

# 1. INTRODUCTION

## 1.1 Background of the study

Nepal being the agricultural country, at present agriculture is the main source of livelihood of majority of people. About 81% of economically active population is entirely dependent upon agriculture contributing 40% of total domestic product (CBS 2001). They usually live in village areas and rely on agriculture for their income and employment (Rijal 2003). Majority of people who are poor and illiterate are involved in this field so it occupies a strategic position in the Nepalese economy (MoAC 1999). Beside this sericulture as an industry could be quite advantageous as this profession could be taken in relation with the agriculture. Apart from this the geographical structure and climatic condition is also very favorable for sericulture.

Sericulture was first discovered in China (Encarta 1998) and later it was introduced to other many countries and began to flourish as an industry. Nowadays many countries like Japan, China, South Korea, Soviet Union, India and Italy are the major sericultural countries. Singh (2004) stated in the survey of the International Silk Association and the Japan Raw Silk cocoon that silk harvesting is low in China and India than in Japan and South Korea.

Sericulture as being a village- based industry providing employment opportunity to various person, it is a quite good profession for developing and under developing country like Nepal. In Japan, this industry made a remarkable coverage for the loss in Second World War in various fields toward the national economy (Ganga and Chetty 1991). According to the study made by Food and Agriculture Organization (1973), the production of raw silk is expected to decline in the developed countries and to increase in the developing countries. In Nepal too for achieving higher economic growth, agriculture and sericulture should be taken into parallel with each other (APROSC/JMA 1996).

Sericulture is derived from two words “sericos” meaning silk and “culture” meaning rearing. So it refers to mass scale rearing of silk producing organisms in order to obtain silk from them (Fenemore 1992). Sericulture is a labour intensive activity including both entomological aspect (rearing of silkworm) and agricultural aspect (cultivation of mulberry leaves). Hence in this technology, caterpillars that produce silk are fed on the leaves of mulberry to produce cocoons and it supply valuable natural silk. For this

silkworm rearing technology varies to suit the requirements under different ecological conditions making it cost effective and more productive (Shekharappa et al. 1994). Nepal lies well within the sericultural belt (20° to 40° N), so the climatic conditions for silkworms and mulberry plants are favourable and the manpower required for this industry is also easily available.

The ultimate motivation for farmers to take up any cash crop including sericulture is high profit. High yielding silkworm races feed on nutrient rich mulberry varieties yield desirable gains (Arunachalam 1994). The main foundation for rearing silkworm is a good mulberry garden (Benjamin and Nagaraj 1987). Disease resistant races of silkworm lead to higher percentage of hatchability and hence cocoon production. It is well known that quality and quantity of silk are the results of quality and quantity of mulberry leaves given to the worms. Cocoons with no wrinkles/grains and highly weighted cocoons give long and smooth silk. The proper care and attention taken during the rearing of worms and mounting phase results in higher percentage of cocoon weight. Hatching percentage also depend upon the vitellogen-deficient eggs developed in male hosts of silkworm (Yamashita et al. 1980) specially. Hatchability percentage and disease resistant race is also related to the amount of silk produced by the worms.

## **1.2 Silk**

Silk is a fine, soft and lustrous fiber produced by the larvae of silkworm during the eve of its pupation so called a fibrous protein of animal origin. Silk is produced within the worm through a special gland called silk gland. The shimmering appearance of silk is due to the triangular prism-like structure. Silk's good absorbency makes it comfortable to wear in warm weather while its thermal regulation makes our body warm in cold days. Hence is best for human to wear in any season (Fenemore 1992). Apart from this, silk is produced by other insects too like spider but the silk produced by spider is adhesive and fragile. Hence it should not be used in textile industry and is not considered as of good quality. *Bombyx mori* Linnaeus is the most widely reared silk spinning insect in the context of Nepal. There are many varieties of silkworm but the silk produced by *B. mori* is of highly priced, strong and lustrous. Sericulture is not only an industry of silk but it also supplies many other byproducts like bad cocoons, faeces of worm, damaged silk thread, mulberry plants dead worms etc.

### **1.3 Varieties of silkworm**

There are two types of silkworm: mulberry silkworm and non-mulberry silkworm. Mulberry silkworm is a monophagous plant depending entirely upon mulberry plant (*Morus alba*), while non-mulberry silkworm feeds on other plants too such as Sal (*Shorea robusta*), Bar (*Zijuphus jujuba*), Arjun (*Terminalia arjuna*) etc. Mulberry feeding silkworm belongs to family Bombycidae and non-mulberry feeding silkworm belongs to family Saturnidae. Silk of mulberry silkworm are best for the textile industry as they produce fine filaments of silk inside the cocoon. *Bombyx mori* is the only mulberry silkworm and non-mulberry silkworms are tasar, eri and muga and they produce tasar silk, eri silk and muga silk respectively. Other than these nowadays following pure lines are being used for the commercial purposes in Sericulture Development Division, Khopasi from which higher percentage of best quality cocoons could be harvested like Nepal-107, Nepal-108, Nepal-109, Khopasi-101, Khopasi-102, Khopasi-103, J-1, J-2, C-1, C-2 etc.

### **1.4 Biology of mulberry silkworm**

Depending upon the climatic conditions, some population of silkworm are univoltine, bivoltine and multivoltine. Univoltine race can be reared only once in a year, bivoltine two times in a year and multivoltine more than two times in a year. Silkworm exhibits complete metamorphosis with the following four life stages:

#### **1.4.1 Egg**

The number of eggs laid is highly influenced by the quality of nutrition/quantity of leaves of mulberry plant obtained by the caterpillars during feeding. Normally each female lay about 300-400 small, smooth, sub spherical yellowish eggs. These are seed like in groups usually covered by gelatinous secretion of female. They are of either yellowish or brownish in color. After the incubation, egg hatched into larvae.

#### **1.4.2 Larvae**

Under the influence of suitable temperature yellowish eggs changes into brownish and hatching takes place in 8-12 days. Then the larvae matured feeding entirely upon the mulberry leaves. Larvae are elongate, cylindrical with a small anal horn. The head bears a pair of short antenna, 2 eye spots, mouthparts and a spinneret. The newly hatched larvae

are 0.5 cm long while the fully grown caterpillar is 7.5cm long. During fifth instar the caterpillar eats voraciously and becomes transparent and waxy in appearance. The worms moult for four times in its life time and in this phase it should not be disturbed otherwise the quality of silk produced by them would decline. Between the moults they eat a lot. Their one and only job is to eat and grow.

### **1.4.3 Pupae**

After the fifth instar the larvae stops feeding and starts spinning cocoon in the mounting frame around its body. The mature larvae transform into somewhat rounded pupa after 3-4 days of spinning of cocoon. The cocoon is made up of 3 distinct layers: the outer floss layer, middle compact shell and the inner pelode layer. The pupa is non-feeding and non-motile stage. The pupae of silkworm are of obtect type as the appendages are firmly pressed against the body. It is usually white in colour.

### **1.4.4 Adult**

Pupa takes 10-12 days to transform into the adult. The moths secrete an alkaline saliva from its mouth with which it moisten one end of the cocoon and comes out. The moths neither feed nor fly. It only flutters. Body is differentiated into head, thorax and abdomen. It has a pair of bipectinate antenna and two pair of wings. Sexual dimorphism is appeared in their abdomen as the abdomen of female is slightly larger than that of male.

