



AMOUNT OF CHITIN, CHITOSAN AND CHITOSAN BASED ON CHITIN WEIGHT IN PURE RACES OF MULTIVOLTINE AND BIVOLTINE SILKWORM PUPAE *Bombyx mori* L.

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ABSTRACT

The pupae of silkworm are an alternative source of chitin which consequently yields chitosan. Chitin, a linear homopolymer of β - 1 - 4 - linked N - acetylglucosamine, is the second most abundant biopolymer next to cellulose. It is the major structural polysaccharide in the insect's exoskeleton. Among the different races of mulberry silkworm, multivoltine pure races contain higher chitin of 3.225 per cent was present in male pupae and 3.078 per cent was observed in female pupae. Similarly higher chitosan per cent was observed in male 2.449 per cent and 2.354 per cent in female pupae. Male silkworm pupae contain higher per cent of chitin (3.217%) than the female pupae (3.023%). Chitosan is also found to be higher (2.451 %) in male pupae than in female pupae (2.291 %).

KEYWORDS: Chitin, Chitosan, Silkworm pupae, Pure races, Polysaccharide and Exoskeleton

INTRODUCTION

Silk industry is an important agro based industry in India. India is the second largest producer of silk next to China. Among many valuable products in sericulture, direct use of mulberry fruits for jam, jelly and wine production are of prime importance. The use of silkworm excreta for extraction of chlorophyll (Raju, 1996), drugs and triacontanol (a plant growth promoting substance) is also feasible. The handicraft items produced from cut cocoons which have immediate market value are also gaining momentum in recent years. Pupa, which constitutes the major portion of the cocoon weight, is an inevitable by-product generated in large quantity (75-85%) during the green cocoon production. Pupa is one of the important byproduct in reeling industry (Choudhury, 2003; Dandin and Rajan, 2005). Dry pupae contain 45-49% of protein (Fagoone, 1983) and 23-24% of oil (Jolly *et al*, 1974), thus forming an important biosource of oil and proteins (Singh and Suryanarayana, 2003). After extraction of these

proteins and oil, the remaining waste is used for the extraction of chitin, which is also another important by-product from sericulture industry (Katti *et al.*, 1996). Chitin and chitosan have immense applications in various fields such as food industry, cosmetics, agriculture, water treatment, biomedicine, textile, biotechnology, paper industry; wound healing agents, etc., (Shahidi, 1994; Xu *et al.*, 1996; Chen *et al.*, 2002). The present study aims at utilizing the pupae left as reeling waste and unutilized pupae in grainage industry, where they are just dumped as a waste, resulting in greater environmental pollution.

MATERIALS AND METHODS

Estimation of chitin and chitosan

Pupae of pure races of Bivoltine and multivoltine silkworm races were used for the extraction of chitin and chitosan. Then the yield of chitin and chitosan from different races of silkworm pupae were calculated.

$$\text{Chitin (\%)} = \frac{\text{Chitin obtained (g)}}{\text{Dry weight of the pupae (g)}} \times 100$$

$$\text{Chitosan (\% in the pupae)} = \frac{\text{Chitosan obtained (g)}}{\text{Dry weight of the pupae (g)}} \times 100$$

$$\text{Chitosan (\% in the chitin)} = \frac{\text{Chitosan obtained (g)}}{\text{Weight of the chitin (g)}} \times 100$$

RESULT**Per cent yield of Chitin, chitosan and chitosan based on chitin weight in different types of silkworm pupae
Pure Multivoltine Races**

The data on per cent yield of chitin, chitosan and chitosan based on chitin weight among the three pure multivoltine races viz., Pure Mysore, Nistari and C-nichi are furnished in the Table.1 The multivoltine pure races showed significant difference among the chitin present in the male pupae. Higher chitin per cent was observed in pure mysore male pupae which was 3.407, whereas in Nistari and C-nichi it was 3.224 and 3.044 per cent respectively. There was no significant difference found among female pupae chitin. The chitin percentage in Pure Mysore, Nistari and C-nichi were 3.185, 3.082 and 2.968 per cent, respectively. Among the sexes, male pupae possessed higher chitin per cent of 3.225, whereas it was 3.078 per cent in female pupae. Higher chitosan percentage was observed in Nistari (2.528%), which was followed by Pure Mysore (2.524%) and they were on par. Lower chitosan in

male pupae were observed in C-nichi of about 2.297 per cent. Higher chitosan percentage was observed in Pure Mysore (2.441%), which was followed by Nistari (2.414%) and they were on par. Lower chitosan in female pupae was recorded in C-nichi of about 2.206 per cent. Considering the sexes, male pupae possessed higher chitosan per cent (2.449%) than the females (2.354%).

No significant difference was observed in chitosan percentage based on chitin weight in male pupae. Higher values are observed in Nistari (78.640 %), which was followed by C-nichi (75.530 %). Lower values were observed in Pure Mysore (74.043 %). No significant difference was observed in chitosan percentage based on chitin weight in females; higher values are observed in Nistari (78.340 %), which was followed by Pure Mysore of about (76.653 per cent). Lower values were observed in C-nichi (74.347). Considering the sexes, male pupae possessed higher chitosan per cent (76.447) than the females (76.256). Among multivoltine pure races Pure Mysore contain higher chitin and chitosan. Male silkworm pupae contain higher chitin content than the female.

