Evaluation of different strains of mulberry silkworms /*Bombyx mori* L. / for their adaptability and silk yield in Ethiopia

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Abstract

Mulberry silkworm, Bomyx mori L. is one of the silkworm's underutilization in Ethiopia. However, rearing of selected strains of this mulberry silkworm race that adapt to different agroecologies is very essential for improving silk cocoon quality, yield and profitability. In this experiment, four Kenyan (ICIPE) bivoltine races (Kenya-1, Kenya-3, Kenya-4 and Kenya-5), two Korean bivoltine races (Korea-1 and Korea-3), two Vietnamese multivoltine (Mult-yel and Mult- wh) mulberry silkworm strains were evaluated in different locations (Melkassa, Alagae, Wondo- Genet and Jimma) which represent different agro-ecologies of Ethiopia. The experiment was laid out in Completely Randomized Design (CRD) in three replications. Thus, different silkworm strains showed statistical significant silkworm characteristic ranges in different locations which include egg hatchability (63.67% to 91.00%), larval duration (21.67days to 32.00 days), total life cycle duration (44.94 days to 79.67 days), single weight of larva (1.328 grams to 3.567 grams), effective rate of rearing (56.22% to 92.0%), single cocoon weight (0.726 grams to 1.600 grams), single shell weight (0.108 grams to 0.355 grams) and silk ratio (14.71 to 22.76%). However, a bivoltine mulberry silkworm strain known by Kenya 1 (ICIPE1) have showed an outstanding performance compared to other strains in all locations especially in cocoon parameters. Therefore, it is recommended for future research and development efforts on mulberry sericulture in Ethiopia.

Key words: *mulberry, silkworm, strains, performance, cocoon*

Introduction

The silkworm, *Bombyx mori* L spins valuable silk fibre, making it one of the most beneficial insects to mankind, and is becoming an attractive multifunctional material for both textile and non-textile uses (Murthy *et al,* 2013).The practice of silk production involves diverse activities from the cultivation of host plants to silk processing, which engage people of all spectrums. Further, the by-products also find uses ranging from fertilizers in rural areas to pharmaceutical industries (Legay, 1958).Thus, silk production has the potential to make a significant contribution to the economy of many countries where there is surplus labor, low-costs of production and a willingness to adopt new technologies (Hajare *et al.*, 2007).

In Ethiopia, silk has strong attraction to the people starting from early period of Axum Kingdom. However, the silk yarns used were imported from India, Arabia and China **(**Spring and Hudson, 2002**).** Currently, Ethiopia is the second populous country in Africa after Nigeria. There is a general trend of increasing rate of unemployment in the country. Therefore, sericulture, which is an agro based labor intensive and environment friendly cottage industry, can be an efficient and effective agricultural endeavor for the country. The business holds a ray of hope at village level for Ethiopian citizen migrating to cities searching for jobs **(**Kedir *et al.*, 2014**)**. As a result, silk production from mulberry silkworm is practiced bits in bits in different parts of the country **(**Metaferia *et al.*, 2006**).**

However, rearing of superior silkworm strains that well adapt to the local environment is an important method for enhancing cocoon quality, increasing cocoon yield an improving economic benefit **(**Nguku *et al.,* 2009**)**. Differences in agro-ecologies across regions including significant distinctions in temperature and humidity require a type of silkworm strain which is both hyper silkgeneous and adversity resistant (Basavaraja *et al,* 2005). Rearing performance in silkworms is also affected by ecological, biochemical, physiological and quantitative characters, which influence growth and development, quantity and quality of silk they produce in different geographical locations (Virk *et al,* 2011; Ramesh *et al,* 2012; Anandakumar and Michael, 2012 and Reddy *et al,* 2012). The success in silkworm rearing depends on the various factors including successful implementation of technological and managerial tools along with high yielding and best-suited mulberry varieties and silkworm strains (Rajan and Himantharaj, 2005). In addition, the *B. mori* insect is an oligophagous herbivore and depends mainly on the quality of mulberry leaves and environmental conditions for its development (Murthy *et al,* 2007).

