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# NUTRACEUTICAL EFFECTS OF SEAWEEDS IN AUGMENTING SILK PRODUCTION IN THE SILK WORM BOMBYX MORI

Jaculin Praba C.<sup>1</sup>, Pamdmalatha C.<sup>1</sup>, Nagarajan K. and Ranjit Singh A. J. A.<sup>2</sup>\*

<sup>1</sup>Department of Animal Science, Manonmaniam Sundaranar University, Tirunelveli.

<sup>2</sup>Department of Biotechnology, Prathyusha Engineering College, Chennai.

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\*Corresponding Author Dr. Ranjit Singh A. J. A.

Department of
Biotechnology, Prathyusha
Engineering College,
Chennai.

#### **ABSTRACT**

The present study on the nutritive potential of the methanolic extracts of *Sargassum wightii* on the growth of the larvae of Bombyx mori is found to be the first report. A dose dependent influence of the extract of the sea weed on the growth of silk worm larvae was revealed from this study. This suggests that the sea weed extract of *Sargassum wightii* is a good neutraceutical agent to promote the growth of *B.mori*. The contents of the extract might have influenced the palatability of mulberry leaves thereby increasing the digestive potential and metabolic process of *B.mori*. Form this result it is suggested that *Sargassum wightii* extract can be given as feed supplementing neutraceutical to *B.mori*.

**KEYWORDS:** Sericulture; *Bombyx mori*; Sea weed; Neutrceutical; *Sargassum wightii*; Marine algae.

### 1. INTRODUCTION

In integrated farming system sericulture is an important component; which is an agro-based rural industry, with tremendous potential for employment generation in rural areas. It provides not only periodical return within a short period of time but also assures potential for family employment opportunities round the year (Jeyapaul et al., 2003). The main outcome from sericulture is the silk for fashionable clothing's. The silk is the conversion of the nutrients present in the mulberry leaves which are the role source of food for silk worm *Bombyx* mori L. Nutrition is known to play a key role in the larval growth, development and also an the manifestation of economic characters of cocoon. The rate of food consumption and leaf quality influence significantly larval growth rate, weight gain and

probability of survival (Amala Rani et al., 2011). The feeding of nutritionally enriched leaves showed better growth and development of silk worm larvae compounds with valuable pharmaceutical potential, Sheeba et al., (2005). Sea weeds are believed to be the rich nutrient source for higher tropic levels. But the available literature revealed that there is no work on the supplementation of sea weeds to enhance the growth of *B.mori*. So in the present study the seaweed *Sargassum wightii* which has been used as nutrient source for plants and aquaculture organisms was experimented to find out whether they can influence the growth of the larvae of *B.mori*.

#### 2. MATERIALS AND METHODS

#### Seaweeds collection

The marine brown algae *Sargassum wightii* was collected from China Muttom coast of Kanyakumari district, Tamilnadu, India. The seaweed was washed thoroughly with fresh water thrice to remove the extraneous materials and then it was shade dried for duration of 15 days. Then the seaweed was powdered in an electrical mixture and further subjected to various extraction processes.

#### Silkworm rearing method

#### Selection of larvae

For the present study 3<sup>rd</sup> instar stage of *B.mori* reared in a private farm at Pavoorchathiram, Tirunelveli district, Tamil Nadu, India was chosen. The healthy 3<sup>rd</sup> instar larvae were isolated and separated into 1 control group and 2 test groups. Each group contained 30 larvae. The rearing operations were carried out according to Krishnaswami et al., (1971). The control larvae were fed with normal mulberry leaves alone. The experimental groups (T<sub>1</sub> & T<sub>2</sub>) were fed with mulberry leaves treated with the extract of the sea weed *Sargassum wightii* at a concentration of 10% & 20% respectively. The larvae were fed with sea weed extract from 3<sup>rd</sup> instar stage to 5<sup>th</sup> instar stage.

#### Preparation of Sea weeds extract

The marine brown algae *Sargassum wightii* was collected from Chinna Muttom coast of Kanyakumari district, Tamil Nadu, India. The sea weed was washed thoroughly with fresh water thrice to remove the extraneous materials and then it was shade dried for duration of 15 days. Then the sea weed was powdered in an electrical mixture and further subjected to various extraction process (Fig.1).

The powdered sea weed was subjected to percolation by soaking in methanol (1:5 ratio) for a duration of seven days. After seven days of incubation, the filtrate was concentrated separately in a rotary vacuum evaporator and then the solvent free residue was collected and stored in plastic containers and this served as the methanolic extract.