TABLE 1. Estimation of chitin, chitosan and chitosan (based on chitin weight) in the pupae of multivoltine pure races

Races	Chitin *(%)			Chitosan *(%)			Chitosan ** (%)		
	Male	Female	Mean	Male	Female	Mean	Male	Female	Mean
Pure Mysore	3.407 a (10.637)	3.185a (10.280)	3.296a (10.459)	2.524 a (9.138)	2.441a (8.989)	2.483a (9.064)	74.043a (59.402)	76.653a (61.108)	75.348a (60.255)
Nistari	3.224 ab (10.340)	3.082 a (10.111)	3.153 ab (10.225)	2.528 a (9.149)	2.414 a (8.937)	2.471 a (9.043)	78.640a (62.570)	78.340a (62.296)	78.490a (62.433)
C-nichi	3.044 b (10.043)	2.968 a (9.920)	3.006 b (9.981)	2.297 b (8.714)	2.206 b (8.542)	2.251 b (8.628)	75.530a (60.358)	74.347a (59.575)	74.938a (59.967)
Mean	3.225 10.398	3.078 10.1036	3.152 10.2217	2.449 9.000	2.354 8.8226	2.402 8.9115	76.071 60.7769	76.447 60.9932	76.259 60.8851

* - The values are based on the dry pupal weight.

**- The values are based on dry chitin weight

In a column, means followed by a common letter are not significantly different at the 5% level by DMRT
Figures in parenthesis are arc sine transformed values**TABLE 2.** Estimation of chitin, chitosan and chitosan (based on chitin weight) in the pupae of bivoltine pure races

Races	Chitin *(%)			Chitosan *(%)			Chitosan ** (%)		
	Male	Female	Mean	Male	Female	Mean	Male	Female	Mean
CSR2	3.456a (10.707)	2.922a (9.842)	3.189a (10.275)	2.641a (9.351)	2.120a (8.371)	2.381a (8.861)	76.600a (61.125)	72.563 a (58.438)	74.586a (59.782)
CSR4	2.932b (9.857)	3.105a (10.146)	3.019ab (10.001)	2.213b (8.549)	2.204a (8.532)	2.209ab (8.540)	75.575a (60.618)	70.917 a (57.381)	73.246ab (58.999)
DIAZO	2.801b (9.633)	2.776a (9.587)	2.789b (9.610)	2.044b (8.218)	2.050a (8.229)	2.047b (8.224)	72.967a (58.675)	73.871 a (59.270)	73.419ab (58.972)
CSR50	3.123 ab (10.171)	2.963a (9.910)	3.043 ab (10.040)	2.041 b (8.209)	2.127a (8.384)	2.084b (8.296)	65.417b (53.981)	71.809 a (57.948)	68.613b (55.965)
Mean	3.078 (10.092)	2.943 (9.871)	3.010 (9.982)	2.235 (8.581)	2.126 (8.379)	2.180 (8.480)	72.642 (58.600)	72.290 (58.259)	72.466 (58.429)

* - The values are based on the dry pupal weight.

**- The values are based on dry chitin weight

In a column, means followed by a common letter are not significantly different at the 5% level by DMRT
Figures in parenthesis are arc sine transformed values**Bivoltine Pure Races**

The data on per cent yield of chitin, chitosan and chitosan based on chitin weight in four pure bivoltine races viz., CSR2, CSR4, DIAZO and CSR50 were assessed and results are presented in the Table 2. There was significant difference in chitin values among the four races of bivoltine pure races. Among the four races higher chitin value was observed in CSR2 (3.189 per cent) and it is on par with CSR50 (3.043 per

cent) and CSR4 (3.019 per cent). Lower value was observed in Diazo (2.789 per cent). Considering the sexes, male pupae possessed higher chitin per cent (3.078) than the female pupae (2.943).

Chitosan percentage also significantly differed among four races of bivoltine pure races. Among the four races higher chitosan value was observed in CSR2 (2.381 %) followed by CSR4 (2.209 %) and they are on par. Lower chitosan was observed in CSR50 (2.084 %) which was on par with

DIAZO (2.047%). Considering the sexes, male pupae possessed higher chitosan per cent (2.235) than the females (2.126).

Chitosan based on chitin weight significantly differed among four races of bivoltine pure races. Among the four races higher chitosan value was observed in CSR2 (74.586 per cent) followed by DIAZO (73.419 %) and CSR4 (73.246%) and all were on par with each other. Lower chitosan per cent based on chitin weight was observed in CSR50 (68.613%). Considering the sexes, male pupae possessed higher chitosan per cent based on chitin weight (72.642) than the females (72.290). Among four pure bivoltine races viz., CSR2, CSR4, DIAZO and CSR50, CSR2 male pupae contain higher chitin and chitosan per cent. Male pupae contain higher chitin and chitosan content.

DISCUSSION

In the present investigation, it was found that chitin content in male pure multivoltine races was 3.225 per cent whereas in female it was 3.078 per cent. In bivoltine pure races chitin content was 3.078 and 2.945 per cent in male and female, respectively. In cross breed silkworms 3.19 and 3.036 per cent in male and female, respectively. In bivoltine hybrids chitin content was 3.103 and 2.854 per cent, and in eri silkworm pupae chitin content was 3.488 per cent in male and 3.206 per cent in females. Among mulberry races, multivoltine pure races contained higher chitin content. Male pupae contained higher chitin than the females. The present investigation is supported by Aruga (1994) whom have reported that the dried silkworm pupae contain 3.73 per cent of chitin irrespective of races and sex. Ni and Liang (1999) have reported that the silkworm pupae contain 2.71 per cent of chitin. Katti *et al.* (1996) reported that the chitin yield of dried silkworm pupae is 3-4 per cent. Zhang *et al.* (2000) reported that the pupae contain 4 per cent of chitin based on dry weight.

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