Available Online at http://www.journalajst.com

ISSN: 0976-3376

Asian Journal of Science and Technology
Vol.06, Issue, 10, pp. 1836-1839, October, 2015

ASIAN JOURNAL OF
SCIENCE AND TECHNOLOGY

RESEARCH ARTICLE

STUDIES ON THE POPULATION DENSITY FOR THE FIFTH INSTAR LARVAE OF THE COMMERCIAL SILKWORM, *BOMBYX MORI* L

***[1]Reddy P. Lakshminarayana, [2]Babu, M. A. Shanthan, [1]Ravi, G., [1]Sujatha, B.,**
[1]Satyanarayana, B. and [1]Sankar, S. Naik

[1]Department of Sericulture, Sri Krishnadevaraya University, Anantapur - 515 003, India
[2]Regional Sericultural Research Station, CSB, PB No. 50, Anantapur – 515 001, India

ARTICLE INFO

Article History:
Received 19th July, 2015
Received in revised form
12th August, 2015
Accepted 23rd September, 2015
Published online 17th October, 2015

Key words:
Commercial silkworm,
Bombyx mori,
Fifth instar,
Larval density.

ABSTRACT

Population density during the ultimate larval instar stadium is one of the crucial issues for successful commercial cocoon crop of the mulberry silkworm, *Bombyx mori* L. Experiments were conducted with two commercial silkworm hybrids; one from the multivoltine x bivoltine hybrid, PM x CSR2 and the other from the bivoltine x bivoltine hybrid, CSR2 x CSR4 to examine the optimum population density requirements for commercial rearing during fifth instar larval period. Results clearly indicated that the fifth instar larval eating period of the silkworm is directly related to the fifth instar silkworm larval density while the fifth instar larval weight is related inversely. The two commercial silkworm hybrids studied differ in their optimum larval population density requirements during the fifth larval instar period for maximum fifth instar larval growth (weight) and minimum fifth instar larval duration. Based on all the population density regimes studied, the results revealed three fifth instar larval density zones; (1) un-economic fifth instar larval density zone, (2) optimum fifth instar larval density zone and (3) loss fifth instar larval density zone. For PM x CSR2, the optimum fifth instar larval density zone of 70 to 90 number of larvae/feet^2 was more suited while that for CSR2 x CSR4, the fifth instar larval density zone was 60 to 70 number of larvae/feet^2. Results were discussed on the basis of contemporary commercial silkworm rearing in India.

INTRODUCTION

The mulberry silkworm, *Bombyx mori* L. is reared commercially only on mulberry (*Morus* sp.) foliage for the silk production. Silkworm rearing is described as the combination of science and art. In India, a sericulture farmer cultures a population of silkworm larvae, ranging from 25000 (50 DFLs) to 500000 larvae (1000 DFLs) per each batch of commercial rearing (Babu, 2014, Srinath, 2014) and harvests five to ten cocoon crops every year based on the type of hybrid. The uniformity in larval growth and development during the commercial rearing and the minimum time/labour intake for the completion of larval period aiming at production of good cocoon crop is the utmost important aspect of commercial silkworm rearing (Babu, 2014, Srinath, 2014). The commercial multivoltine x bivoltine hybrid, PM x CSR2 and the bivoltine x bivoltine hybrid, CSR2 x CSR4 are popular in the contemporary Indian sericulture. The growth in the fifth instar larvae is rather high and visible to that of the larvae of earlier instars. Therefore, the investigations pertaining to larval population density were concentrated in the fifth instar period alone. It is reported that fifth instar larval period recorded high growth, especially on the fifth (for multivoltine x bivoltine; Reddy, 1993) and on sixth day (for bivoltine x bivoltine hybrids; Rahmatulla, 2012) of fifth instar. Citations on the rearing spacing are available based on the space required for 100 DFLs (Jolly, 1987, Dandin *et al*., 2000, Ganga and Chetty, 2010). However, larval density related studies are lacking. The information is available only for bivoltine hybrid silkworm rearing. Rajan *et al*. (2003) recommended that the fifth instar bivoltine hybrid silkworm larvae should be provided with a density of 50 to 70 larvae per feet^2 during fifth instar period to reduce secondary contamination, to support enough growth, to obtain better cocoon yield and to improve silk quality. However, such records are not available for multivoltine x bivoltine silkworm hybrid rearing. Thus, conclusive studies on fifth instar larval density requirements are lacking. The results on the optimum larval population density (number of larvae per feet^2) requirements during the fifth instar larval period of multivoltine x bivoltine and bivoltine x bivoltine hybrid are reported in the present communication, considering two rearing characters *viz*., larval duration and larval weight.

