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Assessment of Growth and Performance of Silk Worms (Bombyx mori L.) on Mulberry Leaves at Jimma, South West Ethiopia

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Abstract

Silkworm (Bombyx mori L.) is essentially monophagous insect feeds solely on mulberry leaves (morus spp.). For performance study seven mulberry silkworm races/hybrids were evaluated at silkworm rearing laboratory at Melko-JARC, six of them are bivoltine and one race was multi-boltine. For bivoltine race eggs were treated with 37% HCl of 1:3 ratios to break the diapauses stage. The objective of the study was to compare larval duration, number of larva left after each molting, cocoon, pupa and shell weight and shell to cocoon ratio among the seven mulberry silk worm races. When comparing the mean weight of 25coocon among the silk worm races the largest was 30.8gm (Kenya 1) and the lowest was 19.7gm (Yellow).And the largest mean weight of 25shell observed was 6.9gm (Kenya 1 and Kenya 5), the lowest was 2.4gm (Yellow). For both weight of 25 cocoons and 25 pupa yellow silk worms race was significantly difference among the rest silk worm races ($p < 0.05$). For 25 shell weight of yellow silk worm race was significantly difference among the rest silk worm races and there is also significance difference between China 2 and Kenya 5 silk worm races. There is also significance of hatchability ($p<0.05$).

Keywords: Bivoltine, *Bombyx mori L.,* cocoon, Jimma, mulberry leaf, shell, Silk Worms

Introduction

Sericulture is an agro-based rural industry having tremendous employment potential and foreign exchange earnings (Bothikar *et al*., 2014). It is the rearing of silkworm for the production of raw silk originated in China between 2600 and 2700 BC (Rahmathulla, 2012). Silk is called the queen of textiles due to its glittering luster, softness, elegance and durability. It is a very costly fiber, produced by silk worms (Borisade, 2012). Silkworm is one of the most important domesticated insects where the growth and development is greatly influenced by environmental conditions. Success of silkworm breeds/hybrids largely depends on their adaptability to the environment in which it is destined to be reared. The biological as well as cocoon-related characters are influenced by ambient temperature, rearing seasons, quality mulberry leaf, and genetic constitution of silkworm strains. It is a well established fact that under tropical conditions, unlike polyvoltines, bivoltines are more vulnerable to various stress like hot climatic conditions of tropics, poor leaf quality, and improper management of silkworm crop during summer that is not conducive for bivoltine rearing for technologically and economically poor farmers of India (Suresh *et al.*, 2001; Lakshmi, 2007 and Begum *et al.,* 2008).

Silkworm (*Bombyx mori L.*) is essentially monophagous insect feeds solely on mulberry leaves (morus spp.). Its growth and development as well as cocoon and silk production entirely depends upon the quantity and quality of mulberry leaves (Nagaraju, 2002). The success of sericulture industry depends upon several factors of which the impact of the environmental factors such as biotic and abiotic factors is of vital importance. Among the abiotic factors, temperature plays a major role on growth and productivity of silkworm, as it is a poikilothermic (cold blooded) insect (Benchamin and Jolly, 1986). Silk played an important role in the social and religious life of Ethiopia from the earliest days of the Kingdom of Axum. The silk was imported in large quantities from India, Arabia and China and stored in vast caverns in the central highlands of Ethiopia (Metaferia *et al*., 2007).

In fact introduction of mulberry silkworm into Ethiopia dates back to 1930s by Italian and its activities went into recession shortly. Since then, mulberry spread all over the country and is seen growing in altitude of 1500m to 2,500m in the country. In 1970's some Ethiopian scientists were getting interested in mulberry silkworm and they conducted some research works until 1980s. Since then, very few research works have been made due to low priority given by Ethiopian Government (Metaferia *et al.*, 2006). As a result, failure of timely introduction of adequate technologies, a poor capacity building, lack of integrated and effective extension services, and lack of parent stock of silkworm eggs contributed a lot for the less impact around stakeholders. Therefore, feed plants cultivation and silkworm rearing is practiced by a very small number of users and reeling is performed in domestically prepared handloom to reel coarse silk yarn that is not suitable for the production of quality fabrics. Also in case no parent stock of silkworm eggs are domestically produced and as far as it relies on external supply sources, not only a high quality silkworm egg is hard to be ensured but also it is difficult to meet consumer's requirement in terms of timely delivery.

