IJIE Int. J. Indust. Entomol. 45(1) 22-28 (2022)
ISSN 1598-3579, http://dx.doi.org/10.7852/ijie.2022.45.1.22

ISSN 1598-3579 (Print)
ISSN 2586-4785 (Online)

Growth effects of novel heating system using heater-installed rearing tray on silkworm, *Bombyx mori*

Chan Young Jeong, Kee Young Kim, Nam-Suk Kim, Sang Kuk Kang, Jong Woo Park, Ik Seob Cha, and Seong-Wan Kim*

Industrial Insect and Sericulture Division, National Institute of Agricultural Sciences RDA, Jeollabuk-do 55365, Republic of Korea

Abstract

Silkworms are very sensitive to changes in temperature and humidity, and unless it is a suitable temperature and humidity to grow, the productivity and quality of silkworms are greatly reduced. Therefore, it is very important to manage temperature and humidity for silkworm feeding facilities. In particular, it is essential to install heating facilities in Asian countries with distinct seasonal changes. During the feeding period, many farms manage the temperature and humidity of feeding facilities by installing and using electric heaters inside the facilities. However, it is very difficult to manage the room temperature stably by the heaters. In addition, unlike the temperature inside the facility, silkworms could undergo severe temperature changes as the inside of the rearing tray could not be warm enough. In this study, in order to improve the previous heating method, the new rearing method that directly heats the bottom of the rearing tray was developed. Compared to the previous room-heating system, the novel heater-installed tray (HIT) system significantly reduced the change in temperature during the experimental period. In addition, the number of days of silkworm growth up to harvest was shortened, which was effective in growth performance, and it was also found that silkworms grew more uniformly in HIT system than in previous system. Moreover, as the heater tubes were installed directly under the rearing tray, it quickly dried mulberry leaves and silkworm feces after feeding, and as a result, the environment in the tray was greatly improved with decrease the labor of breeder. In conclusion, these results suggest that the heater-installed rearing tray method greatly improves silkworm quality, increases weight of silkworms, and final profits compared to the previous room heating system with electric heaters.

Int. J. Indust. Entomol. 45(1), 22-28 (2022)

Received : 10 Aug 2022
Revised : 14 Sep 2022
Accepted : 22 Sep 2022

Keywords:
Bombyx mori,
Silkworm breeding seat,
Heater-installed tray

Introduction

The silkworm breeding has a very long history. The oldest record of silkworm breeding is from China more than 4,600 years ago (Kurin, 2002). As the history of silkworm breeding is long, methods and techniques for silkworm breeding have been developed for a long time. The silkworm varieties have diversified according to climate, soil properties, and country

***Corresponding author.**
Seong-Wan Kim
Industrial Insect and Sericulture Division, National Institute of Agricultural Sciences, RDA, Jeollabuk-do 55365, Republic of Korea
Tel: +82-63-238-2846 / FAX: +82-63-238-3832
E-mail: tarupa@korea.kr

(Kumari *et al.*, 2011; Vijayan *et al.*, 2010). The reason why silkworm breeding has such a long history and development is, above all, that silkworms have high industrial value. Since a long time ago, silkworm silk has accelerated the textile industry and brought national economic benefits, and silkworm larvae, silk fiber, and pupae have been used as useful protein sources and medicinal materials (Herold and Scheibel, 2017; Kim *et al.*, 2019; Wang *et al.*, 2020; Jo *et al.,* 2020; Kim *et al.,* 2021). It was announced that silkworms are effective in diabetic nephropathy and play an important role (Zhang *et al.*, 2016). Recently, the technique for producing useful substances such as antiviral proteins in silkworm was developed, and the industrial value of silkworms is drawing attention again (Okano *et al.*, 2000; Ueda *et al.*, 1993). In addition, academia is discussing the possibility of studying human diseases using a silkworm as a model organism (Tabunoki *et al.*, 2016). However, despite many advances, silkworm rearing is laborious and difficult. Most silkworms are well known for eating only fresh mulberry leaves. In order to improve the discomfort of monophagy, efforts are being made to develop methods or varieties of polyphagous silkworms, but no meaningful results have been produced yet (Asaoka, 2000; Iizuka *et al.*, 2012; Zhang *et al.*, 2013). Silkworms are very sensitive to environments, especially temperature and humidity (Rahmathulla and Suresh, 2012). In addition, silkworms are very vulnerable to bacteria, fungi, viruses, and chemicals. In most cases, silkworms damaged from the outside die immediately or have no product value even if harvested. Therefore, it is very important for farmers to create and maintain the proper environment conditions for silkworm rearing.

