Analysis of Boil off Loss in the Cocoon Shells of Bivoltine Breeds in Three Seasons

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ABSTRACT
The mulberry silkworm, Bombyx mori L. is one of the productive insect exploited for silk of the commercial importance. The silk is animal based filament made up of two principal proteins viz., fibroin and sericin. The removal of sericin from the silk bave is known as boil-off loss ratio (B.O.R). It is considered as one of the special qualitative trait during the development of silkworm breeds. The current investigation has been undertaken to record cocoon and reeling parameter as well as boil-off ratio in bivoltine breeds viz., CSR2, CSR4, CSR6, CSR16, CSR17, CSR26, CSR27 and S8 in different seasons namely pre-monsoon, monsoon and post-monsoon. The results of the study inferred that, all the breeds performed better in respect of cocoon weight, shell weight, shell percentage, filament length, renditta and denier during monsoon season as compared to pre-monsoon and post-monsoon. Further, lower significant of boil-off loss ratio was registered by all the breeds during post-monsoon season. However, CSR2 recorded lowest value for this trait among breeds.

Keywords: Bombyx mori L., Bivoltine hybrids, Boil-off loss ratio, Seasons, Commercial traits

INTRODUCTION
The sericulture is closely related with rich heritage of Indian civilization. India trails behind China for production of raw silk and occupies second position in the world (Vijayaprákash & Dandin, 2005). The main intension of silkworm breeding program is to develop high cocoon yielding silk breeds without compromising its quality. At the end of larval period, the silkworm spins a cocoon shell by extruding silk bave to protect pupa from adverse environment condition. The silk bave is composed of central core fibroin protein, which is surrounded by sericin protein. It also contains a meager amount of fat, wax, colouring and mineral matter not exceeding 2-3 % (Carboni, 1952). The main silk substance fibroin is insoluble in alkaline hot water, where as gummy sericin protein is readily soluble in boiling alkaline soap solution (Sadov et al., 1978). Hence, sericin is protein removed in the process of degumming. The cocoon shell has more boil-off percentage when compared to the raw silk.

The percentage of boil-off loss has got paramount importance in reeling and weaving activities (Kannan, 1986). Raw silk neither possesses luster nor softness due to presence of sericin (Roopesh Kumar et al., 2003). In sericulturally advanced countries like China and Japan, silkworm breeders have successfully bred productive hybrids. During the course of breeding, boil-off loss is considered as one of the important quantitative trait and for this trait lower value is preferred. The boil-off ratio for bivoltine is found to be 24% and it is optimum. It is genetically differing among the silkworm strains (Sinha et al., 1992). The degumming loss percentage is higher in multivoltines than bivoltines due to genetic constitution (Sidhu & Sonwalker, 1969). The boil-off loss ratio varies according to the season, which is influenced by the environment (Sonwalker, 1969). The contribution of sericin is highest for mulberry silk (23 to 30%) when compared to non-mulberry silk (Venugopal, 1991). Low boil-off ratio content improved cocoon reeling qualities and is manifested by dominant genes, while recessive genes act towards the opposite direction (Gamo & Hirabayashi, 1984). Several reports are available on boil-off loss ratio in silkworm breeds and hybrids. The boil-off loss ratio is intermediate in bivoltine hybrids and multivoltine x bivoltine hybrids (Basavaraja et al., 2000; Veeranna Gowda et al., 2013; Seetharamulu et al., 2013 and Anil Kumar, 2018). The silk degumming is necessary for process which make enable the penetration of chemical and dye stuff substances easily. After the removal of sericin the silk used in weaving process. The tropical climate like India is characterized by variations in environmental factors and successes of cocoon crop depends on manipulation of temperature and humidity in different seasons. The seasonal variations in the environment has notable impact on phenotypic expression and heritable commercial traits such as cocoon weight, shell weight and shell ratio (Nacheva & Junka, 1989). Realizing the importance of tropical environment for the production of quality silk, several studies were under taken to know the impact of different seasons on mulberry crop and cocoon production (Narayana et al., 1964 and Kasiviswanathan et al., 1970). Further, estimation of boil-off loss ratio (B.O.R) is pertinent during the process of breeding and also one of the important parameter for evaluation of hybrids during race authorization. However, seasonal influence on boil-off loss ratio in silkworm breeds is meager. Hence, the present study is necessitated.

MATERIALS AND METHODS
The disease free layings of eight silkworm bivoltine breeds viz., CSR2, CSR4, CSR6, CSR16, CSR17, CSR26, CSR27 and S8 obtained from NSSO grainage, Mysuru. After brushing the larvae were reared by employing standard rearing techniques (Dandin & Giridhar, 2010) in three different seasons viz., pre-monsoon (March to June), monsoon (July to October) and post monsoon (November to February). After the cocoons harvest, a minimum of ten cocoons (three replication each) were selected from each breed and season wise to record cocoon and reeling parameters namely cocoon weight, shell weight, shell percentage, filament length, denier and renditta. In addition to this, degumming was carried out by boiling the cocoon shells in soap solution by following procedure suggested by Basavaraja et al. (2000).