The objectives of the study were to assess growth and development of silk worms (*Bombyx mori l.*) on mulberry leaves. To select adaptable, high yielding and pest resistant mulberry silkworm races with acceptable silk parameters. To evaluate six different bivoltine and one multi-voltine mulberry feeding silkworm breeds under Jimma conditions. And to compare larval duration, number of larva left after each molting, cocoon, pupa and shell weight and shell to cocoon ratio among the seven mulberry silk worm races.

Materials and Methods

Study Site

Mulberry plants grown in garden and silkworm rearing experiments were conducted in rearing house at Jimma Agricultural Research Center (JARC), Jimma, Ethiopia. Silkworm rearing laboratory at JARC for performance study seven mulberry silkworm races/hybrids were evaluated, six of them are bivoltine and one race was multi-boltine. Mulberry plant was raised simultaneously to serve as feed for the silkworms. For bivoltine race eggs were treated with 37% HCl of 1:3 ratios to break the diapauses stage. Twenty-four hours prior to hatching, eggs were covered with paper to stimulate uniform embryonic growth. Eggs were exposed to light for 30 minutes on the day of hatching. Upon hatching young age instars (1^{st} -3^{rd}) were fed with young shoots chopped to the size appropriate for growing larvae. Whereas late age instars (4^{th} –5^{th}) were fed with medium to mature leaves. Young age larvae were fed with tender, succulent and nutritious leaves which are known to favour the growth and development of silkworms, while mature and coarse leaves were fed to larvae when they grow till ripening. Normal daily feedings of four times per day (at 8:00 AM, 11:00 AM, 2:00 PM and 5:00 PM) were given for each silkworm race. Rearing beds were cleaned every day before 1^{st} feeding. The room temperature and relative humidity (RH) were maintained based on recommendations. Mountages were arranged timely for matured worms. Cocoon yield were harvested after seventh day from mounting.

Completely Randomized Design /CRD/ with three replications were employed for this experiment. Necessary data were collected during the study period which includes number of larvae left after each molting stage under observation (at 1^{st} – 4^{th} instar), total number of larvae reach full maturity, weight of ten matured larvae (gm) at 5^{th} instars at 6 days of age, developmental period (egg, larvae, pupae & adult longevity), date of mounting, date of harvesting, fresh weight of twenty-five cocoons (gm), fresh weight of shells (gm), silk shell to cocoon ratio (%), no. of eggs/ three female adult moth (fecundity), first date of hatching, last date of hatching.

Study Material

Silkworm rearing was conducted following the standard method under natural conditions and also at recommended temperature and humidity conditions wooden compensator made mountages was used for mounting the ripened larvae. After 48 hours of mounting, when the larvae formed hammock, the mountages were turned upside down. Cocoon harvesting was carried out on the 7^{th} day of spinning. The cocoons were deflossed and the defective ones were sorted out. Assessment was carried out on the subsequent day. The survival rate was calculated as the number of live pupae to the number of larvae treated.

Parameters Studied:

Various quantitative traits such as fecundity, cocoon weight, shell weight, silk shell to cocoon ratio, larval weight, number of larva at different stage, and larval duration were calculated. The characteristic features of the selected breeding resource materials are given below.

Silkworm breeds

The six bivoltine and one multivoltine mulberry silkworm races the races were obtained from Melkasa Agricultural Research Center (MARC).The races used were bivoltin (China-2, Keneya– 1, Keneya-3, Keneya-5, Korea-1 and Korea- 3) and multivoltine (yellow).

Silkworm rearing and estimation of genetic traits

Disease free eggs from each strain were reared and cocoons were harvested and maintained until eclosion of moths. Healthy female moths emerging on the peak day of eclosion were allowed to mate for three to four hours and held until oviposition. The bivoltins' eggs were acid treated within 20 hours after oviposition, following the method developed by Yokoyama (1962) to prevent hibernation. The whole process, from silkworm egg incubation to completion of rearing activities, was carried out under hygienic conditions in a silkworm rearing laboratory thoroughly disinfected with bleaching powder and formalin solution.