In Asian countries with clear seasonal changes, it is essential to maintain the proper temperature and humidity for silkworm rearing. In order to prevent low temperatures during the autumnal season, electric heaters are installed inside the facility to prevent silkworms from being damaged by low temperatures in Korea. However, most heating facilities do not make the inside of the facility warm enough in autumn. Therefore, many researchers are constantly striving to improve the current breeding method and raise silkworms under optimal conditions. In this study, a new heating method was introduced to improve the problems of the existing heating method for silkworm breeding. The novel method applied boiler tubes which were installed under the breeding table to directly transfer heat. This study measured and analyzed the period of reaching 5th instar, growth performance, and weight to compare the growth effect of silkworms in new heating methods, and discussed the utilization of newly developed heating methods.

Materials and Methods

Silkworm strains

Silkworm *Bombyx mori*, Baekokjam was used for all experiments, and silkworm breeding was carried out in accordance with the breeding standards of the Rural Development Administration (RDA), except temperature. Fresh mulberry leaves were raised in 16L8D conditions. The experiment with uniformly grown 4^{th} instar silkworms was started, and they were raised until the 4^{th} day of the 5^{th} instar and measured to analyze growth performance.

Heating facility setting method

The previous heating method for raising silkworms installs electric heaters inside the facility. In the new heating method, boiler tubes with a diameter of 25 mm were installed at intervals of 40 cm under the breeding seat. Both heating systems operated heating at dawn when the air became cold. The measurement of temperature was measured by installing digital thermometer at regular intervals in the center of all breeding seats.

Measurement of silkworm growth

The period of silkworm growing was measured from the 2^{nd} day of 4^{th} instar (4L2D) silkworm to the first feeding of the 5^{th} instar silkworm using each heating system. The average and weight distribution of silkworm were calculated with the 4L2D, 7^{th}-fed 5L3D, 8^{th}-fed 5L3D and 5L4D silkworms in the two heating systems. In all experiments, 60 silkworms were randomly collected as 3 repetitive experiments in the conditions.

Statistical analysis

All experimental results are represented by Mean value (M) ± Standard deviation (SD), and a Oneway Analysis of Variation (ANOVA) using IBM SPSS Statistics 23 (IBM, USA) was performed, and the significance of each mean value was tested by Duncan's multiple ($P<0.05$).

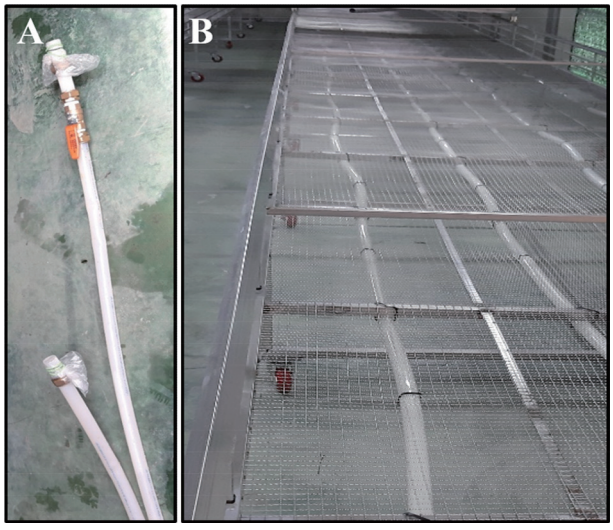

Fig. 1. Boiler tubes used in HIT system and silkworm rearing tray with them. (A) Boiler tubes with a diameter of 25 mm. An operated oil boiler allows hot water to flow through the tubes. (B) Boiler tubes were installed at intervals of 40 cm under the silkworm rearing tray.