# Application of sea weed extract on mulberry leaves

Fresh MR<sup>2</sup> mulberry leaves were collected and separately treated with 10% and 20% sea weed extract. The leaves were separately soaked with each concentration for 15minutes and then dried in air for 10 minutes. The larvae were fed 3 times a day and the larval weight was recorded for control and experimental groups. The total amount of protein in the entire body of control and sea weed *Sargassum witghtii* extract treated worms (4<sup>th</sup> day of 3<sup>rd</sup>, 4<sup>th</sup> and 5<sup>th</sup> instar) were estimated using the method of Lowry et al., (1951).

#### 3. RESULTS

The growth of the control and sea weed extract group larvae treated with 10% and 20% of the extract were monitored from 3<sup>rd</sup> instar stage to 5<sup>th</sup> instar stage. The difference in weight gain/reduction between control and sea seed treated groups are presented in (Table 1). The mean weight of the larvae of control and experimental groups T1 and T2 were  $13.22 \pm 0.14$ g (30 worms)  $12.67 \pm 0.11g$  (30 worms) and  $12.08 \pm 0.02g$  (30 worms) respectively on the first day of experiment (Table 1, Fig 2.). After 3 days the mean weight of extract given groups were comparatively lesser than the control group. But from the 4<sup>th</sup> day onwards the weight of the treated groups was more than the weight of the control group. On the 6<sup>th</sup> day of  $3^{rd}$  instar the weight of the larvae in the control group was  $26.62 \pm 0.36$  g. but in the 10% and 20% Sargassum extract supplemented larvae the mean weight on the  $6^{th}$  day of  $3^{rd}$ instar larvae were  $28.36 \pm 0.16$  g and  $33.64 \pm 0.27$ g. When compared with control, the Sargassum extract fed larvae showed a higher growth rate (than the mulberry leaf alone fed larvae). In the control group the growth difference between 1<sup>st</sup> and 6<sup>th</sup> day of 3<sup>rd</sup> instar larvae was 101.36 % high. But in the Sargassum extract treated larvae the weight gain was 123.83% in 10% treated group and 150.16% in 20% Sargassum extract treated group. The result indicates the growth promoting nutritive potential of Sargassum extract, had promoted the growth higher than the mulberry leaf alone fed  $3^{rd}$  instar larvae of *B.mori* (P < 0.01) (Table 1, Fig.2).

In the 4<sup>th</sup> insatr stage both the control and treated larvae consumed the mulberry leaves but the larvae consumed the mulberry leaves coated with *Sargassum* extract voraciously. This is

reflected in the % of weight gain in the entire of  $4^{th}$  instar phase (Table 2). The weight gain on the  $1^{st}$  day of the  $4^{th}$  instar in the control group was 110.28%. But it was170.45% in 20% treated group. On the  $5^{th}$  day of  $4^{th}$  instar stage the percentage of weight gain was 264.97 in the control larvae and 296.60% in the 10% treated larvae and 342.88% in the 20% treated larvae. The percentage weight gain was almost tenfold increase from the first day (Table 2) (Fig.3). The weight gain in 20% *Sargassum* extract treatment was 90% higher than the control and it is significant at  $P \le 0.05$  level. This observation further confirms that the extracts of the sea weed enhance the metabolism and growth in the larvae of *Bombyx mori*.

In the present study the weight gain in the 5<sup>th</sup> instar larvae of *B.mori* treated with sea weed extract was also monitored. In the present observation due to high environmental temperature in the ambience the duration of the 5<sup>th</sup> instar larval stage was reduced to 4 days. Even during these 4 days of growth phase in the 5<sup>th</sup> instar of the larvae, a progressive increase in weight was observed up to the 3<sup>rd</sup> day in control and sea weed extract treated group. On the 4<sup>th</sup> day of 5<sup>th</sup> instar larvae, a reduction in weight was observed in control and sea weed extract treated groups (Table 3) (Fig.4).

The weight gain on the first day and third day of 5<sup>th</sup> instar was 264.67 and 313.01 on the control larvae. Whereas the weight gain on the first day and third days of 10% treatment was 308.45g and 363.30g. Likewise the weight gain of larvae treated with 20% of *Sargassum* extract on the 1<sup>st</sup> day and 3<sup>rd</sup> day was 387.42g and 418.79g respectively The weight gain of larvae clearly indicates that the methanolic extract of the sea weed *Sargassum* has some bioactive compounds to enhance the growth of the larvae of *Bombyx mori*.



Fig 1: Experimental seaweed – Sargassum wightii.