MATERIALS AND METHODS

Two silkworm hybrids; one from multivoltine x bivoltine hybrid, PM x CSR2 and the other from bivoltine x bivoltine hybrid, CSR2 x CSR4 that are popularly reared commercially in the contemporary Indian sericulture are selected for the

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****Corresponding author: Reddy P. Lakshminarayana,***
Department of Sericulture, Sri Krishnadevaraya University, Anantapur - 515 003, India.

experimentation. The DFLs (disease free layings; each DFL is group of 400 to 500 silkworm eggs laid by a single silk moth on a single day) of commercial silkworm hybrids; PM x CSR2 and CSR2 x CSR4 were procured from the Silkworm Seed Production Centre (SSPC), National Silkworm Seed Organization (NSSO), Central Silk Board (CSB), Mysore, India. The silkworm larvae were reared on the foliage of a popular mulberry variety, V1 (*Morus* sp.) according to Krishnaswami (1986).

Table 1. Details of different larval density treatments employed for two commercial silkworm hybrids, PM x CSR2 and CSR2 X CSR4, number of plastic trays and number of larvae kept from the completion of fourth larval-to-larval moult to completion of silkworm larval ripening. Number of trays required for 100 DFLs rearing (50000 larvae) is also indicated

S. No.	Treatment (Number of larvae/feet^2)	Plastic tray dimension	Plastic tray area	Number of larvae kept/tray	Trays required for 100 DFLs (50000 larvae)
1	40	2' x 3'	6 feet^2	6 x 40 = 240	208
2	50	2' x 3'	6 feet^2	6 x 50 = 300	167
3	60	2' x 3'	6 feet^2	6 x 60 = 360	139
4	70	2' x 3'	6 feet^2	6 x 70 = 420	109
5	80	2' x 3'	6 feet^2	6 x 80 = 480	104
6	90	2' x 3'	6 feet^2	6 x 90 = 540	93
7	100	2' x 3'	6 feet^2	6 x 100 = 600	83
8	110	2' x 3'	6 feet^2	6 x 110 = 660	76
9	120	2' x 3'	6 feet^2	6 x 120 = 720	69
10	130	2' x 3'	6 feet^2	6 x 130 = 780	64

The larval density has been described as number of larvae/feet^2 (Rajan *et al*., 2003). For determination of optimum larval density during fifth instar silkworm rearing period, 10 larval density treatments, starting from 40 number of larvae/feet^2 to 130 number of larvae/feet^2 were considered, each for PM x CSR2 and CSR2 x CSR4 (Table 1). For each treatment/ silkworm hybrid combination, 5 replications were maintained. Till the completion of the fourth larval stadium, the silkworm larvae were reared according to standard rearing method (Krishnaswami, 1986). The larvae were shifted to plastic rearing trays of 2' x 3' dimension immediately after completion of fourth larval-to-larval ecdysis (one tray each for each treatment, each silkworm hybrid and each replication), as detailed in Table 1.

From the experimental treatments, two silkworm rearing parameters *viz*., fifth instar larval duration and fifth instar larval weight on 5th day (for PM x CSR2) and 6th day (for CSR2 x CSR4) were recorded. The fifth instar larval duration was recorded from the peak time of the larvae out of fourth larval-to-larval ecdysis to the peak time of larval ripening and expressed in hours. Similarly, the larval weight, indicating the larval growth in the contemporary Indian mulberry sericulture practices, has been recorded at 10.00 h on the fifth day for PM x CSR2 and on the sixth day for CSR2 x CSR4. One hundred numbers of larvae were selected at random from each treatment/replication and weighed individually on digital weighing balance. From the recorded weights of the larvae, the average larval weight was derived to denote fifth instar larval weight. The data were treated statistically (ANOVA) and all the F values, below 5% are designated as significant and those below 1% level as highly significant. Values that are above 5% level are coined as non-significant.

RESULTS

Results on the impact of fifth instar larval density on the fifth instar larval eating period registered interesting trends, clearly indicating that the fifth instar larval density in the multivoltine x bivoltine hybrid, PM x CSR2 (Fig. 1) is directly related to fifth larval eating period. Similarly, the fifth instar larval density and fifth larval instar eating period in the bivoltine x bivoltine hybrid, CSR2 x CSR4 (Fig. 2) are directly related. Low fifth larval eating periods were recorded under low fifth instar larval density regimes. Further, the fifth instar larval eating period increased according to the increase in fifth instar larval density.