Performance of the strain itself in a given environment indicates its superiority. During evaluation, emphasis was given on the phenotypic expression of traits of economic importance under different temperature conditions. However, as the objective of the study was for greater viability and high productivity merits, equal importance was given on these two traits during selection of parents. The significant variations observed in the phenotypic manifestation for the traits analyzed can be attributed to the genetic constitution of the breeds and their degree of expression to which they are exposed during their rearing. Such variations in the manifestation of phenotypic traits of the breeds studied can be ascribed to the influence of environmental conditions. Variable gene frequencies at different loci make them to respond differently. The results are in line with the findings of Sudhakar *et al*, 2001.

Therefore, appropriate selection of the silkworms strains based on rearing performance and economic qualities in different climatic conditions is essential to select and exploit suitable silkworm strains for improved sericulture practices (Basavaraja *et al,* 2005 and Virk *et al,* 2011). On the other hand, there is no any recommended silkworm strain for mulberry silk production in Ethiopia. Hence, this experiment was initiated with objective of introducing and evaluating different mulberry silkworm strains for their adaptability, better yield and quality silk.

Materials and Methods

Description of the Study Areas

The experiment was conducted in four locations: Melkassa, Wondo-Genet and Jimma Agricultural Research Centers and Algae Agricultural Technical, Vocational, Educational and Training (ATVET) College, Ethiopia. These locations represent some of the agro-ecologies of the country assumed suitable for silkworm development and productivity.

Melkassa Agricultural Research Center (MARC), Central Rift Valley of Ethiopia, which is located at the distance of 15 km in the southeasterly direction from Adama town, which is situated at 8° 24' N latitude and 39^{o}21' E longitude with an altitude of 1550 meters above sea level (MoA, 2000). The main rainy season for this area is from June to September (Kiremt) which contributes about 69% of the total annual rainfall and the second short rainy season (Belg) is from March to May which brings nearly 24% of the precipitation. The third season, which is from October to January (Bega), is dry most of the time but contributes around 7% of the total annual rainfall especially during October and January for the late cessation of Kiremt and early onset of Belg seasons, respectively. For the period 1977-2006, the annual average rainfall was 702 mm and ranged from 450 to 918 mm. The peak months were July and August with an average rainfall of 157.5 and 161.6 mm, respectively. The long-term mean rainfall for the Bega, Belg and Kiremt seasons was 52, 166 and 482.5 mm, respectively. For the period 1977-2006, the daily mean maximum and minimum temperatures were 28.5 and 13.8 °C, respectively. The mean maximum temperature was between 30.9 °C during May and 26.2 °C during August (Gebru and Abebe, 2011). According to the recent agro-ecological classification of Ethiopia (MoA, 2000), the Melkassa Hypo Calcic Regosol ecotope falls in the zone termed hot to warm semiarid lowlands.

Jimma Research center (JARC) which is found at about 345 km from Addis Ababa in South west and lies between 36° 10' E longitude and 7° 40' N latitude. This area experiences annual average rainfall of 1000 mm for 8 to 10 months. The zone has an elevation ranging from 880 to 3360 masl. The temperature of Jimma zone varies from 8-28°C. The average annual temperature is 20°C (Haile A. and Tolemariam T, 2008).

Wondo-Genet Agricultural Research Center located at 7° 1*92’* N latitude and 38° 382' E longitudes with an altitude of 1780 m above mean sea level. The site receives a mean annual rainfall of 1000 mm with minimum and maximum temperatures of 10 and 30°C, respectively. The soil textural class is clay loam with an average pH of 7.2 (Tesfaye, 2005 - unpublished).