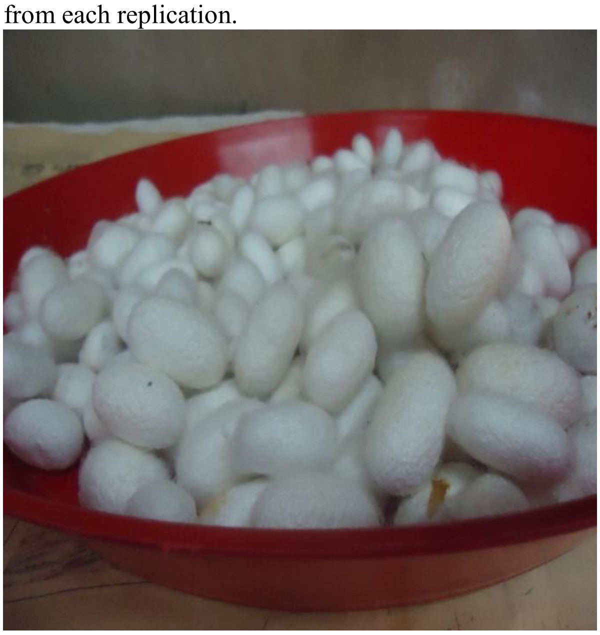
Two hundred worms of each race treated as one treatment and were kept in uniform size wooden tray. Silkworm rearing was conducted for each breed by feeding them the same variety of mulberry leaves from the well-maintained irrigated mulberry garden at JARC. A standard rearing procedure was adopted as recommended by Datta (1992). The young larvae (l^{st}–3^{rd} instars) were reared at 26–28° C with 80–90% RH and late age larvae (4^{th} and 5^{th} instars) were maintained at 24–26° C with 70–80% RH until the 3rd day of fifth instar. Each group was divided into three replication, under standard rearing conditions. Fecundity of each race was computed by taking average number of eggs laid by single female moth of each race.

5th instar larva Larva of China-2

Cocoons were harvested 7–8 days later after completion of cocoon spinning. Harvested cocoons were accessed for survival to pupation. During the process of silkworm rearing, data on larvae and cocoons for the seven silk worm races (larval weight, cocoon, pupa and shell weight, and shell ratio) were collected and calculated according to the equations below:

Larval weight (gm): Mean larval weight (g) recorded for 10 randomly selected larvae at the peak of growth of fifth instar larvae from each replication. This was an indicator of the general health of the larvae.

Cocoon weight (gm): The Mean 25 cocoon weight in grams randomly chosen on the 7th or 8th day of spinning from each replication.



Cocoon Mountage for cocoon production

Shell weight (gm): The Mean 25 shell weight in grams shell randomly chosen from each replication. The shells used were the same cocoons used for the cocoon weight determination.

Pupa weight (g): The Mean 25 pupa weight in grams calculated by subtracting shell weight from cocoon weight for each replication.

Pupa weight = cocoon weight – shell weight

Shell ratio (%): The total quantity of silk available from 25 cocoons was expressed as a percentage using the following equation for each replication.

$$\textbf{Shell ratio (\%)} = \frac{\text{single cocoon shell weight (g)}}{\text{single cocoon weight (g)}} * 100$$

Silkworm Moulting: Molting test was carried out up to 4th molt following standard rearing methods with three replications and 200 larvae/replication. Tender leaves (1st-4th order) on healthy shoots were harvested and fed to young age silkworm larvae up to second molt (Benchamin and Nagaraj, 1987). Silkworm rearing was conducted under standard conditions (Krishnaswami, 1986 & 1990). First appearance of one larva out of molt was considered as commencement of molting (Benchamin and Anantharaman, 1990).

Statistical Analysis
Data collected on various parameters were tabulated using MS excel 2007 and subjected to critical statistical analysis. Statistical analysis was done with assistance of SPSS ver. 20.0 software for understanding the significance of the study by ANOVA. For significant ANOVA post hoc was checked using mean separation. $P < 0.05$ and 95% Confidence interval (CI) were considered significant during the analysis.

Results and Discussion
The Mean of 10 larval weights at 5th instar was varied in the range of 116.60gm to 38.87 gm. The performance of Kenya -3 (116.60gm) for mean 10 larval weight was observed significantly superior over the rest of races followed by Korea - 3 (101.92gm). The lowest mean of 10 larval weight was recorded in Yellow races (38.87gm) as shown in Table 1 below.

Larval duration (Days) the data on the larval duration of silkworm *Bombyx mori L*. are presented in Table 2. Larval duration was observed in the range of 24 days to 21days. In all bivoltine varieties (21 - 23 days) and multivoltine (yellow) race 23-24 days larval duration were recorded (from 1st instar to mountage stage).It is similar to work done by Pakhale, (2014) in India that is larval duration of mulberry silk worms observed was in the range of 22.13 days to 23.27 days. Daily mean temperature and relative humidity of rearing room during larval duration was 20.77^0c and 72.18% respectively during this rearing time. The mean range temperature and relative humidity were 19.33 - 22.0^0c and 64.00 – 75.33% respectively. Work was carried out to evaluate rearing performance of silkworm (Gawade, 2006).