The taxonomic position of silkworm is:

Phylum: Arthropoda

Class: Insecta

Subclass: Pterygote

Division: Endopterygote

Order: Lepidoptera

Suborder: Ditrysia

Superfamily: Bombycoidea

Family: Bombycidae

Genus: *Bombyx*

Species: *mori*

While the systematic position of mulberry plant is:

Division: Phanerogams

Subdivision: Angiosperm

Class: Dicotyledon

Subclass: Apetalae

Family: Moraceae

Genus: *Morus*

Species: *alba*

The various sizes of leaves that were feed at different instar larvae are given below:

### **1.5 Sizes of leaf needed to feed different instar of silkworm**

<u>Age/instars</u>	<u>Size of leaves</u>
1 <sup>st</sup> instar	0.5-1 cm
2 <sup>nd</sup> instar	2 cm
3 <sup>rd</sup> instar	4-5 cm
4 <sup>th</sup> instar	whole leaf
5 <sup>th</sup> instar	whole branch

While rearing the silkworm proper sanitation and maintainance of suitable temperature and relative humidity also must be appropriate. Upto 3<sup>rd</sup> instar the silkworms should be feed thrice a day while for 4<sup>th</sup> and 5<sup>th</sup> instar larvae they were fed five times a day and the number of worms also should be decreased in the rearing tray as they grow faster and there should be enough spacing between them .

## 1.6 Required temperature and RH for different instar silkworm

<u>Age/instar</u>	<u>Temperature</u>	<u>RH</u>
1 <sup>st</sup> instar	27-28°C	85-90 %
2 <sup>nd</sup> instar	26-27 °C	80-85 %
3 <sup>rd</sup> instar	25-26 °C	75-80 %
4 <sup>th</sup> instar	24-25 °C	70-75 %
5 <sup>th</sup> instar	23-24 °C	65-70 %

Delitage operation or sanitation should be maintained seriously in the early instar periods than the late stage as they are more sensitive in early phages and has maximum probability to get infected by various bacterial, viral or other diseases. The larval duration lasts for some 25 days in between which it molts four times. The larvae are voracious eaters but it stop feeding in the middle of molting stage like in between 1<sup>st</sup> and 2<sup>nd</sup> instar, 2<sup>nd</sup> and 3<sup>rd</sup> instar, 3<sup>rd</sup> and 4<sup>th</sup> instar and at last at 4<sup>th</sup> and 5<sup>th</sup> instar. During molting it should not be disturbed or delitage operation should not be conducted. Spacing between the larvae also should be appropriate as they gradually grow in size. After the last molt the larvae find a proper place and anchor itself to the mounting frame to spin cocoon. Then after 7-8 days of mounting the cocoons are ready to harvest. Now these cocoons are called ripe cocoon. In the span of rearing larvae the diseased larvae are separated from the mass to get further contaminated and the cause of the disease were identified as far as possible.

## 1.7 Diseases

Various types of diseases were found during the study period. Generally larvae get diseased due to unsuitable temperature, low food, genetic causes, distractions during moulting etc. Major diseases of silkworm are described below.

### 1.7.1 Cytoplasmic Polyhydrosis Virus (CPV)

This is a viral disease of silkworm that is usually recognized by presence of milky white secretion. It results due to feeding low quality of mulberry leaves. This disease is also known as white flacherie because the faecal matters seem to be white in colour. The

infection becomes severe during 5<sup>th</sup> instar. By maintaining proper sanitation, relative humidity and suitable temperature will protect the larvae from this disease.

### **1.7.2 Nuclear Polyhydrosis Virus (NPV)**

This disease is also called grasserie and infects almost every stage of the larvae. If the leaves get contaminated by the chemical fertilizers or worms are feed with dry leaves of mulberry then NPV is caused.

### **1.7.3 Muscardine**

This disease is characterized by presence of brown strips on the dorsal as well as ventral surface of the worms. These strips get darker at the edges while it becomes lighter at the centre of the body. Muscardine is a type of fungal disease that later gradually makes the larvae dries off. Muscardine is also of various types like yellow, red, black, brown etc

Beside these diseases various other vertebrates like lizards, rats, cats, rodents etc are also the destructors of silkworms during rearing period.

## **1.8 Objectives of the study**

### **General objective**

- ❖ To recommend best quality of cocoon among five selected silkworm races.

### **Specific objectives**

- ❖ To know the maximum hatchability among five races.
- ❖ To test the disease resistant race among them.
- ❖ To determine the quality of produced cocoons.

## **1.9 Rational of the study**

The general purpose of writing the thesis report entitled “Hatchability, disease resistance and quality of produced cocoons among five selected silkworm races in sericulture Development Division, Khopasi” is to recommend the most feasible silkworm race. This will help uplift the economic condition of poor people by increasing the yield of cocoon production locally as silkworm rearing is the job that can be conducted with less money and time. This will give idea to know and identify the hatching ability, disease resistance and hence determine quality of the produced cocoons among five silkworm races in Khopasi. Since they are the hybrid races which are distributed directly to the farmers for commercial silkworm rearing, they should bear the best qualities in them so that maximum cocoon production is achieved. Beside agriculture, cocoon production stands as a best part time job among the under privileged people of Nepal as it does not need much time and money. This job can also be done by every aged people such as from child to old aged ones. This study will also add a step forward to get rid of problems related to diseases of silkworm that will benefit the silkworm rearing procedure.

## **1.10 Limitations of the study**

- ❖ Due to lack of sufficient time study of further generations could not be accomplished.
- ❖ Budget constrain so difficult to carry more research on this topic.
- ❖ Due to regular work carried out within the Sericulture Division, lack of lab facilities was faced.



## 2. LITERATURE REVIEW

### 2.1 In the context of Nepal

From the literatures on silkworm, it has been known that Kafle (1970) studied the feasibilities and probabilities of sericulture in Nepal. In the same year he also studied the occurrence behavior and seasonal cycle of local wild silkworm *Bombyx mori*. Singh (1981) in Nepal studied the effect of food on the yield of raw silk in mulberry silkworm (*Bombyx mori* Linnaeus). After that Devkota (1983) identified qualitative and quantitative variations in the production of cocoon and raw silk by mulberry silkworm, *Bombyx mori* Linnaeus due to different frequencies. After that a qualitative study on the food utilization by the silkworms accounting the N<sub>2</sub> flow from mulberry to silkworms in terms of ecosystem was summarized and food utilized was determined by the gravimetric method (Horie, Yasuhiro et al. 1976). Jolly (1978), a U.N. sericulture consultant under Narcotic Division studied and recommended for the development of non-mulberry silk in Nepal. Later it was told that larval growth of mulberry silkworm depend upon the food utilization (Prasai 2005). The effect of the refrigeration of cocoon on the fecundity of moth and hatchability of eggs of multivoltine mulberry silkworm (*Bombyx mori* L.) vary (Upadhaya 2006).

