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Silkworm pupal residue products foliar spray impact in silkworm (*Bombyx mori* L.)

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Abstract
A field experiment was conducted to study the effect of foliar spray of Silkworm pupal residue protein (SPRP) and Silkworm pupal residue extract (SPRE) in mulberry (V_1) and silkworm. The experiment was conducted with fourteen treatments and three replications in a randomized complete block design. The results of the study indicated that, significantly higher fifth instar larval weight (38.78 g/10), less fifth instar larval duration(156.58 hours) and less fourth moulting duration (25.63 hours) was recorded when PMXCSR$_2$ worms were fed with leaves grown by the foliar spray of 2% SPRP + RDF + 20MT FYM/ha/year (T_6) and it was statistically on par with T_8 (SPRE spray @ 1 % + SPRP spray @ 1 % + RDF+ 20MT FYM/ha/year) which recorded 38,00 g/10,157.53 hours and 25.71 hours, respectively. Higher total sugar (2.52 %) and lipid (3.04 %) content was recorded in T_6 (foliar spray of 2% SPRP + RDF + 20MT FYM/ha/year). Higher effective rate of rearing (99.10 %) was recorded when the PMX CSR$_2$fed with leaves grown by the foliar spray of 2% SPRP + RDF + 20MT FYM/ha/year (T_6).

Keywords: silkworm pupal residue protein, silkworm pupal residue extract, rearing

Introduction
Silk – The queen of textiles is the natural fiber, spells luxury, elegance, class and comfort, which is secreted by silkworm. India is the second largest producer of silk and also the largest consumer of silk in the world. The silkworm is a monophagous insect which depends on the mulberry for its complete growth and development. Due to this, silkworm requires specific quality of leaves during different phases of its growth and this reflects on the importance of mulberry cultivation practices. According to Miyashita (1986) [7] the mulberry leaf alone contributes to the extent of 38.2% for successful cocoon crop production. Due to increasing demands for silk, stress has to be laid on the further improvement of leaf productivity both in terms of quality and quantity (Geetha *et al*., 2017) [4].

The cost of mulberry leaf production is increasing every year because of the usage of high and costly chemical inputs. In mulberry cultivation high dose of nitrogen (350 kg/ha/year) used leads to increased cost of production. Health hazards, environmental, water and soil pollution is the ill effects of using heavy dose of chemical fertilizers for a prolonged period.

In the sericulture, substantial amounts of by-products are generated at different stages can be recycled and utilized. Hence, a proper recovery and optimum utilization of various byproducts of sericulture goes a long way in meeting several demands. Silkworm pupa is a major byproduct of silk reeling industry obtained after reeling of silk. In India, it is estimated that about 40,000 MT of dry pupa is generated per year. Pupa contains about 79.8% protein, 6.68% fat and 5.15% ash. The silkworm pupal residue products and urea can be used as foliar sprays as nitrogen source to improve leaf quality and quantity. Hence, the present study was undertaken to find out the effect of different concentrations of (0, 5, 1 and 2 per cent) silkworm pupal residue protein (SPRP) and silkworm pupal residue extract (SPRE) and urea to

improve the silkworm rearing and biochemical traits.

The pupal of silkworm has vast applications in a variety of field. Effective use of by-products not only increases the rearer's income but also helps in fuller utilization of natural resources. This vital aspect so far seems to have not been taken proper care in sericulture industry. Pupae can be utilized in a better way by producing value added products with the adoption of improved technology. The by-products presently felt as wastes, can put to better use in generating the value based products and there by catapult the industry to a more profitable and economically viable spot. The full utilization of silkworm pupae as different marketable products and such an integrated operation can certainly make the sericulture more practical. The cost of end product *i.e.* the silk can be proportionately brought down by the combination of regulating the processing methods and converting the wastes as useful by-products. The optimal by-product utility concept can be highly useful to sericulture industry, which can help in elevating the socio economic status of the rural poor rearer's. Profitable conversion of wastes/ by-products to high value utilities through phyto and post-harvest technologies, the collaboration of seri scientists with related industries, to locate functional activities for potential applications can reduce the production cost, pollution, recycles resources to cater the ever growing population and their demanding wants.