Results and Discussion

The new heating system reduced the fluctuation of temperature changes in the breeding seats

The heater-installed rearing tray (HIT) system is a new heating method in which boiler tubes are directly installed under the breeding seat to transfer a direct heating effect (Fig. 1). In order to confirm the heating effect of the HIT system, the temperature inside the breeding seat were measured at intervals of 4 hours for 6 days during the experiment. The previous system which is heating a room with electronic heaters showed a drastic decrease in temperature at dawn and a rapid increase in temperature (Fig. 2A). During the experiment, the silkworm in previous heating system experienced a minimum temperature of 17.9 ℃ and a maximum temperature of 30.3 ℃, and the inside of the breeding seats recorded an average temperature of 23.9 ℃. On the other hand, the HIT system made the range of temperature changes inside the breeding seat moderate. The silkworms in the HIT system experienced a minimum temperature of 23.0 ℃ and a maximum of 29.3 ℃, and the average temperature inside the breeder was 25.6 ℃ (Fig. 2B). These results show that the previous heating system using electric heaters inside the facility do not heat the lowered temperature quickly enough, however, the HIT system is less affected by the outside temperature and can directly and quickly raise low temperature of the inside of the breeding seat.

The HIT system accelerated the growth of silkworms

The silkworms show differences in growth speed depending on the environment and the breeding period changes (Osanai, 1978). In particular, silkworms stop eating at low temperatures and greatly adversely affected by growth (Liang, 1985; Zhou and Wang, 1982). Considering the space, food, and labor for silkworm rearing, silkworm rearing is more economical as the breeding period is shorter. In this study, to confirm the growth promotion effect of silkworms in HIT system, the silkworm growth rate was measured and the change was analyzed by comparing the two heating systems.

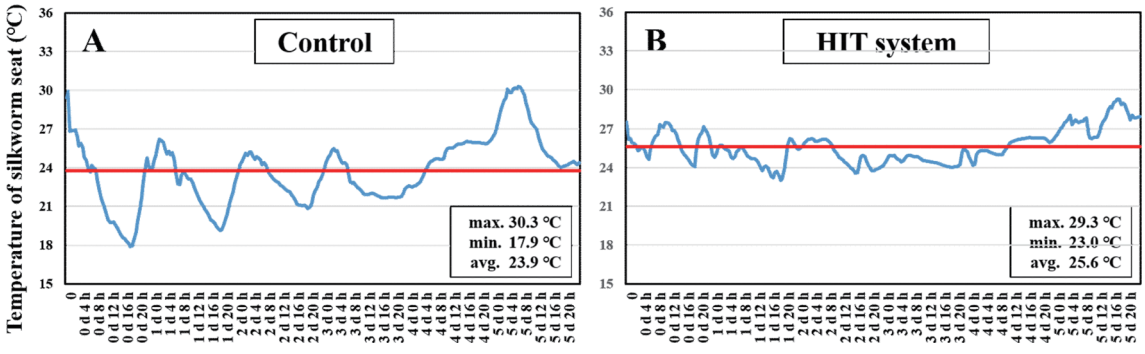

Fig. 2. Changes in temperature of silkworm seat in previous heating system or heater-installed tray (HIT) system. These experiments were conducted with 2nd day of 4th instar. (A) Changes in silkworm seat temperature of the control during experiment. (B) Changes in silkworm seat temperature of the HIT system during experiment. Blue line means the measurement temperature in the silkworm seat and red line means the average temperature in the silkworm seat. A digital thermometer was placed in the middle of the feeding tray, and the temperature was measured at intervals of 4 hours.

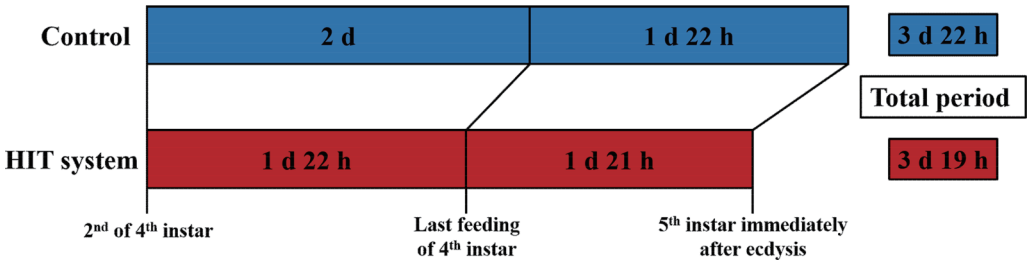

Fig. 3. Comparison of the period of reaching the 5th instar silkworm. The number of days taken from the 2nd day of 4th instar to 5th instar silkworm of the control or HIT system was compared. Compared to the control, it was shortened by 2 hours from the 2nd day of 4th instar silkworms to the last feeding of 4th instar silkworms in the HIT system, and finally, by 3 hours to the 5th instar silkworms.