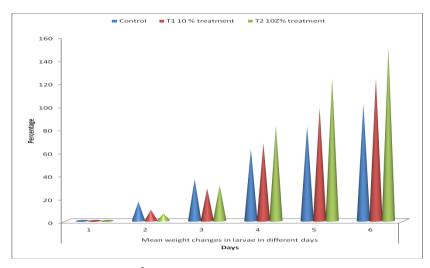


Fig 2: Weight changes in the  $3^{rd}$  instars larvae of *Bombyx mori* treated with Seaweed extract supplemented mulberry leaves.

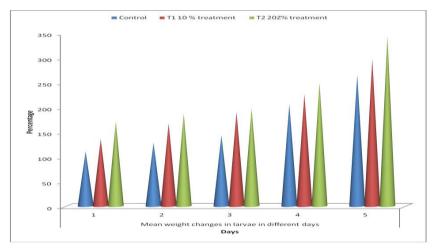


Fig 3: Weight changes in the 4<sup>th</sup> instars larvae of *Bombyx mori* treated with seaweed extract supplemented mulberry leaves.

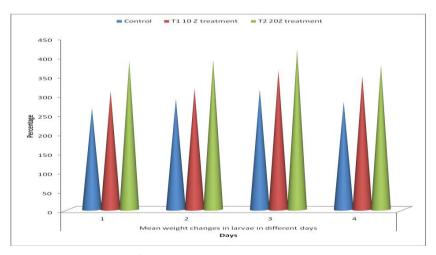


Fig 4: Weight changes in the 5<sup>th</sup> instars larvae of *Bombyx mori* treated with Seaweed extract supplemented mulberry leaves.

Table 1: Weight changes in the  $3^{rd}$  instars larvae of *Bombyx mori* treated with Seaweed extract supplemented mulberry leaves (Value represent mean of 3 groups – each group with 30 larvae).

Group	Mean weight changes in larvae in different days							
	1	2	3	4	5	6		
Control	$13.22 \pm 0.14$	$15.42 \pm 0.36$	$18.02 \pm 0.16$	$21.46 \pm 0.16$	$24.08 \pm 0.41$	$26.62 \pm 0.36$		
	$15.22 \pm 0.14$	(16.64)	(36.30)	(62.32)	(82.14)	(101.36)		
T1 10 %	12.67 ± 0.11	$13.87 \pm 0.1$	$16.22 \pm 0.5$	$21.22 \pm 0.17$	25.04 ±0.01	$28.36 \pm 0.16$		
treatment	$12.07 \pm 0.11$	(9.47)	(28.01)	(67.48)	(97.63)	(123.83)		
T2 10Z%	$12.08 \pm 0.02$	$12.84 \pm 0.13$	$15.82 \pm 0.13$	$22.04 \pm 0.18$	30.017±0.31	$33.64 \pm 0.27$		
treatment	$12.06 \pm 0.02$	(6.29)	(30.96)	(82.45)	(123.59)	(150.16)		

Percentage of weight changes over the 1<sup>st</sup> day of larval weight is given in parenthesis.

Anova: Two-Factor Without Replication

SUMMARY	Count	Sum	Average	Variance
Row 1	6	118.82	19.80333	26.64711
Row 2	6	117.38	19.56333	40.24595
Row 3	6	126.437	21.07283	83.02201
Column 1	3	37.97	12.65667	0.325033
Column 2	3	42.13	14.04333	1.686633
Column 3	3	50.06	16.68667	1.373333
Column 4	3	64.72	21.57333	0.177733
Column 5	3	79.137	26.379	10.15668
Column 6	3	88.62	29.54	13.3644

ANOVA								
Source of Variation	SS	Df	MS	$oldsymbol{F}$	P-value	F crit		
Rows	7.895641	2	3.94782	0.853177	0.454875	4.102821		
Columns	703.3033	5	140.6607	30.39866	9.71E-06	3.325835		
Error	46.27199	10	4.627199					
Total	757.471	17						

Table 2: Weight changes in the 4<sup>th</sup> instars larvae of *Bombyx mori* treated with seaweed extract supplemented mulberry leaves (Value represent mean of 3 groups each group with 30 larvae).

Croun	Mean weight changes in larvae in different days							
Group	1	2	3	4	5			
Control	$27.80 \pm 0.14$	$30.12 \pm 0.21$	$34.23 \pm 0.57$	$40.41 \pm 0.62$	$48.21 \pm 0.82$			
Control	(110.28)	(127.83)	(142.7)	(205.67)	(264.97)			
T1 10 %	$29.84 \pm 0.16$	$33.80 \pm 0.21$	$36.68 \pm 0.16$	$41.25 \pm 0.20$	$50.28 \pm 0.24$			
treatment	(135.52)	(166.77)	(189.50)	(225.57)	(296.60)			
T2 20Z%	$32.67 \pm 0.30$	$34.63 \pm 0.24$	$35.87 \pm 0.40$	$42.15 \pm 0.30$	53.5±0.24			
treatment	(170.45)	(186.67)	(196.94)	(248.92)	(342.88)			

Percentage of weight changes over the 1<sup>st</sup> day of larval weight is given in parenthesis.