### 2.2 In the context of world

Silk had been reared for over 5000 years in China as per Nagaraju and Goldsmith (2002). Watanabe (1966) found that cytoplasmic polyhedrosis virus seem to be more infectious toward silkworm than nuclear polyhedrosis virus. Nakada et al (1994) determined the cocoon measurement and its statistical analysis in the silkworm, *Bombyx mori*, Linnaeus. Verma, Narasimhana et al. (1972) studied the rearing period of silkworm. Sugai et al. (1972) sterilized the male silkworm, *Bombyx mori* L. by the high temperature environment. The leaf quality of 3 improved strains was evaluated by the feeding experiment on the different races of silkworms. This was done on the same year by Das and Sikder. In 1973, the factors responsible for mortality of silkworm were suggested by Nigam et al. (1973) studied on some strange hatching behavior in silkworm *Bombyx mori* L. Hassanein et al. (1974) reported the degree of food assimilation and quantities of

secreted silk from one race to another in *Bombyx mori* worm. The effect of different types of mulberry leaves i.e. trees, lowcut, bush leaves on the weight of silkworm larvae were investigated (Khan et al. 1976). Fertility and hatching was enhanced with reduction of fecundity when pupae of different periods were exposed to high temperature (Singh 1980). Usually some races show higher hatchability when they are treated integrated factor combinations according to Yu, 1982. Later Watanabe (1986) found CPV resistant genes in the silkworms. On a comparative study of the effect of feeding on fresh mulberry leaves and recommended artificial diet on the biology of silkworm *Bombyx mori*. Also in the same year Cornaby et al (1986) studied the feeding behavior of larval silkworms on different shapes of mulberry leaves. In the same year Fonesca et al. studied the influence of the age of mulberry leaf on silkworm growth and development. Hato et al. (1989) studied on resistance of the silkworm, *Bombyx mori* L. for high temperature in proceedings of the 6<sup>th</sup> International Congress of SABRAO (II). Chartterjee et al. (1990) extrapolated that 21 characters do influence the silk yield qualitatively and quantitatively. Thiagarajan et al. (1993) studied on the performance of 26 strains of silkworm *Bombyx mori* (Bombycidae) in different seasons. Singh et al. (1998) studied on cocoon shapes in different crosses of the mulberry silkworm, *Bombyx mori* L. Rahmathulla (1999) found management procedures of climatic factors during silkworm rearing. In the same year Sen et al. told that an extensive study is needed to develop new strains that are disease resistance. Nataraj et al. (2001) accomplished the influence of temperature and relative humidity on the rearing performance and disease incidence in CSR hybrid silkworms, *Bombyx mori* L. Reddy et al. (2002) found implications of temperature and humidity on the adult eclosion patterns in silkworm *Bombyx mori* L. Rhmathulla (2004) found influence of various environmental and nutritional factors during 5<sup>th</sup> instar silkworm rearing on silk fibre characters. Manjula et al. (2005) determined effect of preservation of seed cocoon/pupae at different environmental condition on the reproductive efficiency of new bivoltine silkworms, *Bombyx mori* L. in proceedings of the advances in Tropical Sericulture. Manisankar et al. (2008) studied effect of environmental factors (temperature and humidity) on spinning behavior of silkworm (*Bombyx mori* L.). In the same year Ganesh et al. studied microbial factors in the digestive juice of silkworms. Hussain et al. (2011) studied on effect of relative humidity on factors of seed cocoon production in some inbred silkworm (*Bombyx mori*) lines. Recently Jiang, et al. (2012) explored that the transgenic technology is a powerful tool for improving disease resistant species.

These all literatures will definitely help to draw some of the conclusions about the above mentioned topic. The temperature and relative humidity directly or indirectly affect the hatching capacity of silkworm eggs/seeds. They will give idea about the correct rearing techniques during the span of the field. Determination of cocoon measurement and its statistical analysis also became very convenient due to the older Nepali and world literatures.

### 3. MATERIALS AND METHODS

The present rearing work had been undertaken in Sericulture Development Division, Khopasi, Kavre district of Nepal. It is about 35 km south-east from the capital city Kathmandu. The species of silkworm is *Bombyx mori* L. while the variety of mulberry plant fed to the silkworm is *Morus alba*. Box rearing for large larvae and paraffin paper rearing for small ant worms were done during the study period. The cocoons were mounted in the mounting frame at the late stage of their development. This observation data is based on the work done during spring season when the maximum temperature is 29°C and the relative humidity is 91. The study was carried out with five selected silkworm races namely K<sub>1</sub>, K<sub>2</sub>, K<sub>3</sub>, STD<sub>1</sub> and STD<sub>2</sub> respectively for which following materials had been required.

#### 3.1 Materials required

- ❖ Rearing house, rearing tray and rearing cage
- ❖ Knife
- ❖ Mounting frame
- ❖ Chopping board
- ❖ Bamboo basket
- ❖ Thermometer
- ❖ Hygrometer
- ❖ Balance
- ❖ Feather
- ❖ Heating apparatus
- ❖ Net
- ❖ Long foam rubber strips
- ❖ Paraffin paper

#### 3.2 Chemicals required

- ❖ Ethyl alcohol (70%)
- ❖ Formalin
- ❖ Lime solution
- ❖ Hydrochloric acid
- ❖ Salt

### **3.3 Methods**

#### **3.3.1 Hatchability**

Hatchability will be determined by counting the successfully hatched ant worms. For this, 200 eggs each of five selected silkworm races will be observed in two treatments in the spring season in Sericulture Development Division, Khopasi. Hatching procedures will be overcome only after proper disinfection. For which the eggs should be washed with HCl solution at first and then by cold water, after that only they will be kept for incubation. For uniform hatching the incubated eggs should be covered with a black paper until 8-10 days. The temperature and relative humidity will be kept around 24-25°C and 90-95 % respectively. Then after “hakitate” or feeding the newly hatched larvae will be done by the help of a bird’s feather.

For determining hatchability the formula will be:

Hatchability % = successfully hatched larvae / total number of eggs X 100

#### **3.3.2 Disease resistant race**

The larvae will be reared from hatching till cocoon formation. Finely chopped mulberry leaves will be given to the worms and the sizes of leaves gradually increases as they grow in size.

During rearing, as mentioned already the disease resistant race will be identified by observing the least attacked race by the diseases. The lowest infected race will be considered as highly resistant race. Mostly three diseases namely NPV, CPV and muscardine will be considered in this study.

#### **3.3.3 Quality of the produced cocoons**

Both the physical and commercial characters of cocoons will be concerned for the determination of quality of then produced cocoons. After the 5<sup>th</sup> instar larvae, they undergo moulting and larvae started spinning cocoons. Various parameters like cocoon shape, weight, wrinkles, weight of cocoon shell, shell ratio, double cocoons (number of defective cocoons) etc were recognized for determination of quality of cocoons. Apart from this the shell ratio also determines the quality of the produced cocoons.

The shell ratio will be calculated by the formula:

Shell ratio = weight of cocoons / total weight of cocoon's shell x 100

### **3.3.4 Data analysis**

The data was analyzed by using Microsoft Excel 2010.