Lay out and rearing

As per the rearing recommendations of silkworms by Rajan and Himantharaj (2005), the silkworm rearing room and equipments were cleaned, washed and disinfected with 2% formalin solution at the rate of 800ml per 10m before the commencement of the experiment (rearing).Under this experiment, eight mulberry silkworm strains were considered as treatments. These include four Kenyan (ICIPE) bivoltine strains (Kenya-1, Kenya-3, Kenya-4 and Kenya-5), two Korean bivoltine strains (Korea-1 and Korea-3) and two Vietnamese multivoltine strains (Mult-yel and Mult-wh).The experiment was designed in a Completely Randomized Design (CRD) and the treatments were replicated three times. In each replication, 200 worms were used and allowed to complete their life cycle. Rearing was done in trays measuring 90 x 60cm, placed on rearing racks, 150 x 75 x 200cm that could hold 20 trays each and for every feeding tray equal amount of feeds from same feed plant variety were given. Mulberry *(Bomyx mori)* was cultivated and used as feed source for these silkworms. Tender leaves were fed four times a day until the larvae ends II instar stage and semi tender leaves to III instar larvae, while more matured leaves were fed to IV and V instar larvae.

Data Collection and Analysis

As adopted by Kedir *et al.* (2014), egg count was made before larval hatching. On the sixth day of spinning, the cocoons were harvested, counted and weighed. Data like larval and total life cycle duration (in days) and mature larval weight (in grams) were recorded. Cocoons from all strains were harvested from the mountages and then sorted. Cocoons were weighed using sensitive electronic balance after cutting open the cocoons using a blade to release the pupa and the moulted skin. Then, the cocoon weight (with pupa) and cocoon shell weight (without pupa) were documented. The following formulae were used for analysis of egg hatchability (%), effective rate of rearing (ERR %) and silk or shell ratio (%) calculations.

$$\textit{Egg hatchability to larva} = \frac{\textit{Number of normal eggs} - \textit{Number of non-hatched eggs}}{\textit{Number of normal eaas}}$$

$$\textit{Shell Ratio} = \frac{\textit{The cocoon shell}}{\textit{Weight of the whole cocoon}} \times 100$$

$$\textit{ERR} = \frac{\textit{Number of cocoon}}{\textit{Number of larvae brushed}} \times 100$$

Finally, data were analyzed using SAS software at 5% level of significance (SAS, 2000). Significant means ($p<0.05$) were separated using Least Significant Difference (LSD).

Results

The present study clearly depicted variations in respect of growth and cocoon characters of mulberry silkworm strains, *Bombyx mori.* Data on growth, rearing performance and cocoon traits of mulberry silkworm strains viz., egg hatchability (%), larval and total life cycle durations (days), larval weight (g), effective rate of rearing (%), cocoon weight (g), shell weight (g) and shell ratio (%) of different strains are illustrated below. In this experiment, significant differences were observed on silkworm characters among mulberry silkworm strains in different locations. Larval development periods and total life cycle durations variations were recorded among the different strains. The temperature and humidity records of the study areas are also presented in the table below (table 1).

Table 1. Temperature and humidity records of the study areas during experimental period

Study areas	Temperature			Humidity		
	Min	**Max**	**Mean**	**Min**	**Max**	**Mean**
Melkassa	19	34	27.6	25	76	58.1
Wondogenet	20	28	24.5	35	80	62.3
Jimma	18	32	25.8	41	83	67.6
Alage	22	32	27.1	30	78	60.4

Egg Hatchability

Egg hatchability of mulberry silkworm strains to larval stage was ranging from 63.67% to 91.0% (Table 2).

Table 2. Variations in egg hatchability among mulberry silkworm strains

Treatments	Melkassa	Alage	Wondo-Genet	Jimma
Kenya-1	84.997^{a}	88.333^{a}	86.556^{a}	84.000^{abc}
Kenya-3	63.670^{c}	87.667^{a}	83.889^{a}	77.000^{c}
Kenya-4	66.557^{bc}	88.553^{a}	84.222^{a}	76.000^{c}
Kenya-5	74.110^{b}	90.000^{a}	84.889^{a}	77.000^{c}
Korea-1	69.223^{bc}	90.500^{a}	87.333^{a}	89.000^{ba}
Korea-3	70.223_{bc}	86.333^{a}	85.000^{a}	91.000^{a}
Mult-yel	85.557^{a}	87.887^{a}	84.556^{a}	79.890^{bc}
Mult-wh	72.333^{bc}	85.777^{a}	86.222^{a}	85.813^{bac}
Pr	0.004	0.7204	0.4086	0.0445
CV	6.8563	3.99333	2.37206	7.045656