**ANOVAs:** Two-Factor Without Replication.

SUMMARY	Count	Sum	Average	Variance		
Row 1	5	180.77	36.154	68.34023		
Row 2	5	191.85	38.37	61.6611		
Row 3	5	198.82	39.764	71.55418		
Column 1	3	90.31	30.10333	5.981233		
Column 2	3	98.55	32.85	5.7619		
Column 3	3	106.78	35.59333	1.558033		
Column 4	3	123.81	41.27	0.7572		
Column 5	3	151.99	50.66333	7.106233		
ANOVA						
Source of Variation	SS	df	MS	$oldsymbol{F}$	P-value	F crit
Rows	33.14332	2	16.57166	14.43229	0.002218	4.45897
Columns	797.0362	4	199.259	173.5351	8.35E-08	3.837853
Error	9.18588	8	1.148235			
Total	839.3654	14				

Table 3: Weight changes in the 5<sup>th</sup> instars larvae of *Bombyx mori* treated with Seaweed extract supplemented mulberry leaves (Value represent mean of 3 groups each group with 30 larvae).

Crown	Mean weight changes in larvae in different days						
Group	1 2		3	4			
Control	$50.40 \pm 0.47$	$51.38 \pm 0.21$	$54.60 \pm 0.32$	$50.36 \pm 0.78$			
Control	(264.67)	(288.65)	(313.01)	(280.93)			
T1 10 Z	$51.75 \pm 0.40$	$52.65 \pm 0.30$	$58.70 \pm 0.24$	$56.68 \pm 0.18$			
treatment	(308.45)	(315.55)	(363.30)	(347.36)			
T2 20Z	$58.88 \pm 0.20$	$60.12 \pm 0.24$	$62.67 \pm 0.30$	$57.70 \pm 0.40$			
treatment	(387.42)	(391.72)	(418.79)	(377.65)			

Percentage of weight changes over the 1<sup>st</sup> day of larval weight is given in parenthesis.

**ANOVAs:** Two-Factor Without Replication

SUMMARY	UMMARY   Count   Sum		Average	Variance
Row 1	4	206.74	51.685	3.999033
Row 2	4	219.78	54.945	10.86177
Row 3	4	239.37	59.8425	4.529492
Column 1	3	161.03	53.67667	20.76163
Column 2	3	164.15	54.71667	22.30023
Column 3	3	175.97	58.65667	16.28263
Column 4	3	164.74	54.91333	15.80973

ANOVA							
Source of Variation	SS	df	MS	$\boldsymbol{F}$	P-value	F crit	
Rows	134.8772	2	67.43861	26.22157	0.001082	5.143253	
Columns	42.73963	3	14.24654	5.53936	0.03653	4.757063	
Error	15.43125	6	2.571875				
Total	193.0481	11					

#### 4. DISCUSSION

Sea weeds are reported to be a good source of nutrients (Victoria Rani and Evangeline 2015 Kim 2011; Kim and Chojnacka, 2015; Sivasankari et al., 2006; Linkakumar et al., 2006; and Dhargalkar and Pereina, 2005). A study on the available literature showed that there is no work to evaluate the nutritive potential of the extracts of the sea weed *Sargassum* on the growth of *B.mori*.

There are many reports on the nutritive efficiency of different sea weeds to promote the growth of the plants including mulberry plant (Xavier et al., 2007; Ramamurthy and Sujatha, 2007; Jothi Nayaki, 2009; Victoria Rani and Evangeline, 2015). Ramamoorthy et al., (2016) and Sararapu et al., (2017) reported that the extract of *Sargassum wightii* was active against microbial pathogens of *B.mori* and inhibits the microbial growth according to its concentration.

The present study on the nutritive potential of the methanolic extracts of *Sargassum wightii* on the growth of the larvae of *Bombyx mori* is found to be the first report. A dose dependent influence of the extract of the sea weed on the growth of silk worm larvae was revealed from this study. This suggests that the sea weed extract of *Sargassum wightii* is a good neutraceutical agent to promote the growth of *B.mori*. The contents of the extract might have influenced the palatability of mulberry leaves thereby increasing the digestive potential and metabolic process of *B.mori*. Form this result it is suggested that *Sargassum wightii* extract can be given as feed supplementing neutraceutical to *B.mori*.

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