Materials and Methods

A field experiment was conducted in an established irrigated V_1 mulberry garden planted at a spacing of 90X90 cm in order to study the effect of foliar application of silkworm pupal residue extract on mulberry and silkworm in randomized complete block design with fourteen treatments and three replications at Department of Sericulture, UAS, GKVK, Bengaluru, Karnataka. All the cultural practices were followed as per the package of practices for irrigated mulberry garden (Dandin *et al*., 2010) [3]. Foliar spray of silkworm pupal residue extract, silkworm pupal residue protein and urea were given as per treatments at 30, 40 and 50 days after pruning (DAP) the mulberry garden. The treatments were T_1= Silkworm pupal residue extract (SPRE) spray @ 0.5 % + $T_{14,}$ $T_{2=}$ SPRE spray @ 1 % + $T_{14,}$$T_{3=}$ SPRE spray @ 2 % + $T_{14,}$$T_4$= SPRP spray @ 0.5 % + $T_{14,}$ T_5= SPRP spray @ 1 % + $T_{14,}$ T_6= SPRP spray @ 2% + $T_{14,}$ $T_{7=}$ SPRE spray @ 0.5 % + SPRP spray @ 0.5 % + $T_{14,}$ $T_{8=}$ SPRE spray @ 1 % + SPRP spray @ 1 % + $T_{14,}$ $T_{9=}$ SPRE spray @ 2 % + SPRP spray @ 2 % + $T_{14,}$$T_{10}$= Urea spray @ 0.5% + $T_{14,}$ T_{11}= Urea spray @ 1% + $T_{14,}$ T_{12}= Urea spray @ 2% + $T_{14,}$ T_{13}= Aqueous extract + $T_{14,}$and T_{14}= Control (RDF+ 20MT FYM/ha/year).Preparation of silkworm pupal residue extract: This was prepared by pressing of raw pupae in cotton bag contained water followed by clarified from muslin cloth. Prior to foliar spraying, the pupae extract was analyzed for nitrogen. Nitrogen content of silkworm pupal extract was 11%.

Preparation silkworm pupal residue protein: Collected silkworm pupa was crushed and extract was dissolved in pH 7 buffer and filtered through filter paper. To the filtrate, ammonium sulphate was added slowly and stir constantly for 15 minutes. Then extract was centrifuged at 13,000 rpm. The collected pellets were analyzed for nitrogen. Nitrogen content of silkworm pupal protein was 15%.

The silkworm rearing was conducted at Department of Sericulture, UAS, GKVK, Bengaluru. PM X CSR_2 breed was reared to assess the rearing performance, by feeding the leaves raised with different treatments. The following observations were recorded *viz*., Matured larval weight (g), Fifth instar Larval duration (hours), Moulting Duration, Effective rate of rearing (ERR) (%), Disease incidence (%), Total sugar (%) and Lipid (%).

Data recorded on various parameters were subjected to Fishers method of analysis of variance and interpreted as given by Gomez and Gomez (1984) [5]. The level of significance used in F and t test was P=0.05 for RCBD and P=0.01 for CRD. Critical difference (CD) values were calculated where F test was found significant.

Results and Discussion

Significantly higher mature larval weight (38.78g/10),less fifth instar larval duration (156.58hours), lesser disease incidence (0.89 %) and higher effective rate of rearing (99.10 %) was recorded when PM x CSR_2 worms were fed with mulberry grown by the foliar spray through 2 percent SPRP spray + RDF + 20 MT FYM/ha/year (T_6) and it was statistically on par with T_8 (SPRE spray@1% + SPRP spray@ 1%+ RDF + 20 MT FYM/ha/year) recorded mature larval weight of 38.00 g/10, fifth instar larval duration of 157.53 hours, 1.74 % of disease incidence and 98.25 % of ERR over other treatments (Table 1). Less first (24.20 hours), second (23.69 hours), third (25.52 hours) and fourth moulting period (25.63 hours) was recorded in PM x CSR_2 fed with mulberry grown by the foliar spray through 2 percent SPRP spray + RDF + 20 MT FYM/ha/year (T_6) and it was statistically on par with T_8 (SPRE spray@1% + SPRP spray@ 1%+ RDF + 20 MT FYM/ha/year) where in 24.40 h, 23.78 h, 25.59 h, and 25.71 h of first, second, third and fourth moulting duration was noticed over other treatments (Table 2 & Figure 1). Highest total sugar (2.52 %) and lipid (3.04 %) content was recorded when the PM x CSR_2 was fed with mulberry grown by the foliar spray through 2 percent SPRP spray + RDF + 20 MT FYM/ha/year (T_6) and it was followed by T_8 (SPRE spray@1% + SPRP spray@ 1%+ RDF + 20 MT FYM/ha/year) which recorded 2.46 % of total sugars and 2.96 % of lipid over other treatments. (Table 3 & Figure 2). Ankalagi and Ansari, (1992) [1] reported that, spraying protein based 'Fasal', a 1- tricontanol based product at 1.0 ml/lit on K-2 mulberry for three times at 10 days interval after 15 days of pruning and feeding such leaves to NB_4D_2 has increased the growth performance of silkworm and reduced the larval mortality. Silk protein synthesis starts in fourth and fifth instar of silkworm, there is a huge demand for protein rich food in late instars. Dietary supplementation of the leaf with organic extracts elicited varied responses in the final instar larvae of the mulberry silkworm, *Bombyx mori* L. (Rajeswari and Isaiarasu, 2004) [9]. The nutritional supplement of soluble protein aqueous extracts from waste pupae on larval instars of *Antheraea assama* in different concentrations has an impact on the larval growth and cocoon parameters. The protein extract of muga pupa can be utilized as a supplement on the muga food plant as a growth stimulator in silkworm (Saikia *et al*.,, 1971) [10]. Similar results have also been reported by Vishwanath *et al*. (1997) [11], Jayaprakshrao *et al*. (1998) [6], Basit and Ashfaq (1999) [2] and Narahari *et al*. (2001) [8].