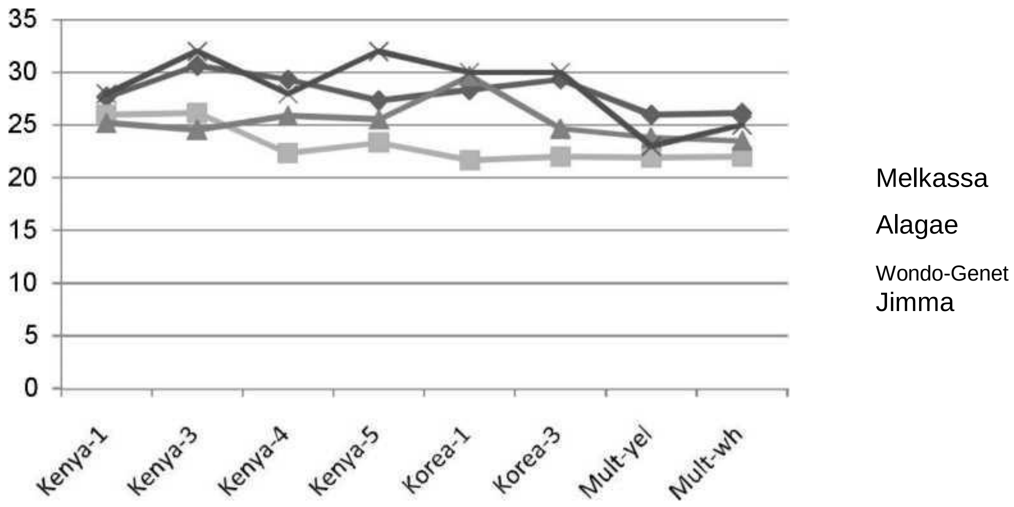
Means followed by the same letter within a column are not significantly different from each other at 5% level of probability.

Statistically significant variation in mean egg hatchability among mulberry silkworm strains was observed in Melkassa and Jimma areas. In Melkassa, Mult-yel and Kenya-1strains showed highest egg hatchability (85.557 % and 84.997 %, respectively) but Kenya-3 strain showed the least one (63.67%). In Jimma, the best egg hatchability was achieved from Korea-3 (91.0%) followed by Korea-1 (89.0 %), Mult-wh (85.81 %), Kenya-1 (84.0 %) and Mult-yel (79.89 %) but the least hatchability was recorded from Kenya-4 (76%) (Table 2).

Larval and total life cycle duration

Larval period and total life cycle duration of the strains revealed statistically significant difference among each other in all locations (Fig. 1). The shortest larval duration was noticed in Alagae from Mult-yel (21.89 days) and Mult-wh (22.0 days) strain. However, the longest larval duration was observed in Jimma from Kenya-5 (32.0 days) strain. In addition, the shortest total life cycle duration was seen in Alagae from Mult-yel (44.94 days) and Mult-wh (45.61 days) strains but the longest was in Jimma from Kenya-1 (79.67 days) strain. Generally, multivoltine strains revealed shorter larval and total life cycle durations compared to bivoltine strains (Fig 1).

a) Larval duration (in days)



b) Duration of the total life cycle (in days)