Table 1. Comparison of the weight of silkworms in previous heating system or HIT system. Each of the 5th instar 60 silkworms after 7th feeding or 8th feeding was used for weight measurement. The error bar represents the standard deviation.

	2nd day of 4th instar		3rd day of 5th instar after 7th feeding		3rd day of 5th instar after 8th feeding	
	Wight (g)	Mean (g)	Wight (g)	Mean (g)	Wight (g)	Mean (g)
Control	0.4315 ± 0.0657	0.4011 ± 0.0623	1.4995 ± 0.2137	1.4448 ± 0.2395	1.6190 ± 0.2459	1.6670 ± 0.2137
	0.3915 ± 0.0618		1.4040 ± 0.1837		1.6660 ± 0.2400	
	0.3805 ± 0.0593		1.4310 ± 0.3212		1.7160 ± 0.1553	
HIT system	0.395 ± 0.0417	0.402 ± 0.0525	2.2000 ± 0.3384	2.214 ± 0.2334	2.3400 ± 0.2136	2.4400 ± 0.2293
	0.404 ± 0.0732		2.1620 ± 0.1942		2.5500 ± 0.2532	
	0.407 ± 0.0427		2.2800 ± 0.1676		2.4300 ± 0.2211	

The mulberry leaf feed amount of the two groups was the same. In the case of the HIT system, the warm air from boiler tube caused the silkworm seat to dry quickly. Rapid drying was prevented by feeding with mulberry shoots, and the amount of mulberry leaves was adjusted so that there was no problem with silkworms eating mulberry leaves. So in the HIT system, the amount of mulberry leaf didn't have to be particularly high. In the previous heating system, the period until 5L3D from 4L2D silkworm took 3 days and 22 hours. It took two days from the 4L2D to the last feeding of 4th instar, and 22 hours a day by 5L3D silkworm (Fig. 3A). However, in the case of silkworms raised by the HIT system, it took 22 hours a day from the 4L2D to the last feeding of 4th instar, and 21 hours a day to reach the 5L3D silkworm, which took a total of 3 days and 19 hours (Fig. 3B). These results show that silkworms in the previous heating method experienced low temperatures in which silkworm growth was inhibited, and thus had low growth performance, however, silkworms grew normally at a relatively moderate temperature in the HIT system.

The weight of silkworms in the HIT system was heavier than that in previous system

In order to determine whether the difference in growth speed due to temperature of the breeding seat leads to a change in the quality of the silkworm, the weight of the 5th instar silkworm after 7th or 8th feeding was measured. The weight of silkworm in the two groups showed a big difference when they were raised up to 5L3D from 4L2D silkworms (Table 1). The average weight of 5th instar silkworms after 7th or 8th feeding in the previous heating method was 1.4448 g and 1.6670 g, whereas the silkworms in the HIT system showed an average weight increase of 53% and 46% with 2.2140 g and 2.4400 g, respectively. These results imply that the HIT system, which regulates a stable temperature environment in silkworm breeding seats, has an effect on the growth performance of silkworm, showing a large difference in

