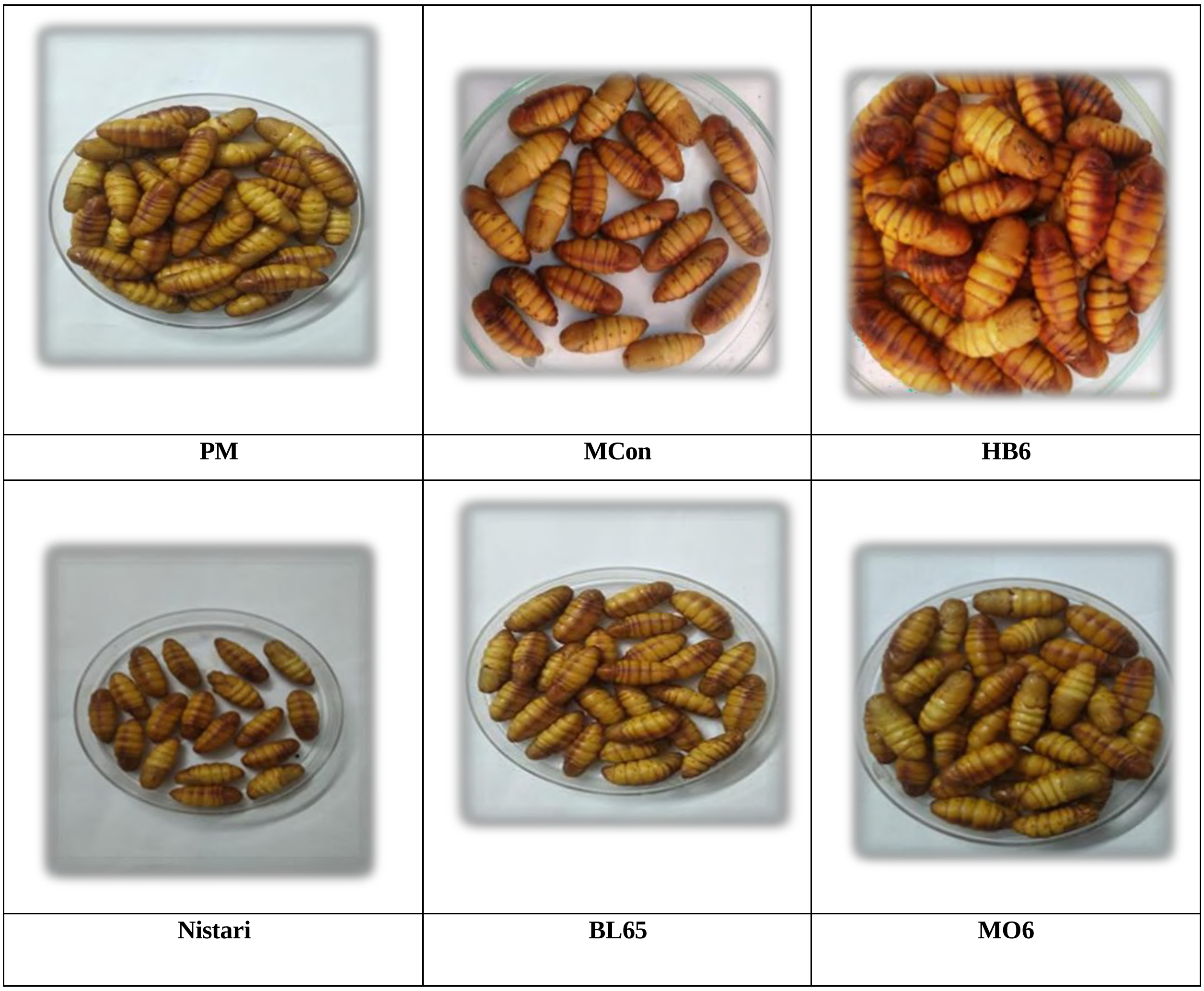


Plate 1: Pupae of Multivoltine pure races

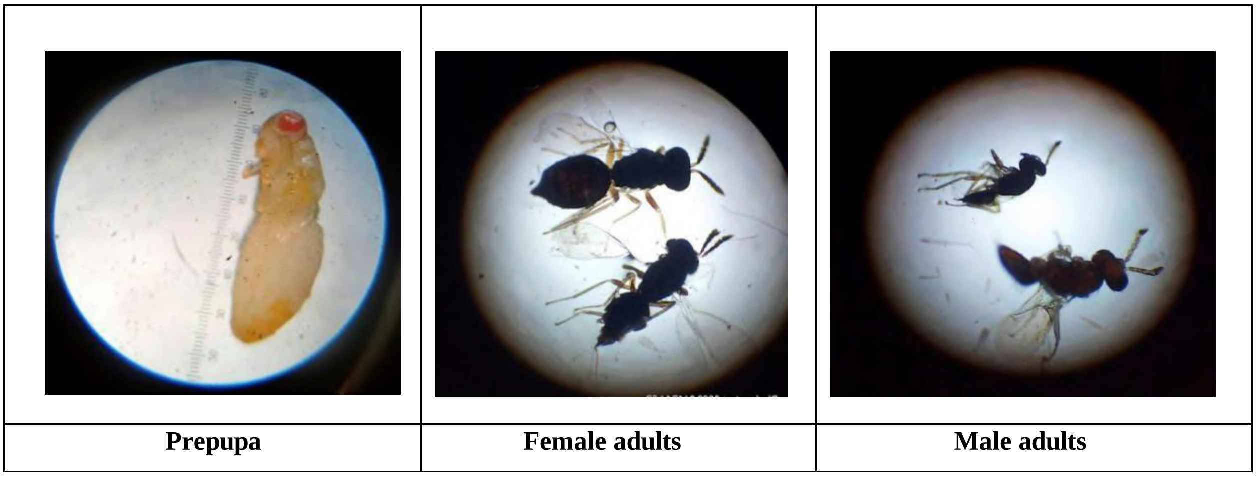


Plate 2: Microscopic views of Tetrastichus howardi progeny adults

III. RESULTS & DISCUSSIONS

Sankar et al.(2016) first reported the use of *Bombyx mori* L as a host for the mass multiplication of *T. howardi*, noting a higher emergence rate of *T. howardi* compared to another insect host. Later, Kishan et al. (2024) used bivoltine breeds of *Bombyx mori* L to assess host specificity and progeny production, with results indicating that CSR26 and CSR57 had higher progeny production.

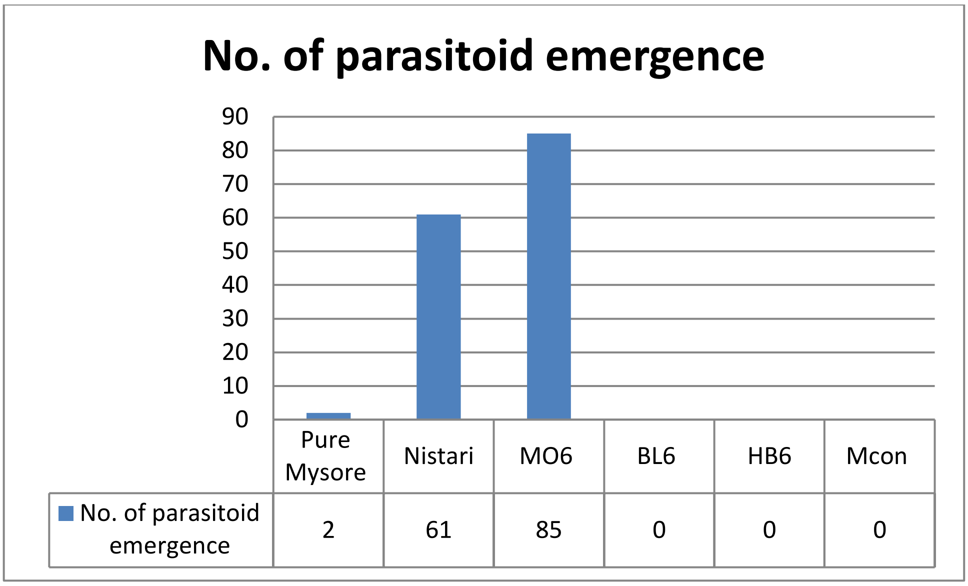
In the present study, several Multivoltine races—Pure Mysore, Nistari, HB6, BL6, MO6, and MCon—were selected to examine their suitability as hosts for the mass multiplication of *Tetrastichus howardii*. The results indicated that multivoltine races are suitable hosts for *T. howardii*, with Pure Mysore, Nistari, and MO6 showing positive outcomes, producing 2, 61, and 85 progeny, respectively. HB6, BL6, and MCon, however, did not produce any progeny. The male to female sex ratios were 17 ± 0.0 for Nistari and 20.25 ± 0.0 for MO6. Since Pure Mysore produced only 2 female progeny, the sex ratio for this race is 0. Previous studies reported total life cycles of 20 ± 1 days and 19 ± 1 days (Kfir et al., 1993). The present study observed similar results, with a life cycle of 19 ± 2 days (Table 1 & Graph 1).

