# Effect of Exposure of the Silkworm, Bombyx mori, To High Temperature on Survival Rate and Cocoon characters

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#### Abstract

The silkworm, *Bombyx mori*, at various larval stages was exposed to high temperature (36°C) for 48 h, and the effects on the survival rate and cocoon characters were investigated. The heat treatment in larvae in each instar caused a reduction in the survival rate, and the heat susceptibility increased with age. Within the same instar (from 1st to 4th instars) heat treatment of the larvae at early or later stages resulted in a much higher survival rate than that of larvae treated at the middle stage. Results of heat experiment conducted at various times during the 5th instar showed that larvae treated in the early half of the middle period and late stage before mounting were more susceptible to heat which reduced their survival rates compared with the larvae treated in the other periods. Heat treatment applied at various times in larvae also reduced the cocoon weight and cocoon shell weight, and the period of heat susceptibility was almost similar to that related to the reduction of the survival rate. When the cocoon weight and cocoon shell weight were reduced markedly by the heat treatment, the cocoon shell ratio tended to show a high value. Heat treatment applied to the larvae in each instar (from 1st to 5th instars) prolonged or shortened the duration of the feeding period by less than one day. Within the same instar, heat treatment at the early stage prolonged the duration of the feeding period, whereas the treatment at later stages shortened the feeding period. The results of the present study indicated that the heat treatment affected the survival rate and cocoon characters depending on the larval stage at which the treatment was applied, and that the 5th instar larvae at the early stage were most susceptible to the heat treatment. Accordingly, for testing the heat resistance of the silkworm, it is recommended to expose 5th instar larvae at the early stage to high temperature  $(36^{\circ}C)$  for 48 h.

**Discipline:** Sericulture **Additional key words:** heat resistance

# Introduction

Sericulture has been one of the major fields of agriculture in upland villages in Japan. Recently, however, due to labor shortage in sericulture, rough rearing of the silkworm under adverse conditions has tended to prevail, resulting in low yield of cocoons and increase of the number of cocoons with poor quality. Against this background, there is a great demand for the development of a technology for labor-ultrasaving and low-cost sericulture, as well as breeding of silkworm varieties resistant to adverse rearing conditions.

In the present study, the effect of high temperature treatment of the silkworm larvae on the survival rate and cocoon characters was analyzed for the development of a method of testing for resistance to adverse conditions.

#### Materials and methods

Silkworm race, N  $137 \times C$  146, reared in early autumn was employed in the present study. Fifty larvae were sampled every day from hatching to mounting, and they were exposed to high tempera-

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ture (36°C) for 48 h in an incubator. During the heat treatment, younger larvae were kept in a plastic petri dish ( $25 \times 18 \times 3$  cm in size), while older larvae were kept in rearing cages ( $43 \times 32 \times 8$  cm in size), with a plastic lid. Moisture in both petri dish and rearing cage was kept at more than 90% RH during the treatment, and the larvae were fed fresh mulberry leaves twice a day.

After the heat treatment, the tested larvae were transferred to ordinary rearing conditions at 25-28°C and they continued to feed until mounting. After pupation, each tested larval group was investigated for the duration of the feeding period, survival rate based on the percentage of pupation, and quantitative characters of cocoon.

#### Results

#### 1) Survival rate

When the larvae in the 1st-3rd instars were subjected to the heat treatment, some of them died sporadically at the larval stage, at the time of mounting, and after spinning. In the larval group in which the heat treatment was applied in the 4th instar, few larvae died within the 4th instar, but most of the dead larvae were in the 5th instar and in the period after mounting. In the larval group in which the heat treatment was applied at the early stage of the 5th instar, dead larvae were mainly in the 5th instar, whereas in the group treated at the late stage of the 5th instar larval death occurred in the mounting period and in cocoons.

Relationship between the larval stage at which the heat treatment was applied and the survival rate is shown in Fig. 1. The survival rate of the larval

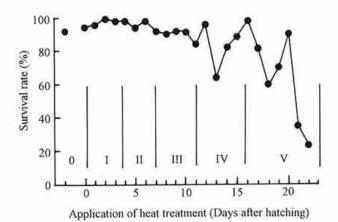


Fig. 1. Effect of heat treatment (36°C for 48 h) in larvae at various stages on survival rate 0: Untreated control, I-V: Instar.

group treated in the 1st-3rd instars tended to be higher than that of the larval group treated in 4th-5thinstars. The susceptibility to heat treatment was stage-specific within the same instar. The 4th instar larvae at the middle stage were most susceptible to the heat treatment which reduced the survival rate than larvae at other stages of the same instar. The 5th instar larvae treated with heat on the 2nd day or the 6th-7th day showed a remarkable reduction of the survival rate, while larvae treated on the 3rd-5th day or at the mature stage displayed a relatively high survival rate.