The parameters namely shell percentage, filament length, denier and renditta were calculated by using following formulae.

Shell ratio = \( \frac{\text{Shell weight (g)}}{\text{Cocoon weight (g)}} \times 100 \)

Filament Length (m) = \( R \times 1.125 \)
Where, \( R \) = Number of revolutions recorded by an epprouvette.

1.125 = Circumference of epprouvette in meter.

Denier = \( \frac{\text{Weight of the filament}}{\text{Length of the filament}} \times 9000 \)

It denotes thickness of the silk filament.
Unit quantity of cocoons required to produce one unit of raw silk.

Degumming

The initial weight of ten cocoon shells of bivoltine breeds (seasons wise) were recorded before degumming process. Selected cocoon shells were subjected to liquor bath consists of mixture of neutral soap and sodium carbonate. The amount of soap and sodium carbonate required for degumming for one gram of cocoon shell are follows:

- 40 ml of liquor solution (Water)
- Soap 7 gram per liter
- Soda ash 1 gram per liter

The cocoon shells of respective breeds were immersed in the beaker containing liquor bath where the temperature was maintained at 90 to 95°C for about one hour. The material was turned up and down to achieve uniform and effective degumming. Afterwards it was boiled in distilled water for about half an hour. Finally, the material was washed in tap water and then dried. The final weight of the material was recorded.

The boil-off loss percentage was calculated by using the formula:

$$\text{B.O.R.} = \frac{\text{Initial dry weight} - \text{final weight after degumming}}{\text{Initial dry weight}} \times 100$$

The data obtained in the investigation was statistically analyzed by employing one-way factorial completely randomized design at 5% level of significance OPSTAT online statistical package developed by O.P.Sheoran, Programmer, Computer section Chaudhury Charan Singh Hisar Agriculture University, Hisar, Haryana State, India.

RESULTS AND DISCUSSION

Cocoon weight

Cocoon weight is an important attribute for the yield as this character mainly depends on the race/breed and its ability to convert mulberry leaf into cocoon. The bivoltine breeds differ one another for cocoon weight and even season wise. The highest cocoon weight of 1.831 was recorded in S8 during monsoon season and it was lowest in CSR16 (1.520g) during pre-monsoon season (Table.1). In plants and animals, the degree of variations were the phenotypic expression is dependent on the responsiveness of the different genotypes to diverse environment conditions (Griffing & Zsiros, 1971; Orozco, 1976 and Strickberger, 2002). The variations in the cocoon weight among the bivoltine silkworm breeds differ due to their genetic constitution and interaction with dynamic environment. Better expression for this trait in the selected breeds in monsoon season is due to congenial environment. It is also clear by the earlier studies that many of the quantitative tarits in silkworm respond differently to inherent polygenic system. Hence, differential response of the genotype of silkworm (*Bombyx mori* L.) was noticed in response of changing environmental factor in different seasons. The present finding are in conformity with the observations of Anatha and Subramanya (2010) who have reported that the higher value of cocoon weight was recorded in multivoltine race Daizo (1.19g) and bivoltine breed CSR2 (1.92g) in monsoon season and it was lowest in Pre (0.72g) and C108 (1.54g) in pre monsoon season. Similar results were also observed by Nagalakshamma and Nagajyoothi (2010) in some new evolved multivoltine breed MU303 and Pure Mysore race in monsoon and pre- monsoon seasons, respectively. For this traits lower value was recorded in pre-monsoon is due to unfavorable environmental conditions prevailing during the different seasons are in conformity with earlier findings of Subramanya (1985), Rajanna (1990) and Mariba shetty (1991).

Shell weight

It is an important economic character which indicates the silk content. The expression of the trait differ among silkworm breeds and
seasons. The maximum shell weight of 0.426 g registered by S8 in monsoon season. In contrast, it was minimum in CSR16 (0.317 g) during pre-monsoon season. All breeds performed better for this trait during monsoon season (Table 1). Zhoo et al. (2005) observed that metamorphosis of larvae into pupae and adult depends on shell weight which is influenced by maintenance of environment conditions during different larval stages. It is clear from the investigation that all the silkworm breeds expressed better for this traits during monsoon season is due to favorable environmental conditions which is characterized by low temperature and high humidity. These results are on par with the findings of Nagalakshamma and Nagajyothi (2010) who have opined that higher shell weight was recorded in MU303 (0.20 g) in monsoon season and it was lowest in Pure Mysore (0.14 g) during pre-monsoon season. This type of trend was observed for the said trait by Antha and Subramanya (2010) in multivoltine and bivoltine breeds during monsoon and pre-monsoon seasons, respectively. Likewise, expression for the said trait differ in silkworm breeds in relation to different seasons were also noticed by Kalpana (1992), Nanjundaswamy (1997) and Veeraiah (1999).