However, the response of fifth instar larval density from 40 number of larvae/feet^2 to 90 number of larvae/feet^2 (6 density regimes, Fig. 1) to the fifth instar larval eating period was identical, without any statistical differences among themselves. When the bivoltine x bivoltine silkworm hybrid, CSR2 x CSR4 is examined, the fifth instar larval densities of 40 number of larvae/feet^2 to 70 number of larvae/feet^2 have alone resulted in identical fifth instar larval durations (Fig. 2), with no differences statistically. Thus, the densities of 40, 50, 60, 70, 80 and 90 larvae/feet^2 appears to be on safer side for obtaining minimum fifth instar larval duration for PM x CSR2. For CSR2 x CSR4, however, the fifth instar larval densities of 40, 50, 60 and 70 number of larvae/feet^2 only are to be considered in this aspect. The fifth instar larval durations in these fifth instar larval density regimes recorded to be 122 to128 h for PM x CSR2 and 148 to 158 h for CSR2 x CSR4. Highest fifth instar larval duration of 163 h was recorded for PM x CSR2 and 169 h for CSR2 x CSR4 under maximum fifth instar larval densities of 130 number of larvae/feet^2.

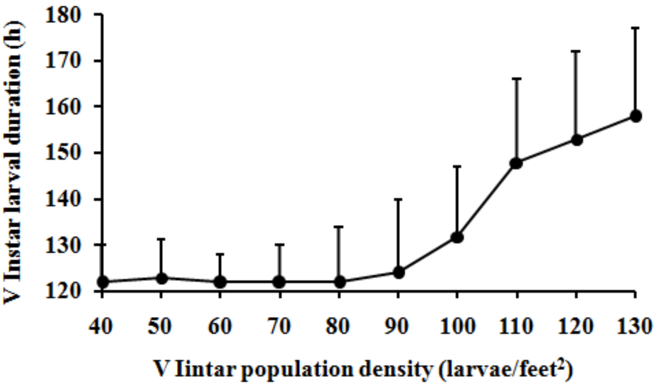

Fig. 1. Impact of fifth instar larval population density (number of larvae/feet2) on the fifth instar larval eating period (in hours) duration of PM x CSR2. Values are mean of 5 replications ± SD. Values at larval population density of 40 to 90 number of larvae/feet2 are not significantly different among themselves while the other larval density (100 to 130) cases are significantly different over the initial 6 density regimes

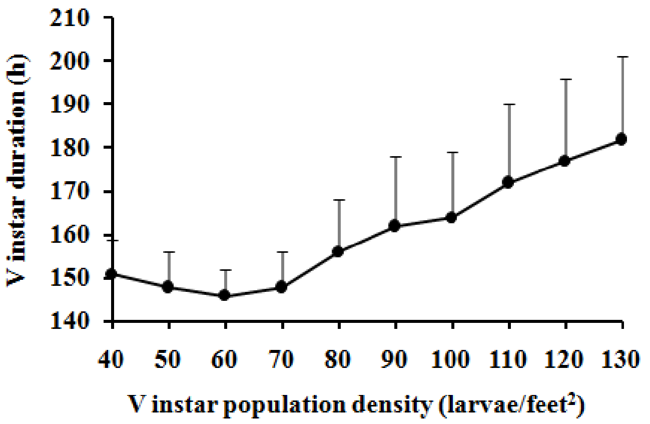

Fig. 2. Impact of fifth instar larval population density (number of larvae/feet2) on the fifth instar larval eating period (in hours) of CSR2 x CSR4. Values are mean of 5 replications ± SD. Values at larval population density of 40, 50, 60 and 70 number of larvae/feet2 are not significantly different among themselves while the other larval density cases are significantly different (p < 0.01) over the initial 4 density regimes