## 4. RESULTS

### 4.1 Hatchability

The hatchability percentage of five selected silkworm races are found to be variable. The number of newly hatched worms among the total number of eggs and is then multiplied by 100 for calculating the hatching percentage. The hatching percentage of five selected silkworm races were found to be:

Table 1.Hatchability % in five races

Hatchability	K1	K2	K3	STD1	STD2
%	97	92	90	87	85

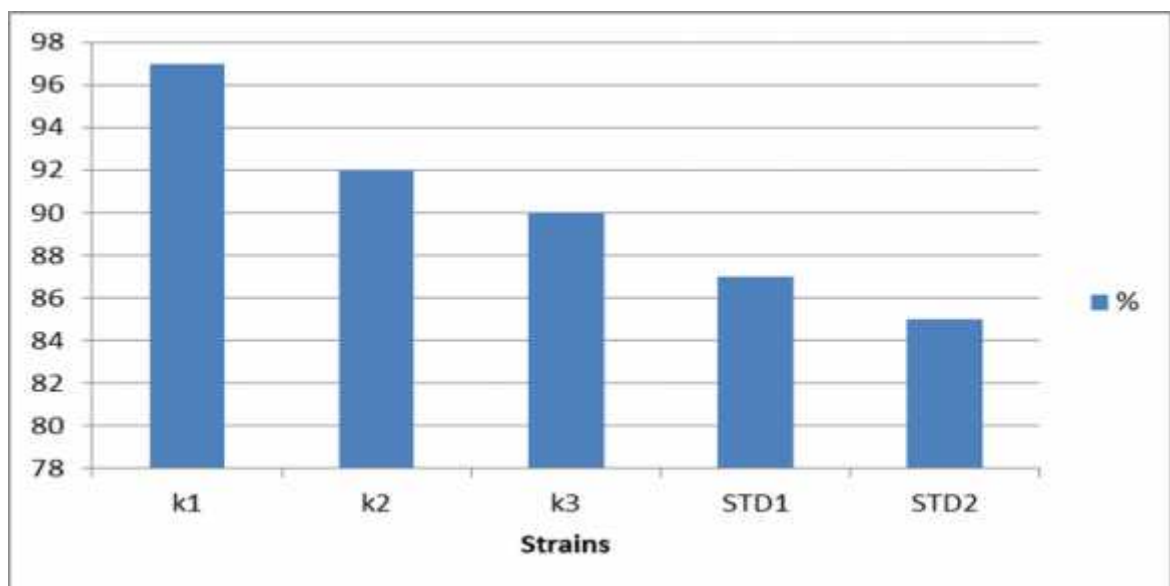


Figure 1: Hatchability of five silkworm races

This shows the hatching percentage and it was found to be maximum in K1 with 97% while it was minimum in STD2 with 85 %.

## 4.2 Disease resistant races

The record on diseased larvae on five trays of each variety are :

Table 2. Number of diseased larvae in five races.

Date	K1	K2	K3	STD1	STD2
3/4/2071	X	X	6	X	X
3/5/2071	5	5	2	1	X
3/6/2071	5	2	4	X	X
3/7/2071	3	1	X	1	X
3/8/2071	5	1	X	X	X
3/9/2071	1	2	2	2	X
3/10/2071	2	1	X	X	X
3/11/2071	X	1	1	X	X
3/12/2071	4	X	X	1	2
3/13/2071	X	4	1	X	X

