Silkworm Seed Production

Dr.H.B.Mahesha, Yuvaraja's College, University of Mysore, Mysuru.

The aim of a grainage is the production of quality seeds. This process involves different steps and they are described as below.

Procurement of seed cocoons in the seed market:

The basic criteria for procurement of seed cocoons are that it should be pebrine free and confirm to the norms, especially regarding the pupation rate. The norms are fixed according to the season for the characters such as cocoon weight, pupation rate and racial characters. The cocoons must be uniform with regard to size, shape and colour. The grainage authorities must enquire about the nature of the rearing, environmental conditions during the rearing and healthyness of the cocoons before the selection of cocoons have been made.

In the cocoon market, the grainage authorities will purchase the required quantity of multivoltine cocoons in an open auction with the presence of govt. market officer as per law. Half quantity of the bivoltine cocoons should also purchased from bivoltine cocoon markets for the preparation of hybrid seeds.

The seed cocoons are packed loosely in perforated boxes or bamboo baskets in small quantities and are transported during cooler hours of the day. If the live cocoons are tightly packed, they get heated up due to respiration. Exposure of hot sun results in the dead cocoons and poor emergence.

Preliminary examination/ selection and sorting of seed cocoons

In order to produce good quality and healthy eggs the seed cocoons used for the purpose must be of high quality and in good health, and therefore the seed cocoons arriving at the grainages are subjected to rigid selection. In selection only sound and uniform cocoons conforming to the characteristics of the race of the parental stock are selected and defective and deformed, under and over sized cocoons, double, stained and dead, uzi infested, thin end, open end, melted cocoons etc. are rejected.

Advance detection of pebrine disease, if any before the commencement of operation of each batch helps in averting great loss to the grainages. This is facilitated by investigations at three stages

1. Pupa test
2. Forced eclosion test
3. First day moth examination.

Of the three tests, only pupa test is explained in detail.

Pupa Test

In this test, the gut of pupa is a more reliable test for pebrine detection than the entire pupa. A sample of pupae is tested from each batch of cocoons.

For this purpose, the pupa is cut ventrally just below the wing bud by a scissor by holding the pupa between thumb and for finger in left hand. After cutting the pupa is pressed gently. The midgut oozes out as a brown body from the cut portion. This midgut is collected and crushed with few drops of potassium hydroxide in a moth crushing set. The fluid is taken on the slide and examine under the microscope with 600 X magnification. If the stock is suffering from pebrine, the entire batch of cocoons is rejected and sent to market. Such cocoons should never be used for preparation of silkworm seed under any circumstances.

Preservation of seed cocoons

If the seed cocoons preserved improperly, it has an impact on the eclosion (emergence) rate, fecundity and viability of moths. The cocoons are preserved in

single layer in well ventilated rooms under natural light and dark conditions. Exhaust fans in cocoon preservation and emergence rooms are essential to expel foul gases and dust. Different component races are to be preserved in separate rooms.

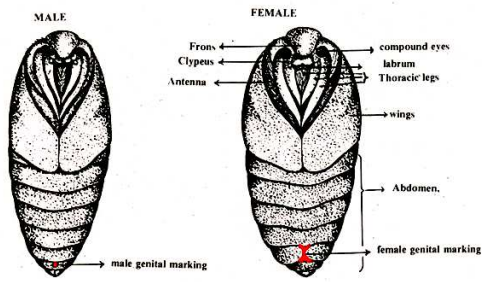
Sex Separation at pupal and moth stage:

In the case of preparation of industrial hybrid seeds, the sexes must be separated before selfing occurs in the same parental race. This sexing may be carried out either in the larval or pupal or moth stage.

In case of pupal stage sexing, the cocoons are deflossed and cut at one end to remove the pupa for determination of sex. Cocoon cutting improves eclosion per cent age in case of bivoltines, where the shell is hard and compact. Multivoltine cocoons are however, more flossy with loose shell and one prove to pupal damage if cocoon cutting resorted to. Male and female pupae are separated in the pupal stage based on the following differences.

Identification of male and female pupae

Male	Female
1. Smaller in size	1. Larger in size
2. Narrow pointed abdomen	2. Broad abdomen
3. Small dot like mark on the ventral side near the top demarcation line of 9th abdominal segment.	3. X mark seen on the ventral side of 8th abdominal segment



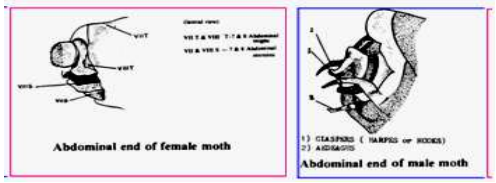
male and female pupa – *Bombyx mori*

In case of sex separation at moth stage, generally the males emerge first and can be picked before the females emerge and kept separately. Later when the females also emerge, one should be ready to pick out males and females separately so to prevent their mating within the same parental stock. For sex separation the main differences between the sexes are given below.