Table 1. Mean parameters along the life cycle of mulberry silk worm races at Melko-JARC silk worm rearing house, (Sept – November, 2015)

Mulberry silk worm races	Hatch ability (%)	No of larvae after 1st molt/out of 200	No of larvae after 2nd molt	No of larvae after 3nd molt	No of larvae after 4th molt	No of larvae reach full maturity
China - 2	70	194	174	174	170	162
Kenya - 1	76	200	198	185	178	177
Kenya - 3	76	188	180	176	174	172
Kenya -5	76	190	174	163	167	154
Korea -1	90	196	168	152	154	152
Korea -3	89	191	175	160	170	159
Yellow	85	195	185	167	153	152

Table 2. Mean of parameters and rearing performance of mulberry silkworms feeding the some mulberry leaves at Melko-JARC silk worm rearing house condition, (July – Sep. 2015).

Mulberry silk worm races	wt of 10 5th instars larvae (gm)	Larval life /Feeding duration (days)	No of days take to harvest cocoon	wt of 25 cocoon (gm)	wt of 25 pupa (gm)	wt of 25 shell (gm)	wt of 25 shell to 25 cocoon ratio , %	No of eggs per female moth / Fecundity
China 2	97.93	21-22	53	34.6	29.1	5.5	16	306
Kenya 1	97.70	21-22	46	37.7	30.8	6.9	18	297
Kenya 3	116.60	21-22	48	36.1	30	6.1	17	341
Kenya 5	63.47	21-22	48	34.9	28	6.9	20	330
Korea 1	95.68	21-22	49	33.1	27.5	5.6	17	407
Korea 3	101.92	21-22	50	35.4	29.3	6.1	17	310
Yellow	38.87	23-24	39	19.7	17.3	2.4	12	305

As showed in table 2 the longest time in days taken for the silk worms to harvest cocoon was 53days (china 1) and the shortest was 39days for Yellow silk worm races. Concerning hatchability the highest percentage observed was 90% (Korea 1) and 89% (Korea 3) while the lowest was 70% (China 2) the rest Kenya 1, Kenya 3 and Kenya 5 were 76%. A single female moth can lay an average of maximum 407eggs Korea-1 race and a minimum of 297eggs Kenya-1 race was observed. That is, there is also no significant difference of fecundity among the silk worm races p= 0.635.

Table3: Summary of GLM multivariate analysis of variance of the seven mulberry silk worm races at Melko-JARC (July – Sep. 2015).

Dependent Variable	df	Mean Square	F	P-value
Hatchability	6	5.714	0.000	0.000*
First molt	6	0.667	3.500	0.025*
No of larva after first molt	6	0.000	1.8920	0.00*
Second molt	6	4.714	99.000	0.000*
No of larva after second molt	6	185.746	1.340	0.304
Third Molt	6	2.111	22.167	0.000*
No of larva after third molt	6	1138.556	1.877	0.156
Forth molt	6	0.429	0.000	0.000*
No of larvae after 4^{th} molt	6	799.651	1.246	0.342
Mean wt 10 larvae at maturity	6	1552.753	9.512	0.000*
Montage duration	6	0.000	0.000	0.000*
No of larvae reach full maturity	6	2845.413	10.827	0.000*
wt of 25 cocoon	6	79.847	16.399	0.000*
wt of 25 pupa	6	50.767	19.289	0.000*
wt of 25 shell	6	4.134	8.708	0.000*
Mean No of eggs per female moth (Fecundity)	6	1981.030	0.727	0.635

* Significant at $p < 0.05$

Table 3 shows that significance of GLM multi comparison of parameters among the seven mulberry silk worm races. Dependent variables with $P < 0.05$ were significance while $P > 0.05$ was not significance. And for some significance parameters further post hoc test was done table 4 below.