Table 1: Feeding influence of V_1 leaf obtained through silkworm pupal Residue protein (SPRP) and Extract (SPRE) spray on larval traits of PM X CSR_2

	Treatments	Mature larval weight (g/10 larvae)	Fifth instar larval duration (h)	Effective rate of rearing (%)	Disease incidence (%)
T_1	SPRE @ 0.5 % + T_{14}	37.34	160.10	94.52	5.46
T_2	SPRE @ 1 % + T_{14}	37.52	159.32	95.81	4.18
T_3	SPRE @ 2 % + T_{14}	36.86	158.06	97.08	2.92
T_4	SPRP @ 0.5 % + T_{14}	37.26	159.60	95.18	4.80
T_5	SPRP @ 1 % + T_{14}	36.44	158.90	96.85	3.13
T_6	SPRP @ 2 % + T_{14}	38.78	156.58	99.10	0.89
T_7	SPRE @ 0.5 % + SPRP @ 0.5 %+ T14	36.53	158.34	95.14	4.85
T_8	SPRE @ 1 % + SPRP @ 1 %+ T_{14}	38.00	157.53	98.25	1.74
T_9	SPRE @ 2 % + SPRP @ 2 %+ T_{14}	36.68	158.51	96.79	3.20
T10	Urea spray @ 0.5 %+ T_{14}	36.28	159.70	94.09	5.89
T11	Urea spray @ 1 %+ T_{14}	36.30	159.14	94.00	5.99
T12	Urea spray @ 2 %+ T_{14}	36.82	158.60	94.66	5.34
T13	Aqueous extract + T_{14}	35.96	158.37	91.84	8.14
T14	RDF + 20 MT FYM/ha/year	35.27	161.27	90.50	9.48
	F – Test	*	*	*	*
	S. Em ±	0.60	0.82	0.87	0.39
	C.D @ 1%	1.81	2.44	2.62	1.19

Note: *-Significant

Table 2: Feeding influence of V_1 leaf obtained through silkworm pupal Residue protein (SPRP) and Extract (SPRE) spray on on Moulting duration (hours) of PM X CSR_2

	Treatments	Moulting duration (h)			
		I Moult	II Moult	III Moult	IV Moult
T_1	SPRE @ 0.5 % + T_{14}	24.66	24.20	26.18	25.91
T_2	SPRE @ 1 % + T_{14}	24.59	24.06	26.00	25.85
T_3	SPRE @ 2 % + T_{14}	24.48	23.81	25.64	25.78
T_4	SPRP @ 0.5 % + T_{14}	24.60	24.31	26.02	26.00
T_5	SPRP @ 1 % + T_{14}	24.50	24.07	25.80	25.95
T_6	SPRP @ 2 % + T_{14}	24.20	23.69	25.52	25.63
T_7	SPRE @ 0.5 % + SPRP @ 0.5 %+ T14	24.51	23.81	25.60	25.74
T_8	SPRE @ 1 % + SPRP @ 1 %+ T_{14}	24.40	23.78	25.59	25.71
T_9	SPRE @ 2 % + SPRP @ 2 %+ T_{14}	24.59	23.92	25.63	25.72
T10	Urea spray @ 0.5 %+ T_{14}	26.01	26.33	27.42	27.00
T11	Urea spray @ 1 %+ T_{14}	25.97	26.26	27.30	26.91
T12	Urea spray @ 2 %+ T_{14}	25.92	26.08	27.15	26.75
T13	Aqueous extract + T_{14}	27.98	27.34	27.04	27.00
T14	RDF + 20 MT FYM/ha/year	28.20	27.50	27.15	27.34
	F – Test	*	*	*	*
	S. Em ±	0.64	0.73	0.54	0.88
	C.D @ 1%	1.93	2.21	1.60	2.66

Note: *-Significant

Table 3: Feeding influence of V_1 leaf obtained through silkworm pupal residue protein (SPRP) spray on total sugars (%) and lipid (%) contents of PM X CSR_2