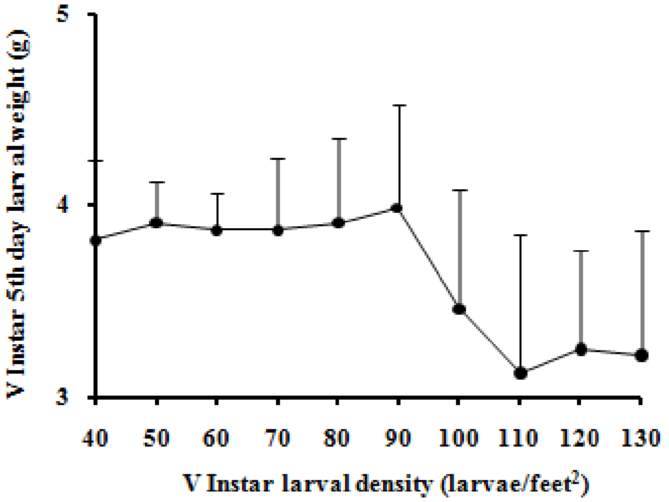

Fig. 3. Impact of larval population density (number of larvae/feet^2) in the rearing bed on the fifth instar 5th day larval weight (in grams) of PM x CSR2. Values are mean of 5 replications ± SD. Values at larval population density of at 40 to 90 number of larvae/feet^2 are not significantly different among themselves ($p < 0.01$) while the other larval density treatments are significantly different over the initial 6 density treatments

The relationship between the larval density (number of larvae/feet^2) during fifth larval instar period for PM x CSR2 and the weight of fifth day larva of fifth instar is inversely related (Fig. 3). Thus, the initial 6 larval densities (40 to 90 larvae/feet^2) have shown more larval weights, however, with no statistical significance. On the other hand, larvae reared in the later 4 larval density regimes (100 to 130 larvae/feet^2) have registered less larval weights. The differences between these four treatments and that of the other 6 treatments are statistically highly significant ($p < 0.01$). Results on the same lines with CSR2 x CSR4 (Fig. 4) also indicated a highest larval weight of 4.5 g with the fifth instar larval population density of 40 to 70 number of larvae/feet^2. These larval weights are not significantly different from each other statistically within these 4 larval density (40, 50, 60 and 70 number of larvae/feet^2) regimes while the larval weights of the other fifth instar larval density (80, 90, 100, 110, 120 and 130 number of larvae/feet^2) regimes are significantly different and low ($p < 0.01$; Fig. 4). Lowest larval weight of fifth instar sixth day of 3.4 g has been recorded with a fifth instar larval population density of 130 number of larvae/feet^2 for CSR2 x CSR4. The results thus, implied that the fifth instar larval population density at 40 to 70 number of larvae/feet^2 are desired compared to the other fifth instar larval density treatments.

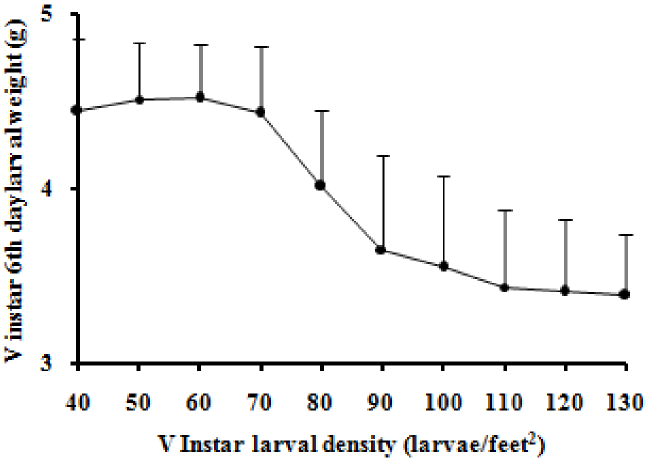

Fig. 4. Impact of larval population density (number of larvae/feet^2) in the rearing bed on the fifth instar 5th day larval weight (in grams) of CSR2 x CSR4. Values are mean of 5 replications ± SD. Values at larval population density of at 40, 50, 60 and 70 number of larvae/feet^2 are not significantly different among themselves ($p < 0.01$) while the other larval density treatments are significantly different over the initial 4 density treatments

DISCUSSION

From the results, it is clear that the larval density provided during the rearing of fifth larval instar period in terms of number of larvae/feet^2 has definite relationship for the two rearing characters studied in the bivoltine x bivoltine, CSR2 x CSR4 and the multivoltine x bivoltine PM x CSR2 hybrids. The fifth instar larval period was directly related to fifth instar larval density while the fifth instar larval weight related inversely. Added to the above general results, it is observed that initial low fifth instar larval densities have little impact on the traits studied. Thus, fifth instar larval densities of 40, 50, 60, 70, 80 and 90 number of larvae/feet^2 have recorded statistically non-significant results for PM x CSR2 while the same of 40, 50, 60 and 70 larvae per feet^2 have recorded identical results or results having no differences among themselves, statistically for the bivoltine x bivoltine hybrid, CSR2 x CSR4. In other words, the multivoltine x bivoltine hybrid, PM x CSR2 has responded with statistically non-significant differences in results for the first 6 larval density regimes (40, 50, 60, 70, 80 and 90 larvae per feet^2) and bivoltine x bivoltine hybrid, CSR2 x CSR4 has responded with statistically non-significant differences in results for the first 4 larval density treatments (40, 50, 60 and 70 larvae/feet^2) only. These observations clearly indicated that the bivoltine hybrids require low levels of fifth instar larval density regimes for better results while the multivoltine x bivoltine hybrid, PM x CSR2 can withstand fifth instar larval densities to the maximum extent of 90 number of larvae/feet^2. The results and discussion made so far indicated that there exist only two fifth instar larval density zones; the better fifth instar larval density zone and the loss fifth instar larval density zone.