Shell percentage
It is one of the important economic parameter contributing for silk productivity which indicates the silk content. This character varies in different breeds over different seasons. The highest shell percentage of 23.78% exhibited by S8 in monsoon season. In contrast it was lowest in CSR16 (19.59%) during pre-monsoon season (Table 1). All the breeds scored better for this trait during monsoon season because of congenial environment conditions which reflects on growth and development of silkworm resulting in enhanced shell percentage. These results are on the line with the earlier observations of Anantha and Subramanya (2010) who have reported that the higher value of shell percentage was registered in multivoltine race Diazo (16.65%) and in bivoltine breed CSR2 (23.12%) in monsoon season. On the other hand, pre and C108 recorded lowest value of 11.86% and 17.86% for the said trait, respectively in pre-monsoon season. Similar observations were noticed in multivoltine and bivoltine breeds during monsoon and pre-monsoon seasons by Nagalakshamma and Nagajyothi (2010) and Anil Kumar (2009).

Filament length
Filament length is one of the major contributory for quantitative trait in silkworm (Miyahara 1978 and Yokoyama, 1979). This character differ among silkworm breeds and even seasons wise. The maximum filament length of 1090 m expressed in S8 during monsoon season. In contrast, it was minimum in CSR16 (786 m) during pre-monsoon seasons. All the breeds exhibit better for the said trait during monsoon season followed by post-monsoon and pre-monsoon season (Table 1). It is evident from the result that favorable environment conditions of monsoon and post-monsoon seasons accelerated the silk protein synthesis which might have resulted in longer filament length. These results are in agreement with the findings of Masrat Bashir et al. (2014) who have reported that longer filament length of 1252 m registered in silkworm breed SK-1 (Marked) during monsoon season and it was shortest in CSR2 (710 m) in pre-monsoon season. Shorter filament length during pre-monsoon season might be due to high temperature and low humidity causes dehydration of silk protein resulting in more extrusion of liquid silk leads to wastage. Similar observation were made in multivoltine breeds MU303 (638 m) and Pure Mysore (394 m) during monsoon and pre-monsoon seasons, respectively by Nagalakshamma and Nagajyothi (2010).

Renditta
This trait mainly depends on cocoon weight and shell weight content. The silkworm breeds exhibit variations for this trait in different seasons. The lowest renditta was recorded in S8 (6.09) during monsoon season. As against to this, it was highest in CSR16 (7.89) during
post-monsoon season (Table.1). Better expression for this trait were recorded in all the breeds during monsoon season when compared to remaining two seasons. It clearly depicts favorable environment conditions during monsoon enhance renditta. Perhaps highest value for this trait was observed among selected silkworm breeds might be due to prolonged larval duration and lower metabolism during pot-monsoon season. These results corroborate in earlier findings of Anantha and Subramanya (2010) who have opined that lowest renditta was recorded in multivoltine breed Diaz (10.81) in monsoon season and it was highest in Pure Mysore (17.16) in pre-monsoon season. Similar trend was observed for this trait in multivoltine and bivoltine breeds during monsoon and pre-monsoon seasons, respectively (Anil Kumar, 2009). Likewise, expression for the said trait differ in silkworm breeds over different seasons were also noticed by Kalpana (1992), Nanjundaswamy (1997) and Veeraiah (1999).

**Denier**

It denotes the thickness / size of the silk filament. This character differ among bivoltine breeds in different seasons. Generally, multivoltine race gifted with low denier whereas bivoltine with higher denier (Anil Kumar, 2009). The lowest denier of 2.81 expressed in S8 during pre-monsoon season. On the other hand it was highest in CSR6 (3.26) during post-monsoon season (Table.1). The better expression for this trait in all the breeds during pre-monsoon followed by monsoon and post-monsoon seasons. The results of the study revealed that expression for this trait was found to be better in pre-monsoon season due to high temperature and low humidity. The present findings are on par with the earlier observations of Anantha and Subramanya (2010) who have opined that lowest denier was registered in multivoltine breed Pre (1.61) and bivoltine breed C108 (2.55) during pre-monsoon season. While it was highest in multivoltine breed Diaz (1.89) during monsoon season. The expression differs in the silkworm breeds for this trait over different seasons were also noticed by Kalpana (1992), Nanjundaswamy (1997) and Veeraiah (1999).