	Treatments	Total sugars (%)	Lipid (%)
T_1	SPRE @ 0.5 % + T_{14}	2.30	2.75
T_2	SPRE @ 1 % + T_{14}	2.38	2.85
T_3	SPRE @ 2 % + T_{14}	2.41	2.91
T_4	SPRP @ 0.5 % + T_{14}	2.39	2.80
T_5	SPRP @ 1 % + T_{14}	2.43	2.93
T_6	SPRP @ 2 % + T_{14}	2.52	3.04
T_7	SPRE @ 0.5 % + SPRP @ 0.5 %+ T14	2.42	2.79
T_8	SPRE @ 1 % + SPRP @ 1 %+ T_{14}	2.46	2.96
T_9	SPRE @ 2 % + SPRP @ 2 %+ T_{14}	2.44	2.85
T10	Urea spray @ 0.5 %+ T_{14}	2.29	2.63
T11	Urea spray @ 1 %+ T_{14}	2.33	2.66
T12	Urea spray @ 2 %+ T_{14}	2.36	2.71
T13	Aqueous extract + T_{14}	2.30	2.61
T14	RDF + 20 MT FYM/ha/year	2.26	2.55
	F – Test	*	*
	S. Em ±	0.06	0.12
	C.D @ 1%	0.19	0.35

Note: *-Significant

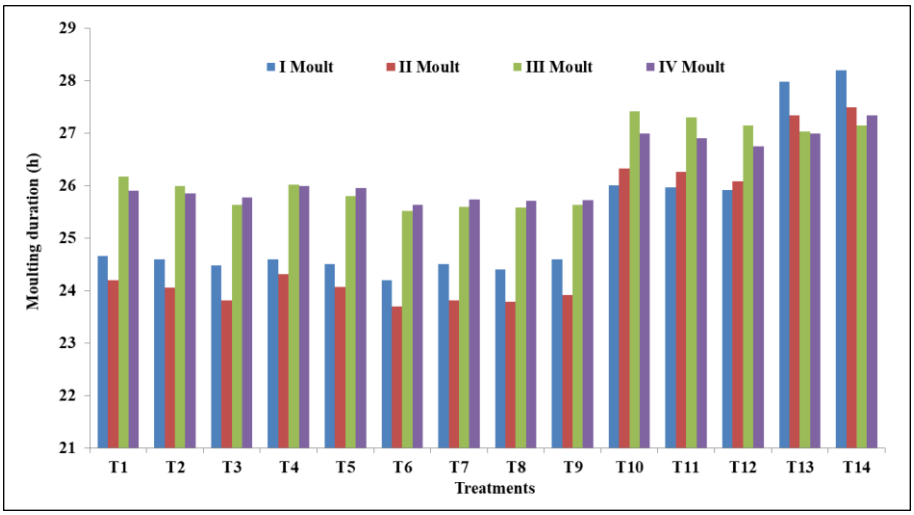

Fig 1: Feeding influence of V_1 leaf obtained through silkworm pupal residue protein (SPP) spray on moulting duration (hours) of PM x CSR_2

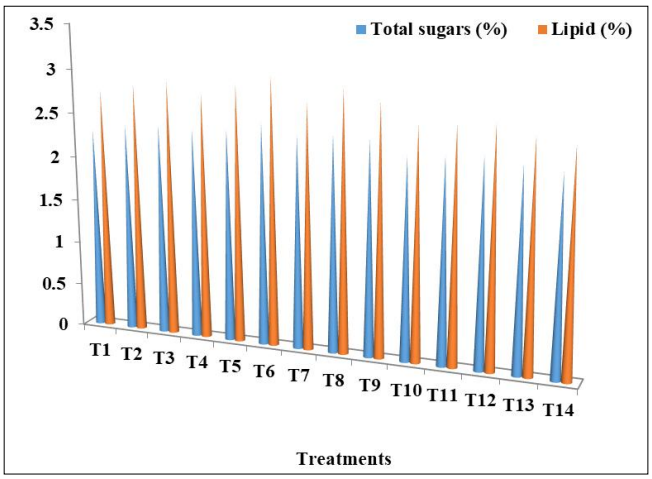

Fig 2: Feeding influence of V_1 leaf obtained through silkworm pupal residue protein (SPRP) spray on total sugars (%) and lipid (%) contents of PM x CSR_2

Conclusion

The effect of silkworm pupal residue protein and silkworm pupal residue extract on mulberry is to be conducted under different agro-climatic regions covering all the seasons on improved mulberry varieties and on improved silkworm hybrids. The evaluation of silkworm pupal protein as a plant growth stimulator for mulberry under field conditions showed that the application of silkworm pupal protein @ 2% along with recommended dose of NPK + FYM has significantly increased the larval rearing biochemical traits.

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