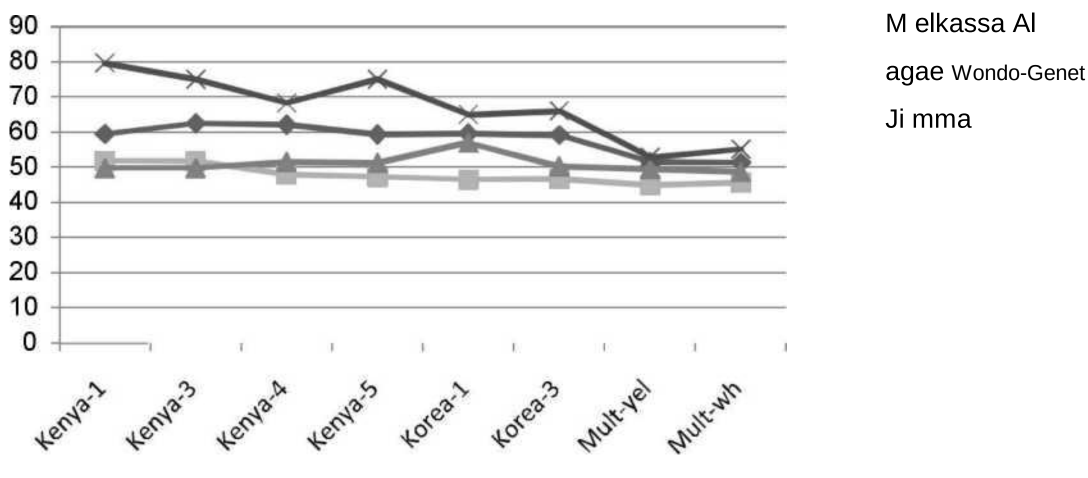


Fig 1. Larval (Fig. a) and total life cycle (Fig. b) (in days) duration of different mulberry silkworm strains

Effective Rate of Rearing

The experiment revealed a range of effective rate of rearing (ERR) was obtained in Alagae from Korea-3 strain (54.17 % and hig hest E R R w as recorded from Mult-yel (92.0 %) an d Mult-wh (92.0 %) (Fig. 2). Multi voltine mulberry silkworm strains (Mult- yel and Mult-wh) have better or at par ERR with other bivoltine strains in Melkassa and Wondo-Genet. The reverse was true in Alagae and Jimma areas. However, strains named Kenya-1 and Kenya-5 were doing well in regard to ERR (≥70%) consistently in all locations compared to other strains.

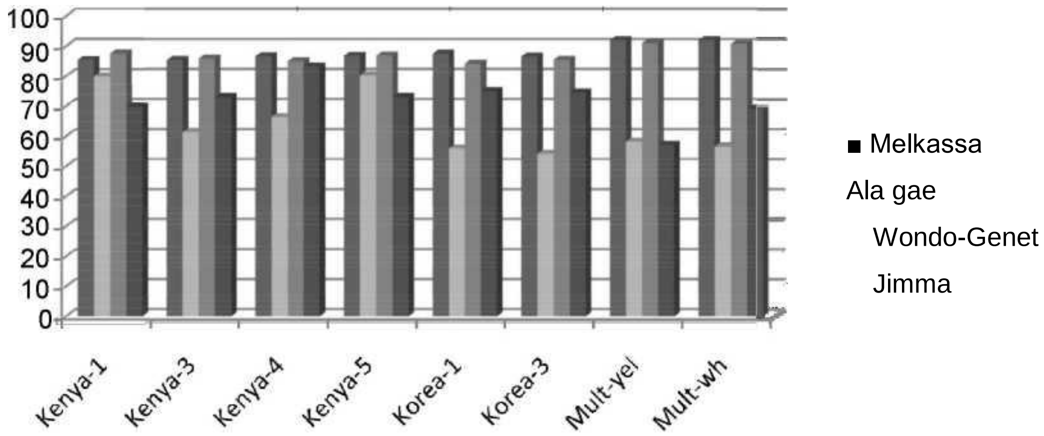


Fig 2. Variability of different mulberry silkworm strains in effective rate of rearing

Matured Larval Weight

Weight of a single matured silkworm larva was significantly different among mulberry silkworm strains in all locations (Table 3).

Table 3. Variations in larval weight (in grams) of different mulberry silkworm strains

Treatments	Melkassa	Alage	Wondo-Genet	Jimma
Kenya-1	3.5333^{a}	2.11667^{a}	2.65667^{a}	3.2300^{a}
Kenya-3	3.100^{bc}	2.10000^{a}	2.03333^{b}	3.4233^{a}
Kenya-4	2.9200^{c}	1.78867^{b}	2.06667^{b}	3.5667^{a}
Kenya-5	3.1633^{bc}	1.63333^{c}	2.64333^{a}	3.5267^{a}
Korea-1	3.1767^{bc}	1.66667^{c}	1.97667^{b}	3.1433^{a}
Korea-3	3.4100^{ba}	1.60000^{c}	2.02222^{b}	3.4233^{a}
Mult-yel	2.2167^{d}	1.43333^{d}	1.37667^{c}	2.0867^{b}
Mult-wh	2.2033^{d}	1.40000^{d}	1.32833^{c}	2.2523^{b}
Pr	<0.0001	<0.0001	<0.0001	<0.0071
CV	6.2904	3.384016	5.813441	10.85261

Means followed by the same letter within a column are not significantly different from each other at 5% level of probability.