Identification of male and female moths

Male	Female
1. More active	1. Less active
2. Smaller in size	2. Larger in size
3. Bigger antennae	3. Smaller antennae
4. The caudal end has a pair of hooks known as harpes helping in copulation	4. The caudal end has a median knob like projection with sensory hairs-which is protruded & retracted to expel the pheromones (Ovipositor)

FEMALE & MALE MOTHS



ABDOMINAL END OF FEMALE & MALE MOTHS

Different races of silk moths emerge at different intervals and it is about 12-14 days also it is depending on the environmental conditions (Temperature). The cocoon/pupa preservation rooms should be kept dark a day before emergence and bright light is switched on in the early morning by about 5 or 6 AM on the day of emergence. Each day's emergence will be over by about 8 AM. The emergence of each lot generally extends over 3-4 days. After collecting all the moths, the room should be kept dark again until next day morning. The males generally emerge earlier and should be picked up and kept in separate trays for mating purpose.

The moths that have paired (selfed) due to error in sex separation, deformed and weak moths should be rejected while picking. The healthy moths can be collected from the paper cover easily. If excess males are found, they can be stored at 5°C for later use. The excess females can also be stored likewise which however is not advisable.

Synchronization

Moths of the component races are made to emerge on the same day, so that male and female moths are readily available for hybridization. This is referred to as synchronization. Planning for synchronization should start at the brushing time of the parent races. However, there may be some difference in spinning due to rearing conditions of the farmers. In such cases, emergence of moths in the two races may be adjusted by selecting cocoons of matching date. In case of synchronizing batches are not available; emergence of the earlier batches can be delayed by refrigerating the cocoons. The cocoons or pupae should be refrigerated preferably on the 7^{th} or 8^{th} day after spinning *i.e.*, 4-6 days after pupation, at 5-10°C. Such refrigeration should be limited to 3 days for females and 7 days for males.

The emerged moths can also be refrigerated for the same purpose. The moths may be refrigerated at 5°C up to 10 days in case of males and 2-3 days in case of females. The refrigeration of females either in the pupal or moth stage should be done only as last resort. Refrigeration should be restricted to any one stage, either in the pupal or moth stage.

Coupling and decoupling

As soon as the moths emerged they start to get pair. For the preparation of hybrid seed sexing should be carried out before pairing.

The female moths are spread in a tray and the male moths of the desirable hybrid component are evenly distributed over the females. Generally excess males are distributed to facilitate quick mating. In about 15 minutes, the male and female moths pair. The excess males and the unpaired females are picked up and allowed to mate in a separate tray. The excess males are collected for cold storage. Coupled moths are left undistributed for three hours under normal day light conditions. The temperature and humidity should be maintained at 23-25°C and 75% respectively. It is advisable to conduct mating between 8 AM to 12 Noon.

After the required period of coupling, the pairs are separated by holding the female moth and gently sliding the male. This facilitates easy separation without injury to the female reproductive organs. The tray containing decoupled female moths may be gently tapped to induce urination.

Although 3-4 hours of mating is sufficient, it is desirable that the coupling period is extended to 6-8 hours to get maximum number of eggs laid in shortest period.

Oviposition

This is nothing but the process of egg laying by the female moth.

When eggs are required to be laid on cards, the mated females are placed on the egg sheets and each moth is enclosed in a cellule. Cellules are conical in shape and are usually made of black PVC. The inner surface of the cellule should be smooth to prevent clinging of moths. Egg cards are designed to accommodate 20-28 moths, serially numbered on each sheet. The cellule isolates the eggs laid by each moth, facilitate individual moth examination and elimination of eggs laid by diseased moth.

In case of preparation of loose eggs the cellules are not required. In this case, a unit number of female moths are allowed to lay eggs on starched paper or cloth, with in a wooden or plastic frame. The number of moths vary from 30-200 according to the health of the batch and convenience.

Moths are allowed to lay eggs for 24 hours in a dark room where the temperature and humidity are maintained at 25°C and 75-80% respectively. In case of moths do not lay eggs until next day morning they are again coupled with a new male moth. In such case some female moths lay eggs, are removed in to the moth preserving boxes. Then mother moths are examined for the pebrine disease and eggs free from pebrine disease are qualified for rearing.

After mother moth examination, the egg sheets or loose eggs in a container are dipped in 2% formalin solution or 500 ppm of ClO_2 (Chlorine Dioxide) solution. This helps in eliminating the possible risk of surface contamination of eggs by various pathogens and formalin increases the adhesive capacity of eggs to the egg sheets. Subsequently, the sheets are washed and dried under shade and preserved under optimum temperature (25°C) and RH of 80%.

Now the eggs are ready for incubation.