Table 4. Summary of LSD multiple comparison of post hoc tests

Dependent Variables	Mulberry silk worm races	Mean	Mean $\pm$ SE	95% CI
weight of 25 cocoon (gm)	china2	34.6a	34.6$\pm$ 1.802	(36.402, 34.798)
	Kenya 1	37.7a	37.7$\pm$ 1.802	(39.502, 35.898)
	kenya3	36.1a	36.1$\pm$ 1.802	(37.902, 34.298)
	kenya5	34.9a	34.9$\pm$ 1.802	(36.702, 33.098)
	korea1	33.1a	33.1$\pm$ 1.802	(34.902, 31.298)
	korea3	35.4a	35.4$\pm$ 1.802	(37.202, 33.598)
	yellow	19.7b	19.7$\pm$ 1.802	(21.502, 17.898)
weight of 25 pupa (gm)	china2	29.1a	29.1 $\pm$1.325	(30.425, 27.775)
	Kenya 1	30.8a	30.8 $\pm$1.325	(32.125, 29.475)
	kenya3	30a	30 $\pm$1.325	(31.325, 28.675)
	kenya5	28a	28 $\pm$1.325	(29.325, 26.675)
	korea1	27.5a	27.5 $\pm$1.325	(28.825, 26.175)
	korea3	29.3a	29.3 $\pm$1.325	(30.625, 27.975)
	yellow	17.3b	17.3 $\pm$1.325	(18.625, 15.975)
weight of 25 shell (gm)	china2	5.5a	5.5 $\pm$0.563	(6.063, 4.937)
	Kenya 1	6.9ab	6.9 $\pm$0.563	(7.463, 6.337)
	kenya3	6.1ab	6.1 $\pm$0.563	(6.663, 5.537)
	kenya5	6.9b	6.9 $\pm$0.563	(7.463, 6.337)
	korea1	5.6ab	5.6 $\pm$0.563	(6.163, 5.037)
	korea3	6.1ab	6.1 $\pm$0.563	(6.663, 5.537)
	yellow	2.4c	2.4 $\pm$0 .563	(2.963, 1.837)

Means with the same letter(s) in the same column are not significantly different from each other at $P < 0.05$

As shown in table 4 above mean separation of 25 cocoons, 25 pupa and 25 shell weight in gm, (post hoc tests) among the seven mulberry silk worm races. For both weight of 25 cocoons and 25 pupa yellow silk worms race was significantly difference among the rest silk worm races ($p < 0.05$). for weight of 25 shell yellow silk worm race was significantly difference among the rest silk worm races and there is also significance difference between China 2 and Kenya 5 silk worm races.

The mean of 25 cocoon weight was recorded in the range of 37.70gm to 19.70 gm. The highest was recorded in the race Kenya - 1 (37.70 gm) over the rest of the races tested. Whereas, the lowest mean of 25 cocoon weight was recorded in the Yellow race (19.70gm). There is significant of cocoon weight ($P = 0.000$).

The mean of 25 shell weight was recorded in the range of 6.90 gm to 2.40 gm. The highest mean of 25 shell weight was recorded in the races Kenya – 1 and Kenya - 5 (6.90gm) over the rest of the races tested. When comparing the cocoon to shell ratio between Kenya – 1 and Kenya - 5 Kenya – 5 is 20% while Kenya -1 was 18 % (Table 2).Whereas, the lowest mean of 25 shell weight was recorded in yellow race (2.40 gm). There is significant of shell weight at $P < 0.05$.

Cocoon weight and shell weight are the most important characters evaluated for productivity (Gaviria *et al.*, 2006). Shell weight percentage indicates the amount of raw silk can be reeled from the given quantity of fresh cocoons and shell weight percentage varies according to age and breed of silkworm. The study showed that there is significance of cocoon weight, shell weight and pupa weight among those mulberry silk worm races ($P=0.000$). Yellow mulberry silk worm race was significantly difference among the rest silk worm races in cocoon weight, shell weight and pupa weight and there was also significance difference between China 2 and Kenya 5 silk worm races. According to Bothikar *et al*. (2014) suggestion to increase cocoon production and to reduce labour cost, it is advisable to choose silkworm strains and mulberry variety which is suitable for particular set of a condition hence, the investigations were made to evaluate superior variety by further evaluation.

As indicated in Table 2 mean of 25 shell weight to 25 cocoon weight ratio percentage the maximum was 20% (Kenya-5) and the minimum was 12% in Yellow race, the bivoltine races have in a range of 20% to 16%. Similar result was reported by Bothikar *et al*. (2014) the shell cocoon percentage of mulberry silk worm races varies between 19.93 to 17.84 %.

Conclusion and Recommendation

The results obtained from the study shows that the effectiveness bivoltine mulberry silk worm races for cocoon production. Generally those silk worm races perform well and have good promise to invest on this sector and the environment is good for mulberry plants. Sericulture program should be encouraged and supported by Regional, Federal governments of Ethiopia and other NGOs as a good opportunity for source of income generation and job creation for youth and women and poverty alleviation in rural, urban and semi-urban areas of the country. Being the sector is new in most areas of the country, farmers, extension workers, urban agriculture offices, urban youth and women have to be supported in terms of training and resources.

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