In general, larval weight of bivoltine strains was higher than multivoltine strains in all areas. Among the strains, the highest larval weight (3.53 gram) was obtained from Kenya-1 strain in Melkassa area. This strain also exhibited better larval weights in all locations compared to other strains. However, the least larval weight (1.33grams) was recorded from Mult-yel strain in Wondo-Genet area (Table 3).

Cocoon Traits: With respect to cocoon traits (cocoon weight, shell weight and shell or silk ratio), there was statistically significant difference among silkworm strains in all locations (Table 4). In general, better cocoon traits were recorded from bivoltine strains compared to multivoltine ones. In specific terms, highest cocoon weight of 1.60 grams and 1.5747 grams was obtained from Kenya-1strain in Jimma and Melkassa areas, respectively. However, the lowest cocoon weight was obtained from Mult-yel (0.7289 grams) and Mult-wh (0.7256 grams) strains in Wondo-Genet area. Similarly, maximum shell weight was obtained from Kenya-1 strain in Jimma (0.3547 grams) and Melkassa (0.330 grams) areas but the minimum was from Mult-yel (0.1111 grams) and Mult-wh (0.1078 grams) strain in Wondo-Genet area. Moreover, best shell ratio was documented from Kenya-1 strain train in Jimma (22.152 %) and in Wondo-Genet (21.583 %) areas but the least was from Mult-yel and Mult-wh strain in all locations (Table 4).

Table 4. Differences in cocoon traits among mulberry silkworm strains

Treatments	Melkassa			Alage			Wondo-Genet			Jimma		
	Single cocoon weight (gram)	Single shell weight (gram)	Silk ratio (%)	Single cocoon weight (gram)	Single shell weight (gram)	Silk ratio (%)	Single cocoon weight (gram)	Single shell weight (gram)	Silk ratio (%)	Single cocoon weight (gram)	Single shell weight (gram)	Silk ratio (%)
Kenya-1	1.5747[a]	0.3300[a]	20.9939[a]	1.18667[a]	0.27000[a]	22.7579[a]	1.23556[a]	0.26667[a]	21.5829[a]	1.60000[a]	0.35467[a]	22.152[a]
Kenya-3	1.3800[bc]	0.2833[b]	20.5337[a]	1.17667[a]	0.26333[a]	22.3837[ba]	1.21889[a]	0.25000[b]	20.5094[ba]	1.34933[b]	0.27200[bc]	20.099[ba]
Kenya-4	1.3397[c]	0.2730[b]	20.4344[a]	0.90667[b]	0.18667[b]	20.5713[b]	1.21222[a]	0.24333[b]	20.0698[b]	1.47200[ba]	0.26800[c]	18.202[bc]
Kenya-5	1.5467[ba]	0.3267[a]	21.130[a]	0.83110[d]	0.14443[c]	17.3981[c]	1.21556[a]	0.24667[b]	20.2920[ba]	1.53733[a]	0.33467[a]	21.794[a]
Korea-1	1.5500[ba]	0.3200[a]	20.6475[a]	0.87333[c]	0.16000[c]	18.3228[c]	1.23222[a]	0.25000[b]	20.2934[ba]	1.34933[b]	0.27200[bc]	20.099[ba]
Korea-3	1.5300[ba]	0.3267[a]	21.367[a]	0.86667[c]	0.15333[c]	17.6885[c]	1.23556[a]	0.25333[b]	20.5073[ba]	1.50400[a]	0.32667[ba]	21.722[a]
Mult-yel	0.9403[d]	0.1483[c]	15.7737[b]	0.80777[d]	0.12333[d]	15.2640[d]	0.72889[b]	0.11111[c]	15.2654[c]	0.85023[d]	0.12843[d]	15.218[c]
Mult-wh	0.9270[d]	0.1363[c]	14.7054[b]	0.80443[d]	0.12000[d]	14.9023[d]	0.72556[b]	0.10778[c]	14.8741[c]	1.04137[c]	0.16177[d]	15.628[c]
Pr	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	0.0019
CV	7.7314	7.327	3.881	2.038582	6.160211	5.858746	1.615740	3.43454	4.069879	6.365308	11.92655	10.39329