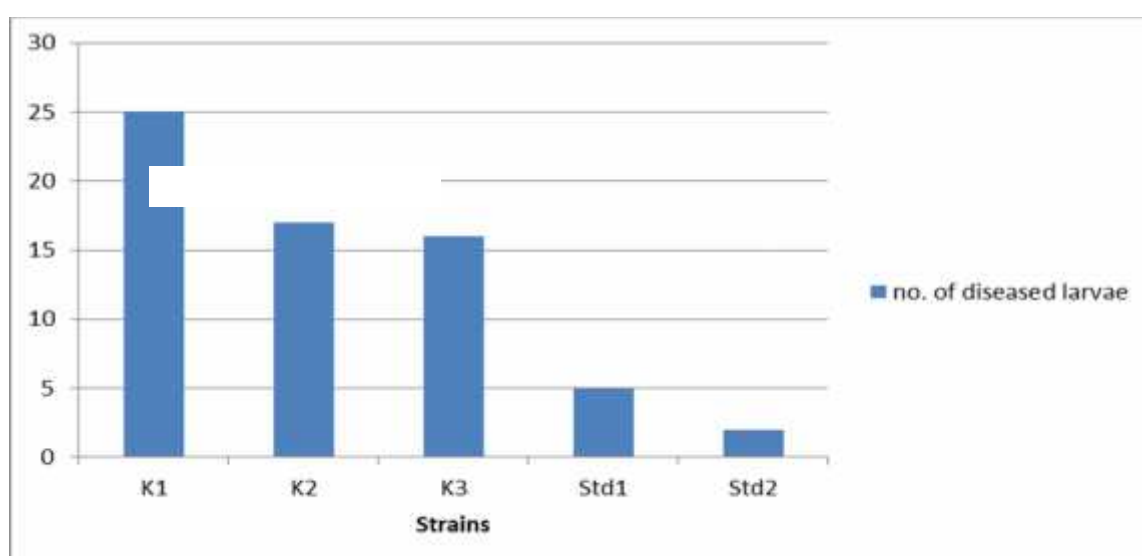


Figure 2: Number of diseased larvae in five races



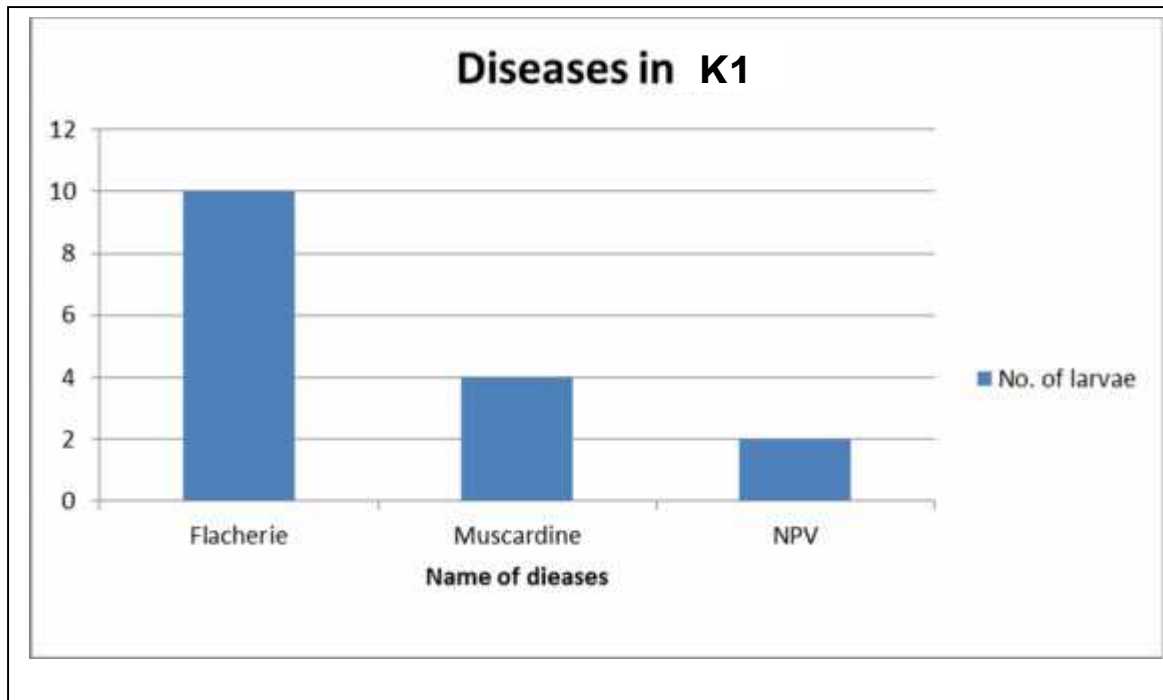


Figure 3: Diseases privileged in K1 only

From the above table the total number of diseased worms in each race was found to be 25 in K1, 17 in K2, 16 in K3, 5 in STD1 and 2 in STD2. Among those five races K1 got highly infected. There are altogether 10 flacherie, 4 muscardine and 2 NPV. Presence of milky white secretion by the larvae and those larvae feeding low amount of mulberry leaves is considered CPV disease. The larvae which have lost the caudal horn located at the posterior part of the larval body and greenish anterior part is considered NPV disease. While the appearance of dark brown line segments on both dorsal and ventral surfaces of the body is recommended as muscardine (Singh 2058). In this study as already mentioned the disease resistance race will be identified by observing the least attacked race by the diseases. The lowest infected race will be considered as highly resistant race. Therefore, race STD2 was highly resistant to diseases and K1 was least resistant. The diseases were recognized by the experts of Entomology Division, NARC, Khumaltar and supervisor Mr. Singh on the basis of external features and symptoms of the diseases and mostly flacherie and muscardine were the prime factors that cause death of the larvae from table 3.

### 4.2.1 Hatchability vs disease resistant race

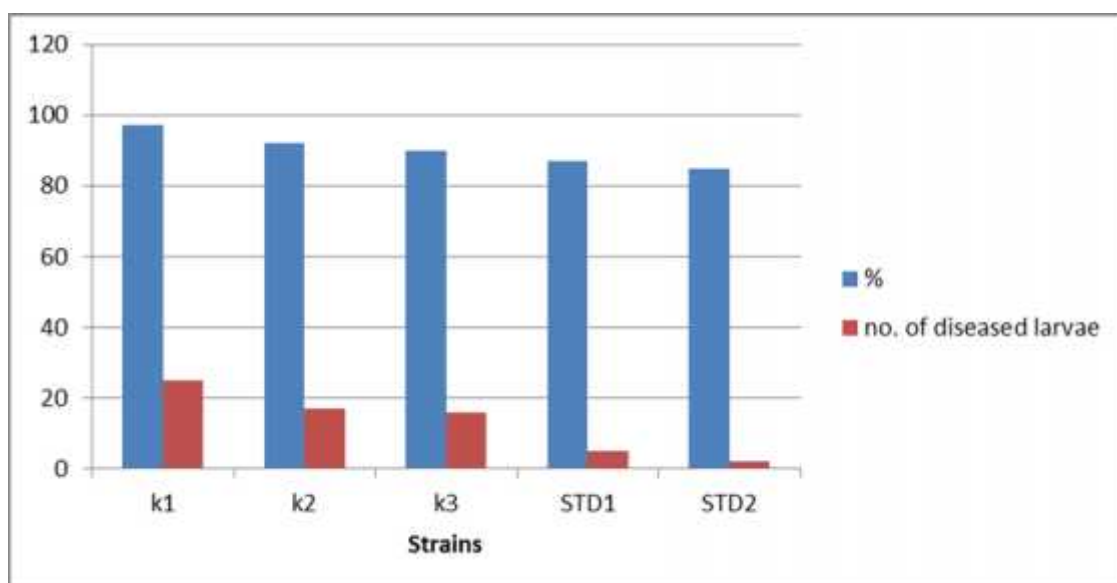


Figure 4: Hatchability and disease resistant race

This chart clearly shows that if hatchability is more, the disease is also found to be more and is considered as least resistant race. While if hatchability is less, then there is less number of diseases and finally it is considered as more resistant. Therefore K1 race is recommended as least resistant and STD2 is considered as highly resistant among the five races.

### 4.3 Quality of the produced cocoons

The shell ratio value gives the exact idea about the quality of cocoon. The race with highest shell ratio was considered as the most feasible race among five selected races while the race with lowest shell ratio as the least feasible one. After the 5<sup>th</sup> instar larvae, they undergo moulting and larvae started spinning cocoons. Beside the shell ratio, various parameters like cocoon shape, weight, wrinkles, weight of cocoon shell, shell ratio, double cocoons (number of defective cocoons) etc were also recorded.

For determining the quality of cocoons the formula for shell ratio is:

$$\text{Shell ratio} = \frac{\text{weight of cocoon}}{\text{total weight of cocoon's shell}} \times 100$$

### 4.3.1 Physical characteristics of silkworm cocoon

Table 3. Physical characters of silkworm cocoon

Items	K1	K2	K3	STD1	STD2
Colour	White	White	White	White	White
Shape	Oval	Oval	Oval	Oval	Oval
Grains/wrinkles	X	X	X	X	X
No. of defective cocoons	X	X	X	X	X
Double cocoons	X	1	X	X	X
Urinated cocoons	X	X	X	X	X

The quality of the produced cocoons can also be identified by following table by using the shell ratio.

Table 4. Quality of produced cocoons by using shell ratio

Items	K1	K2	K3	STD1	STD2
Total larvae after hatching	200	200	200	200	200
Total no. of good cocoons	175	183	184	196	198
Good cocoon %	87.5	91.5	92	97.5	99
Total no. of bad cocoons	25	17	16	5	2
Bad cocoon %	12.5	8.5	8	2.5	1
Average single cocoon wt. (gm)	2.21	2.15	2.1	1.96	1.86
Wt. of 10 male cocoons (gm)	98	170	260	20	5
Wt. of 10 female cocoons (gm)	212	350	290	130	6.4
Wt. of 10 male cocoon's shell (gm)	10	20	30	5.1	1.5
Wt. of 10 female cocoon's shell (gm)	30	35	40.3	2.9	1.3
Male shell ratio (%)	9.8	8.5	5.1	3.9	3.3
Female shell ratio (%)	7	10	7.1	4.4	4.9
Average shell ratio (male + female)	8.4	9.2	7.8	4.1	0.6

From the above data the quality of the produced cocoons was determined for this the shell ratio was calculated. The shell ratio was found to be highest for K2 race while it was lowest for STD2 race. Therefore K2 is regarded as the best quality of cocoon that can be reared among that five races of silkworm from this study.

Hence from above data it has been clear that K1, K2 and K3 were found to be of good quality while STD1 and STD2 were bad with respect to K1, K2 and K3.