Morphometric characteristics of *T. howardi* were recorded during the adult stage, and significant differences were found among the progeny from the different Multivoltine races. In males, the morphometric characteristics ranged from 1.47 ± 0.16 to 1.55 ± 0.03 in body length, 0.63 ± 0.07 to 0.66 ± 0.06 in body width, 1.03 ± 0.09 to 1.26 ± 0.06 in wingspan, 0.54 ± 0.07 to 0.68 ± 0.08 in head width, and 0.78 ± 0.08 L - 0.66 ± 0.06 W to 0.79 ± 0.03 L – 0.56 ± 0.10 W in abdomen size. In females, the morphometric characteristics ranged from 1.96 ± 0.0 to 2.17 ± 0.17 in body length, 0.71 ± 0.06 to 0.89 ± 0.0 in body width, 1.34 ± 0.12 to 1.59 ± 0.12 in wingspan, 0.7 ± 0.0 to 0.79 ± 0.02 in head width, and 0.83 ± 0.0 L – 0.81 ± 0.0 W to 1.12 ± 0.20 L – 0.84 ± 0.06 W in abdomen size.

Among the Multivoltine hosts, progeny from MO6 showed a larger body size in both males and females (body length, body width, wingspan, head width, and abdomen size) compared to progeny from other hosts, followed by Nistari and Pure Mysore. The comparative reproduction performance of *Tetrastichus howardi* in Pure Mysore, Nistari, HB6, BL6, MO6, and MCon showed that progeny production was significantly influenced by the different multivoltine hosts. Significantly higher progeny production was registered in MO6 (85, with 4 males and 81 females), followed by Nistari (62, with 4 males and 58 females), while Pure Mysore produced only 2 progeny, both of which were female.

Table1. Effect of host (Bombyx mori L.) Multivoltine Races on the progeny production of Tetrastichus howardi

Host	**Pupal weight (g)**	**Developmental duration (days)**	**Progeny production (No.)**		**Total Progeny production (No.)**	**Sex ratio (♀/♂)**	**Female longevity (days)**	
			Male	Female			Progeny	Parent
Pure Mysore	1.30 ± 0.10	20 ± 0.0	-	2 ± 0.0	2 ± 0.0	0.0	17 ± 0.0	11.80 ± 1.20
Nistari	0.79 ±0.27	22 ± 0.4	4 ± 0.0	58.0 ± 0.0	62.0 ± 0.0	17 ± 0.0	18 ± 1.41	10.0 ± 1.22
MO 6	1.25 ± 0.10	19 ± 0.70	4 ± 0.0	81 ± 0.0	85 ± 0.0	20.25 ± 0.0	21.0 ± 0.41	13.0 ± 1.3
BL 65	-	-	-	-	-	-	-	-
HB6	-	-	-	-	-	-	-	-
Mcon	-	-	-	-	-	-	-	-
*** Data are the means of ten replications (Mean ± SD).**								



Graph 1. Numbers of progeny produced by different bivoltine breeds of Bombyx mori L.

Table 2. Influence of host (Bombyx mori L.) Multivoltine races on the morphometric characteristics of Tetrastichus howardi

Host breed	Body length (mm)		Body width (mm)		Wing span (mm)		Head width (mm)		Female abdomen (mm)		Male abdomen (mm)	
	Male	Female	Male	Female	Male	Female	Male	Female	Length	Width	Length	Width
Pure Mysore	-	1.96 ± 0.0	-	0.89 ± 0.0	-	1.55 ± 0.0	-	0.7 ± 0.0	0.83 ± 0.0	0.81 ± 0.0	-	-
MCon	-	-	-	-	-	-	-	-	-	-	-	-
HB 6	-	-	-	-	-	-	-	-	-	-	-	-
Nistari	1.55 ± 0.03	1.97 ± 0.06	0.63 ± 0.07	0.71 ± 0.06	1.03 ± 0.09	1.34 ± 0.12	0.54 ± 0.07	0.75 ± 0.03	1.06 ± 0.05	0.65 ± 0.06	0.79 ± 0.03	0.56 ± 0.10
BL 65	-	-	-	-	-	-	-	-	-	-	-	-
MO 6	1.47 ± 0.16	2.17 ± 0.17	0.66 ± 0.06	0.85 ± 0.06	1.26 ± 0.06	1.59 ± 0.12	0.68 ± 0.08	0.79 ± 0.02	1.12 ± 0.20	0.84 ± 0.06	0.78 ± 0.08	0.66 ± 0.06

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