Means followed by the same letter within a column are not significantly different from each other at 5% level of probability

Discussion

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 and humidity plays a major role on growth and productivity of silkworm, as it is a poikilothermic (cold blooded) insect (Benchamin and Jolly, 1986). There is ample literature stating that good quality cocoons are produced within a temperature range of 22 27=C and significant deviations from these levels makes the cocoon quality poorer. However, polyvoltine races reared in tropical countries are known to tolerate slightly higher temperature (Hsieh, 1995), which is also true with crossbreeds, that have been evolved specially for tropical climate.

In the present study, performance of mulberry silkworm strains was tested across different environments which generally depicted a minimum and maximum temperature and humidity ranges of 19-34 °C and 25-83%, respectively. The different silkworm strains showed statistical significant differences among themselves in terms of important silkworm characters in the study locations at $p<0.05$. For example, significant variation in duration to complete different life stages was observed which include larval duration (21.67days to 32.00 days) and total life cycle duration (44.94 days to 79.67 days. Related studies conducted by Shah *et al.* (2007), studied the performance of mulberry silkworms and different seasons and different varieties and confirmed such type of variations. Moreover, significant variability among silkworm strains on egg hatchability (63.67% to 91.00%) and larval weight (1.328 grams to 3.567 grams) and effective rate of rearing (56.22% to 92.0%) were recorded which was also in conformity with Qader *et al* (1992) who observed such differences among silkworm stains through feed nutritive value studies.

In regard to cocoon parameters, this study showed single cocoon weight (0.726 grams to 1.600 grams), single shell weight (0.108 grams to 0.355 grams) and silk ratio (14.71 to 22.76%). This result is in agreement with findings of Nguku *et al* (2009) who studied performnace of different mulberry silkworm strains in neighboring country (Kenya).

In general, all differences could be justified because rearing performance in silkworms is affected by ecological, biochemical, physiological and quantitative characters, which influence growth and development, quantity and quality of silk they produce in different geographical locations (Virk *et al,* 2011; Ramesh *et al,* 2012; Anandakumar and Michael, 2012; and Reddy *et al,* 2012). Among geographically determined factors, Legay (1958) and Scriber and Slansky (1981) stated that temperatures in the range of 21 -27 °C with relative humidity (RH) of 70-85% are required for silkworms effective growth and cocoon productivity. It is known that the productivity of silkworms is affected by temperature and humidity and is verified among the silkworms reared in the all locations. In addition, the nutritive value of leaves especially the moisture content may also vary to contribute to variability in performance of silkworm strains at different agro-ecological zones (Jayaramiah and Sannappa, 1998).

Conclusion

The performance of the mulberry silkworm strains across different locations can be recognized to be variable which will affect mulberry silk production and productivity in general. Therefore, rearing conditions necessitates that the silkworm strain to be reared should be both high yielder and adversity resistant.

In Summary, multivoltine strains showed significantly lower life periods as compared to bivoltine strains in all locations. They also exhibited significantly lower larval, pupal and shell weights. On the other hand, bivoltine strains have performed better than multivoltine strains in regard to commercial traits. However, the bivoltine strains have also revealed significant differences among each other. Among them, a bivoltine mulberry silkworm strain known by Kenya 1 have showed an outstanding performance compared to other strains in all locations. As a result, it is recommended for future research and development efforts on mulberry sericulture in Ethiopia.

However, a cocoon weight up to 1.60 grams that was obtained from this experiment in different locations is lower compared to the findings by Nguku *et al.* (2009) who recorded up to 2.14 grams from improved strains. Therefore, further research and improvement works are required to meet international productivity levels. Important differences in climatic conditions of different silk production areas will also require detailed investigation in future.

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