## 5. DISCUSSION

Silkworm has been reared for over 5000 years in China (Nagaraja and Goldsmith 2002). Silk production increased steadily in the context of the world (Herald 2007). Sericulture is very important for the development of especially villages in Nepal too (Krishnaswami et al. 1973). Mulberry (*Morus* spp: Moraceae, Utricales) is the sole host plant for silkworm (Zhisen et al. 1999). Mulberry variety also play a great role in determination of cocoon characteristics (Bari et al. 1985) and thus the silk production (Krishnaswami et al. 1973). The larval hatching is better in vitellogenin- deficient eggs of the silkworm (Yamashito et al. 1980). The temperature and relative humidity also are the major factors that influence the silkworm and are provided with genetic resistance to prevent from diseases (Zafer et al. 2013). Normally insect reared at 36-37°C temperature are mostly free of disease and can survive (Aruga et al. 1963). Along with disinfection, use of resistant variety of silkworm is usually preferred (Nataraju et al. 2005).

The mulberry leaves with artificial diet containing autumn harvested leaves are more advantageous than those ones reared on other seasons (Watanabe 2003). Apart from this silkworms are provided with antimicrobial factors in the digestive juice. Also insect integument act as barrier for microbes such as bacteria, virus and protozoa that inhibit the growth and penetration of microorganisms (David 1967). A study revealed that bioassay showed maximum resistance to mosquito species (Huang, et al. 2015). For improving disease resistant species, the transgenic technology is a powerful tool (Jiang et al. 2012). Among many bacterial diseases cytoplasmic polyhedrosis virus seem to be more infectious toward silkworm than nuclear polyhedrosis virus (Watanabe 1966). This is because silkworm possesses CPV resistant genes with them (Watanabe 1986). The resistance to CPV infection is generally controlled by polygenes (Watanabe, 2002). Beside this Buhroo (2012) told that they have also immune mechanism to resist various diseases. Sometime disease very often causes severe economic loss (Sinha et al. 2008). The success of cocoon production depends on disease management procedure (Nirupama 2014). The cocoon spinning behavior varies from strain to strain (Kiyosawa et al. 1999).

On the basis of the results obtained during the study period, the lowest and highest hatchability were found to be STD2 and K1 race respectively under the controlled condition of temperature and relative humidity.

While K1 race is recommended as least resistant to disease as it have less number of diseased larvae and STD2 is considered as highly resistant to disease as it have highest infection among the five selected races. At last as per the results obtained from the shell ratio, the quality of produced cocoons were found to be best for race K2 and least good for STD2 race.

## 6. CONCLUSION AND RECOMMENDATIONS

### CONCLUSIONS

The quality of the silk or cocoons depend upon both the quality and quantity of mulberry leaves given to the worms during feeding. Hatchability was greatly affected by the variance of temperature and relative humidity. The climatic conditions also make the rearing of larvae easier if suitable techniques were applied. Disease resistance of silkworm races were also determined by the polygenes and the environmental factors. The worms possess resistance genes located usually in the digestive juice. The variation in hatchability, disease resistance and thus the quality of produced cocoons might result due to various factors like sanitation during rearing, amount of leaves given, quality of leaves, techniques of rearing, use of pesticides in mulberry cultivation etc. On the basis of shell ratio value the quality of produced cocoons were identified. Although from this research K1 was considered as highest hatchability, K2 as best cocoon quality and STD2 as disease resistant race, they might vary as per the place of rearing, season, rearing method etc.

### RECOMMENDATIONS

The recommendation of the present investigation “Hatchability, disease resistance and quality of the produced cocoons among five selected silkworm races in Sericulture Development Division, Khopasi” are summarized as below:

- ) Proper size of mulberry leaves for different instar larvae should be given as the small ant worms are not able to eat large and matured leaves.
- ) The balance between the environmental conditions and proper sanitation should be strictly maintained as they might get attacked by diseases.
- ) Covering the eggs with a black soft cloth must be done for uniform and profitable hatching results.
- ) The weight of the respective cocoon and cocoon's shell must be done with proper care and attention as there might be misunderstanding between the races while calculating the shell ratio.
- ) The chemical pesticides should not be encouraged in the mulberry orchard as they were directly eaten by the silkworms. In case there must be proper care in choosing



the pesticides to the sprayed, their dosages, time of application and waiting period after application of pesticides. Instead bio-pesticides should be advisable.

## 7. REFERENCES

- Ahashan, J and Singh, S.P 1982. Handbook of Economic Zoology, Chand and Company, New Delhi, India.
- Ahsan, M.M. and Sen, S.K. 1974. Tasar culture. Ambika Publisher, Bombay, India, 266 p.
- Akiduki, Gaku. 2010. Egg extract promotes cell migration and growth in primary culture of early embryos in the silkworm. Applied Entomology and Zoology, *Bombyx mori* .**117**.
- Ali-Ali, A. and Mahdi, M.T. 1973. Rearing a Japanese strain of *Bombyx mori* L. (Lepidoptera: Bombycidae) in Iraq **5**:33-48.
- Anonymous, 2001. Annual Technical Report. 1999-2000. Entomology Division, Nepal Agricultural Research Council, Khumaltar, Lalitpur, Nepal.
- Anonymous, 2003. Study visit in Sericulture institute of India. Entomology Division, Nepal Agricultural Research Council, Khumaltar, Lalitpur, Nepal.
- APROSC and John Meller Associates. JMA. 1996. Nepal Agriculture Perspective Plan (Final Report). National Planning Commission, Kathmandu, Nepal.
- Aramwit, P. 2010. Properties and antityrosinase activity of sericin from various extraction methods. Biotechnology and Applied Biochemistry. **55**.
- Arunachalam, V. 1994. How far can improvement strategies in sericulture be patterned on those in agriculture? In: Srirama Reddy, G. (Ed.). Silkworm Breeding, Proceedings of the National Workshop, Oxford and IBH Publishing, New Delhi, 354 p.
- Ashiru, M.O. 2002. The effect of mulberry varieties on the performance of Chul Thai five silkworm races. Discovery and Innovation **14**: 77-83.
- Bari, M.A., Ali, R. and Quiyyum, M.A. 1987. Effect of different plantation spacing on leaf yield of mulberry varieties. Proceedings of the 12<sup>th</sup> Annual Bangladesh Science Conference. Dhaka (Bangladesh). BAAS, 9 p.

Basi, M.A., Islam, R. and Salam, M.A. 1985. Feeding effects of three mulberry varieties on Nistari Race of Silkworm, *Bombyx mori* L. Bangladesh Journal of Zoology (Bangladesh) **13** (1):13-17.

Benchamin, K.V. and Nagaraj, C.S. 1987. Silkworm rearing techniques. In: Jolly, M.S. (Ed.). Appropriate Sericulture Techniques. International Center for Training and Research in Tropical Sericulture, Mysore, India. 176 p.

Benchamin, K.V. and Sen, Rathna. 1990. Is local variety of mulberry nutritively inferior to Kanva-2? Indian Silk **29** (3): 35-6.

Bizhannia, A.R., Ghanipoor, M. and Qotbi, A.A.A. 2008. Investigation on improvement possibility of resistance production traits in 3p, 2p and p generations in 3 Japanese pure lines of silkworm *Bombyx mori* L. using individual selection. Asian Journal of Animal Veterinary **3**: 443-447.

Boraiah, G. (Ed.) 1994. Lectures on Sericulture, Second Edition. SBS Publishers Distributers, Banglore, India, 307 p.