#### 2) Duration of feeding period

Fig. 2 shows the effect of heat treatment on the duration of the feeding period after hatching to mounting. The duration of the feeding period in control larvae was 23 days, while in the treated larvae the period varied from 22 to 24 days depending on the larval time at which the heat treatment was applied. For instance, the 4th instar larvae on the 2nd day were susceptible to heat, resulting in the prolongation of the duration of the feeding period, whereas in the larvae at the late stage of the same instar, the duration of the period was shortened. Heat treatment applied to the 5th instar larvae at the early stage (1st to 3rd days) tended to prolong the duration of the larvae treated at the later stage (after 6th day).

These results indicated that heat treatment applied to larvae in each instar at the early stage tended to prolong the duration of the feeding period compared with the larvae treated at the middle stage.

#### 3) Cocoon weight and cocoon shell weight

Effect of heat treatment on cocoon weight is

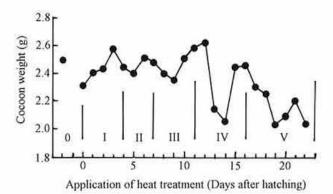


Fig. 2. Effect of heat treatment in larvae at various stages on duration of feeding period 0: Untreated control, I-V: Instar.

shown in Fig. 3. The larvae subjected to heat treatment during the 1st to 3rd instars produced cocoons with a weight ranging from 2.42 to 2.61 g. On the other hand, larvae subjected to heat treatment in the 4th instar produced cocoons with a weight ranging from 2.08 to 2.75 g, while the 5th instar larvae

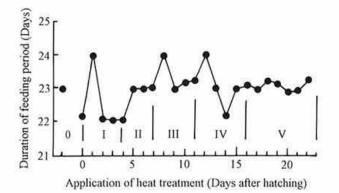


Fig. 3. Effect of heat treatment in larvae at various stages on cocoon weight
0: Untreated control, I-V: Instar.

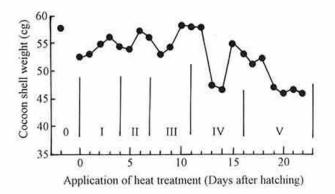


Fig. 4. Effect of heat treatment in larvae at various stages on cocoon shell weight
0: Untreated control, I-V: Instar.

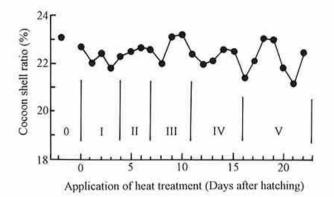


Fig. 5. Effect of heat treatment in larvae at various stages on cocoon shell ratio 0: Untreated control, I-V: Instar.

subjected to the heat treatment produced cocoons with a weight ranging from 2.06 to 2.44 g. Thus, a relatively large variation in cocoon weight was induced when the heat treatment was applied to 4th instar larvae, while the cocoon weight was most reduced by treatment of 5th instar larvae.

Heat susceptibility of the 5th instar larvae leading to a reduction of the cocoon weight increased gradually after ecdysis to the middle stage (3rd to 4th days) when the susceptibility was highest, and then decreased slightly to the later stages. Thus, the application of the heat treatment at the middle of the 4th or 5th instar promoted considerably the induction of a low cocoon weight.

Effect of heat treatment on the reduction of cocoon shell weight was very similar to that on the reduction of the cocoon weight as shown in Fig. 4, with variations depending on the larval stage at which the heat treatment was applied. When both cocoon weight and cocoon shell weight were reduced markedly by the heat treatment in larvae, the cocoon shell ratio tended to show a high value as depicted in Fig. 5.

## Discussion

High temperature is one of the main factors causing environmental stress in the silkworm. Kato et al.<sup>1)</sup> and Nagayasu et al.<sup>3)</sup> reported that the effect of high temperature on the survival rate, duration of feeding period, and cocoon characters of the silkworm varied with the larval stage at which the heat treatment was applied. Similar results were obtained in the present study.

Kato et al.<sup>2)</sup> compared the resistance to high temperature among various silkworm races, and identified 2 types of races, resistant and susceptible. Crossing tests between resistant and susceptible races indicated that the resistance to high temperature is a heritable character. Similar results were reported by Ninagi et al.<sup>4,5)</sup>. These studies suggest that it may be possible to breed silkworms resistant to high temperature.

Shirota<sup>6)</sup> selected silkworms resistant to high temperature based on the high percentage of pupation from the larvae treated with heat in the 5th instar, resulting in the development of a resistant race.

In the selection of silkworms resistant to high temperature, it is important to develop an appropriate method of testing for heat resistance. The results of the present study indicate that heat resistance can be evaluated based on the survival rate and cocoon characters of the larvae which were subjected to the heat treatment (36°C for 48 h) at the early stage of the 5th instar. Further studies on the methods of testing silkworm resistance to stresses other than heat should be carried out for the breeding of silkworms resistant to adverse rearing conditions.

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