**Boil-off loss ratio**

The boil-off loss ratio is one of the special character and silkworm breed having lower for this trait is consider during breeding programme. The data pertaining to this trait revealed significant difference between breeds and even seasons. All the breeds scored lower boil off ratio in post-monsoon followed by pre-monsoon and monsoon seasons. Among the breeds, lowest value for the said trait recorded by CSR6 (21.22 \%) during post-monsoon season. Whereas, it was highest in the breed CSR2 (24.90 \%) during pre-monsoon season (Table.2). The lowest value for this trait were observed in all the silkworm breeds during post-monsoon is due to interaction of genetic system with that of dynamic environment. Moreover, post-monsoon is characterized by low temperature and low humidity which alters rate of metabolism with prolonged larval duration. This might have manifest silk protein synthesis in the silkworm larvae and as a consequence lesser quantity of sericin is synthesized as compared to fibroin protein in the silk filament which leads to lower boil-off loss ratio in this season. In contrast, higher value were scored by the silkworm breeds for this traits during pre-monsoon season where larvae exhibit shorter larval duration resulting in more synthesis of sericin than the fibroin. The higher sericin content in the silk filament naturally enhance boil-off loss ratio in this season. Further, variations were also noticed for boil-off loss ratio within the silkworm breeds and it might be due to diverse nature of genetic constitution. These results corroborate in earlier observations of Anil Kumar et al. (2018) who have reported that the boil-off loss percentage is relatively higher in multivoltine breeds than the bivoltine breeds. Similar trend were also noticed in bivoltine silkworm breeds by Veeranna Gowda et al. (2013).
The results of the present investigation inferred that all the silkworm breeds expressed lower boil-off loss ratio in post-monsoon season when compared to pre-monsoon and monsoon seasons. The present finding helps to develop seasonal breeds with lower boil-off loss ratio.

### Table 1: Cocoon and post cocoon parameters of selected breeds in different seasons

<table>
<thead>
<tr>
<th>Breed</th>
<th>Pre- Monsoon</th>
<th>Monsoon</th>
<th>Post-monsoon</th>
</tr>
</thead>
<tbody>
<tr>
<td>CSR 26</td>
<td>22.16 ± 0.3</td>
<td>21.7 ± 0.2</td>
<td>21.1 ± 0.3</td>
</tr>
<tr>
<td>CSR 17</td>
<td>22.27 ± 0.3</td>
<td>21.9 ± 0.2</td>
<td>21.5 ± 0.3</td>
</tr>
<tr>
<td>CSR 16</td>
<td>22.35 ± 0.3</td>
<td>21.9 ± 0.2</td>
<td>21.5 ± 0.3</td>
</tr>
<tr>
<td>S8</td>
<td>22.39 ± 0.3</td>
<td>21.9 ± 0.2</td>
<td>21.5 ± 0.3</td>
</tr>
</tbody>
</table>

### Table 2: Boil-off loss ratio of bivoltine breeds in different seasons.

<table>
<thead>
<tr>
<th>Breed</th>
<th>Pre-monsoon</th>
<th>Monsoon</th>
<th>Post-monsoon</th>
</tr>
</thead>
<tbody>
<tr>
<td>CSR 2</td>
<td>22.69 ± 0.1</td>
<td>24.90 ± 0.1</td>
<td>21.81 ± 0.2</td>
</tr>
<tr>
<td>CSR 4</td>
<td>22.3 ± 0.1</td>
<td>24.78 ± 0.1</td>
<td>21.65 ± 0.1</td>
</tr>
<tr>
<td>CSR 6</td>
<td>22.60 ± 0.1</td>
<td>24.65 ± 0.3</td>
<td>21.22 ± 0.1</td>
</tr>
<tr>
<td>CSR 16</td>
<td>22.55 ± 0.1</td>
<td>23.91 ± 0.1</td>
<td>21.23 ± 0.4</td>
</tr>
<tr>
<td>CSR 17</td>
<td>21.58 ± 0.2</td>
<td>23.72 ± 0.4</td>
<td>21.11 ± 0.3</td>
</tr>
<tr>
<td>CSR 26</td>
<td>23.21 ± 0.2</td>
<td>24.06 ± 0.1</td>
<td>22.11 ± 0.1</td>
</tr>
<tr>
<td>CSR 27</td>
<td>23.27 ± 0.4</td>
<td>24.89 ± 0.2</td>
<td>21.51 ± 0.2</td>
</tr>
<tr>
<td>S8</td>
<td>22.63 ± 0.4</td>
<td>23.97 ± 0.1</td>
<td>21.52 ± 0.6</td>
</tr>
</tbody>
</table>

* ( ) : Angular transformed values, * : Significant at 5% level ± : Standard error values.
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