Booker, L. 2014. Chinese Silk, Japanese silver and Portuguese ships. Algarve History Association.

Buhroo, Z., Shabir, A.W., Malik, M.A. and Ganai, M.A. 2012. A review: Disease resistance in mulberry silkworm *Bombyx mori* L. Asian Journal **6**.

CBS. 2001. Statistical Year Book of Nepal. Central Bureau of Statistics, Kathmandu, Nepal.

Chattopadhuaua, S. 1994. Productivity analysis of hybrids of mulberry silkworms (*Bombyx mori* L.). Indian Journal of Entomological Research **18** (3): 261-263.

Chartterjee, S.N., Nagaraj, C.S. and Giridhar, K. 1990. An approach to silkworm breeding. Central sericultural Research and training institute, Mysore, 11-16 p.

Chen, K.P. and Lin, C.Q. 1991. Resistance of preserved silkworm races to NPV disease. *Acta sericologica* **17**: 45-46.

Cooper, E.K. 1962. *Silkworm and Science*. Lutterworth Press, London, U.K.

Cornaby, B.W. 1976. Feeding behavior of larval silkworms on different shapes of mulberry leaves. *Environmental Entomology* **5** (3): 595-598.

Das, B.C. and Sikdar, A.K. 1972. Evaluation of some improved strains of mulberry by feeding experiment. *Indian Journal of Sericulture* **9** (1): 26-30.

David, W.L. 1967. The physiology of insect integument in relation to the invasion of pathogens in "Insects and Physiology". Beament and Treherena Eds. London, 17-35 p.

Devkota, B. 1926. Quality and quantitative variations in the production of cocoon and raw silk by mulberry silkworm (*Bombyx mori* Linnaeus) due to different feeding frequencies. MSc. Thesis. Central Department of Zoology, Tribhuvan University, Kathmandu, Nepal.

Fenemore, P.G. 1992. *Applied Entomology*. Alka Prakashan

Ganesh, N.P., Ishikawa, T. and Yamamoto, S. 2008. *Bombyx mori* midgut membrane protein P 252, which binds to *Bacillus Thuringiensis*, Cry 1 A, is a chlorophyllidae-binding protein and resulting complex has antimicrobial activity. *Applied Environmental Microbiology*, 1324-1331 p.

Ganga, G. and Chetty, S. 1997. *An Introduction to Sericulture*, Oxford and IBH Publishing Co. Pvt. Ltd.

Gangwar, S.K. 2011. Relative susceptibility of silkworm breeds to different diseases in spring season. *Society of India* **2** (2).

Ghimire, N. 2000. Sericulture the context in Nepal. Paper presented in the National Workshop of Plant Protection Officers, Department of Agriculture, Kathmandu, Nepal, 221 p.

- Hisao, A. 1994. Principles of Sericulture. Oxford and IBH publishing Co. Pvt.
- Irie and Yamashita. 1980. Larval hatching from vitellogenin-deficient eggs developed in male hosts of silkworm. *Nature* **283**: 385-386.
- Jiang, L., Wang, G., Cheng, T., Yang, Q., Jin, S. and Lu, G. 2012. Reisittance to *Bombyx mori* nucleopolyhydrovirus via over expression of an endogenous antiviral gene in transgenic silkworms. *Archives of Virology* **157**: 1323-1328.
- Kafle, G.P. 1970. Notes on occurance behaviours and seasonal cycle of local silkworms *Bombyx huttoni* in Nepal. *Nepal Journal of Agriculture* **15**.
- Kiyosawa, M., Ito, E., Shiari, K., Kanakatsu, R. and Kiguchi, K. 1999. Cocoon spinning behavior in the silkworm, *Bombyx mori*: Comparision of three strains constructing different cocoons in shape. *Zoological Science* **16** (2): 215-223.
- Krishnaswami, M.N., Narasimhanna, S.K. and Kumararaj, S. 1973. Mannual on sericulture. Silkworm rearing UN Food and Agriculture Organization, Rome, Italy, 54-88 p.
- Krishnaswami, S., Noamani, K.R. and Asan, M. 1970. Studies on the quality mulberry leaves and silkworm cocoon crop production. Quality differences due to varieties. *Indian Journal of Sericulture* **9**: 1-10.
- Kumara, R.S. 1971. Further studies on double cocoons in *Bombyx mori L.* *Indian Journal of Sericulture* **7** (1): 70-71.
- Li, M., Yao, W., Hou, Q., Lin, C. and Chen, K.P. 2001. Studies of some characters in the silkworm *Bombyx mori L.* germplasm in China. *Sericologia* **41**:527-535.
- Liu, H.M., Yang, P.P., Wang, H.F., Liu, L.J. and Huang, X. 2015. Resistance level of mosquito species (Diptera; Culicidae) from Shandong Province, China. *International Journal of Insect Science*, 47-52 p.

Manandhar, D.N., Giri, P.P., Paneru, R.B. and Aryal, S. 2002. A Review on mulberry varietal performance, leaf quality and its effect on silkworms and cocoon production. Nepal Agricultural Research Council (NARC), Entomology Division, Khumaltar, Lalitpur.

Mayhew. 2007. The host defence of *Dorsophila melanogaster*. *Immunology* **25**: 697-743.

Meng, Z.Q. 1982. Inheritance of resistance to nuclear polyhedrosis by preoral inoculation in the silkworm (*Bombyx mori* L). *Acta sericologia sinica* **8**: 133-138.

MoAC. 1999. Impact of the agricultural extension program on agricultural production in Nepal. An evaluation of agricultural extension program in six high investments districts of Nepal. Ministry of Agriculture and Cooperatives, Evaluation Project Analysis division, Kathmandu, Nepal, 3-27 p.

Nagaraju, J. and Goldsmith, M.R. 2002. Silkworm genomics-progress and prospects. *Current Science* **83**: 415-425.

Nakao, H. 2010. Characterization of *Bombyx* embryo segmentation process. *Journal of experimental Zoology* **314** (3): 224-231.

Nigam, M.P. 1973. A note on some strange hatching behavior in silkworm (*Bombyx mori* L). *Indian journal of sericulture* **10** (1): 109-110.

Nirupama, R. 2014. Screening and identification of silkworm breeds of *Bombyx mori* L. for their resistant and susceptible against white muscardine. *Munis Entomology and Zoology* **9** (1): 510-517.

Palikhe, B.R., Ynagawa, H. and Yamaguchi, A. 2011. Illustrated technical manual on cocoon assessment. Promotion of quality cocoon production and processing project. JICA/DOIED, Hariharbhavan, Lalitpur.

Palikhe, B.R., Ynagawa, H. and Yamaguchi, A. 2011. Technical manual on silkworm disease management. Promotion of quality cocoon production and processing project. JICA/DOIED, Hariharbhavan, Lalitpur.

Park, K.J. 1973. A study on rearing season and cocoons yield for multirearing of silkworm (Suweon) **14**: 105-111.

Pedigo, L.P. 1996. Entomology and Pest Management. Prentice Hall of India, New Delhi, India.

Plant Protection Society Nepal. 2005. Integrated Pest Management and Plant Protection Strategy Development in Nepal. Hisi Printing Press. Lalitpur, Nepal.