However, this classification of fifth instar larval density zones seems non justifiable for the economic space of the larval density requirements also accounts in the commercial silkworm rearing. If the fifth instar larval density of 40 larvae/feet^2 for CSR2 x CSR4 is considered, the entire larval rearing space requirement per 100 DFLs of 50000 larvae (100 DFLs) would be 50000/40 = 1250 feet^2. Further, if the fifth instar larval density of 50 larvae/feet^2 for the bivoltine x bivoltine hybrid, CSR2 x CSR4 is to be provided, the entire larval rearing space requirement per 100 DFLs of 50000 larvae would be 50000/50 = 1000 feet^2. These two treatments (40 and 50 larvae/feet^2) are on the higher side of requirements and definitely proved to be uneconomical for their higher rearing space. Moreover, Dandin *et al.* (2000) viewed that less crowding is uneconomical with wastage of space and feed. Therefore, the densities of 40 and 50 larvae/feet^2 are designated uneconomical fifth instar larval densities for the bivoltine x bivoltine hybrid, CSR2 x CSR4. With PM x CSR2 too, the first 6 fifth instar larval densities (40, 50, 60, 70, 80 and 90 larvae/feet^2) have no doubt, given good results in terms of fifth instar larval period and larval weight.

However, the uneconomic fifth instar larval densities for PM x CSR2 are restricted for the first three fifth instar larval densities (40, 50 and 60 larvae per feet^2) as the rearing area required for rearing of 100 DFLs at 40 larvae/feet^2 is 1500 feet^2 (50000/40 = 1250 feet^2), the rearing area required for rearing of 100 DFLs at 50 /feet^2 is 1000 feet^2 (50000/50 = 1000 feet^2) and that required for rearing of 100 DFLs at 60 larvae/feet^2 is 834 feet^2 (50000/60 = 834 feet^2). Consolidating, the rearing space of 1250 and 1000 feet^2 for CSR2 x CSR4 and 1250, 1000 and 834 feet^2 for PM x CSR3 are towards higher side and, therefore, could be designated as 'un-economic fifth instar larval density zone'. Certain fifth instar larval density regimes recorded low fifth instar larval weights, higher fifth instar larval duration (present study) and higher unequal larval percentage coupled with higher cocoon melting percentage during fifth instar (Ravi, 2014). For bivoltine x bivoltine hybrid, CSR2 x CSR4, these fifth instar larval densities which recorded undesired results are 80, 90, 100, 110, 120 and 130 larvae/feet^2. Similarly, for multivoltine x bivoltine hybrid, PM x CSR2, the fifth instar larval densities of 100, 110, 120 and 130 larvae/feet^2 recorded un-preferred results and loss to the farmers in commercial silkworm rearing. Therefore, these fifth instar larval density treatments are designated as 'loss fifth instar larval density zone'. Thus, 80, 90, 100, 110, 120 and 130 larvae/feet^2 for the bivoltine x bivoltine hybrid, CSR2 x CSR4 and 100, 110, 120 and 130 larvae/feet^2 for the multivoltine x bivoltine hybrid, PM x CSR2 are conveniently designated as 'loss fifth instar larval density zone'. Finally, the 'optimum fifth instar larval density zone', as per the results obtained for the multivoltine x bivoltine hybrid, PM x CSR2 are the larval densities of 70, 80 and 90 larvae/feert^2 and that for the bivoltine x bivoltine hybrid, CSR2 x CSR4 are the fifth instar larval densities of 60 and 70 larvae/feert^2.

Acknowledgements

The authors gratefully acknowledge the critical review and editing of the manuscript to its best form by Dr. Sivarami Reddy, N., Scientist-D (Retd.), RSRS, Anantapur, India.

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