Prasai, K. 1998. Study on food utilization and larval growth of mulberry silkworm. MSc. Thesis. Central Department of Zoology, Tribhuvan University, Kathmandu, Nepal.

Puri, A.K. 1990. Biological study on Eri-silkworm *Philosomia ricini* Boisd. MSc Thesis. Central Department of Zoology. Tribhuvan University, Kirtipur, Nepal.

Rahmathulla, V.K., Tilak, V.K. and Rajan, R.K. 2006. Influence of moisture content of mulberry leaf on growth and silk production in *Bombyx mori* L. Central Sericultural Research and Training Institute, Mysore, India.

Raju, P.J. and Krishnamurthy, N.B. 1993. Breeding of two bivoltines, MG511 and MG512 of silkworm, *Bombyx mori* L. for higher viability and silk production. *Seriologia* **33**: 577-587.

Rayamajhi, K.B. 1987. Biological study on Eri-silkworm *Philosomia ricini* (Boisd.) and its yielding. MSc Thesis. Central Department of Zoology, Tribhuvan University, Kathmandu, Nepal.

Rijal, S.P. 2003. Effectiveness of farmer's training for cereal crop production in Myagdi and Baglung districts of Nepal. MSc. Thesis. Institute of Agriculture and Animal Science. Rampur.

Chitwan. Nepal.

Sarkar, D. 1998. *Silkworm Biology, Genetics and Breeding*. Vikash Publishing House, New Delhi, India.

Sengupta, K. and Yusuf, M.R. 1976. Studies on the effect of spacing during rearing on different larval and cocoon characters of some multivoltine breeds of silkworm *Bombyx mori* L. *Indian Journal of Sericulture* **13** (1): 11-16.

Satoru, V. 1974. Effect of the air current in the spinning stage of the silkworm surrounded with high temperature and high humidity upon the reelability of cocoon. *Sericultural Science of Japan* **42** (2): 129-134.

Sen, R., Ahsan, M.M. and Datta, R.K. 1999. Induction of resistance to *Bombyx mori* nuclear polyhedrosis virus, into a susceptible bivoltine silkworm breed. *Indian Journal of Sericulture* **38**:107-112.

Shekharappa, B.M., Radhakrishna, P.G., Raghuraman, R. and Dandin, S.B. 1974. Consolidation of new rearing technologies in the field of productivity improvement by 2001. In: *Global Silk Scenario-2001*. IBN Publishing, New Delhi, 221 p.

Singh, A.B. 2058. Mulberry plantation development program. Agriculture Department. Sericulture Development Division, Khopasi.

Singh, M. 1924. Effect of food on the yield of raw silk in mulberry silkworm (*Bombyx mori*, Linnaeus). MSc. Thesis. Central Department of Zoology, Tribhuvan University, Kathmandu, Nepal.

Singh, B.D. and Sengupta, K. 1980. Temperature during pupation influences oviposition, fecundity and hatchability of silkworm *Bombyx mori*. *Science and Culture* **4**:192-193.

Singh, R. 2004. *Elements of Entomology*. Rastogi Publications.

Sinha, R.K., Kamble, C.K. and Koundinya, P. 2008. Mechanism of viral resistance in mulberry silkworm *Bombyx mori*. *Indian Journal of Sericulture* **47** (1): 1-6.



Tembhare, D.B. 1984. A textbook of insect morphology and endocrinology. S. Chand and Company Ltd. New Delhi.

Thapa, R.B. 2006. Sericulture development of fourth National Conference on Science and Technology, Nepal Academy of Science and Technology, Khumaltar, Lalitpur **1**: 2009-2026.

Tribhuwan, S. and Mathur, S.M. 1989. The main factor in mulberry that attracts the *Bombyx mori* L. Silkworm. Indian silk **28**: 39-40.

Venkatesh, R.H. 2007. Factors influencing oviposition of *Bombyx mori* Linnaeus . A Review of Bangladesh journal in Sericulture **1**: 1-12.

Verma, S.K. 1972. Comparative growth of silkworm *Bombyx mori* L. race reared on different mulberry varieties. Indian Journal of Agricultural Science **40** (12): 1097-1107.

Viren, Li. 1980. Land of silk and sericulture. China Reconstruct **24**: 7- 9.

Yamashito, O. and Irie, K. 1980. Larval hatching from vitellogen-deficient eggs developed in male hosts of Silkworm, Nature **283**: 385-386.

Yanagawa, H., Yamaguchi, A. and Palikhe, B.K. 2011. Technical manual on silkworm disease management, Promotion of quality cocoon production and processing project, JICA/DOIED. Harihar Bhavan, Lalitpur, Nepal.

Yiran, L. 1980. Land of Silk and Sericulture. China Reconstruction **39** (5): 7-9.

Yokahama, T. 1973. History of Entomology: Annual Review. Palo Alto, California, USA, 284 p.

Yu, W. and Ming, Y. 1982. Factors affecting the hatchability of eggs of the silkworm, *Bombyx mori* after short period of cold storage and acid treatment. Journal of Zhejiang Agricultural University **2**.

Watanabe, H. 1966. Relative virulence of polyhedrosis viruses and host- resistance in the silkworm *Bombyx mori* L.: (Lepidoptera: Bombycidae). *Applied Entomology and Zoology* **1**: 139-144.

Watanabe, H. 1986. Resistance of the silkworm, *Bombyx mori* to viral infections. *Agriculture, Ecosystems and Environment* **15**.

Watanabe, H. 2002. Recent advances in silkworm biology. *Current Science* **83**.

Watanabe, H. 2003. Genetic resistance of the silkworm, *Bombyx mori* to viral diseases. *Current Science* **83** (4): 439-446.

Watanabe, H. and Aruga, T. 1963. Inference by the heat inactivated virus on the cytoplasmic polyhedrosis virus in the silkworm *Bombyx mori* L. *Journal of Sericulture Science Japan* **32**:51-57.

Watanabe, H. and Aruga, T. 1971. The effect of moult and the development of nuclear polyhedrosis in the silkworm, *Bombyx mori* L. *Journal of Insect Pathology* **4**: 72-76.

Yao, Q., Li, M.W., Wang, Y., Lu, J., Dong, Y. and Chen, K.P. 2003. Screening of molecular markers for NPV resistance in *Bombyx mori* L. *Journal of Applied Entomology* **127**: 134-136.

Yokahama, T. 1973. *History of Entomology: Annual Review*. Palo ALTO, California, USA, 284 p.

Zhang, Y.N., Liu, S.X., Huo, Y.M. and Ou, S.Y. 1982. Identification of the resistance of certain silkworm races to six types of silkworm disease. *Acta sericologica sinica* **8**: 94-97.

Zhisan, J., Mengchang, T., and Jiaming, W. 1999. The determination of flavonoid contents in mulberry and their scavenging effects on superoxide radicals. *Food Chemistry* **64**: 555-559.

## PHOTO PLATES



Photo plate 1. Eggs being laid by female moth



Photo plate 2. Larvae feeding mulberry leaves



Photo plate 3. Pupal stage



Photo plate 4. Mating of adult moths



Photo plate 5. Feeding the ant worms



Photo plate 6. Sericulture Development Division, Khopasi