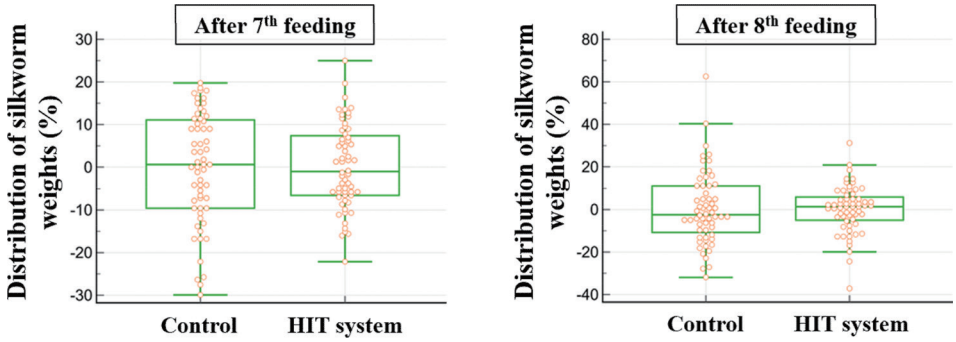

Fig. 4. The inter-quartile range (IQR) of the weight of silkworms by individual in previous heating system or HIT system. Each of the 5th instar 60 silkworms after 7th feeding (A) or 8th feeding (B) was used for weight measurement. The weight of each individual was converted according to the percentage ratio of the average and was indicated by yellow dots.

growth compared to room-heating system.

Comparison of weight distribution in two heating systems

It is important to grow silkworms uniformly as well as robustly in silkworm rearing. If silkworms do not grow uniformly, there is a difference in the feeding time within the same breeding seat, and as a result, it makes a difference in quality of harvested silkworms. As shown in Figure 4, the weight distribution of 5L3D silkworms after the 7th or 8th feeding had a difference in the previous method and the HIT system. The weight of silkworms raised in the HIT system showed a denser distribution of in the median value. Moreover, this tendency appeared stronger in the 5L3D silkworm after 8th feeding. This result suggests that the heating method of the HIT system makes silkworms grow more uniformly than the previous heating method. It is considered that the positive effect from moderate temperature treatment of HIT system during silkworm breeding is a key factor.

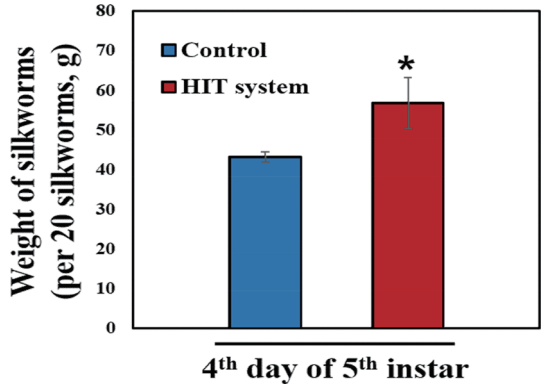

Fig. 5. Comparison of the final weights of silkworms harvested from previous heating system or HIT system. In this measurement, 4th day of 5th instar silkworms were finally harvested. Each of the 20 silkworms was harvested three times, and the weight was measured and averaged. Error bar represents standard deviation. The silkworms harvested from HIT system showed a 31.6 percent increase in weight compared to those from previous heating system. The asterisks represent significant differences with control system ($P<0.05$).

Comparison of weight of the final harvested 5L4D silkworms of two heating systems

This experiment ended with harvesting 5L4D silkworms. The silkworms were raised from 1st day of 4th instar, and the heating methods was treated in different systems for seven days until the 4th day of the 5th instar. As shown in figure 5, the average weight of the 5L4D silkworms in the previous heating method was 43.12 g per 20 silkworms. However, the average weight in the HIT system was finally 56.76 g per 20 silkworms. This was an increase of 31.6% from the results of silkworm breeding in the previous heating method. These results show that a new heating method called HIT system is able to raise silkworms not only robustly and uniformly, but also consequently have a strong positive effect in weight increase on silkworm breeding.

In this study, silkworms were raised by applying the newly developed heater installed tray system called as the HIT system improve the previous heating system for silkworm feeding. In order to conform the difference of the HIT system, the growth period of silkworms, the weight of specific stage, the growth uniformity, and the weight of the final harvested silkworms were measured. Compared to the previous system, the HIT system showed a shortened silkworm growth period, an increase in weight by stage, in growth uniformity, and in weight of the final harvested 5L3D silkworms. In particular, the 5L3D silkworm in

the HIT system showed a 31.6 percent weight increase from the previous system, which resulted in a decisive benefit on farm income. In addition, the HIT system had the advantage of being able to use space to every edge of the facility, being applied to simple facilities that have difficulty heating the whole, and being easy to clean up facilities due to fast drying of residues after feeding. Due to the continued warmth in the HIT system, the drying of the rearing seat was done easily. However, there was no difference between the two systems in the humidity. The humidity remained high while eating mulberry leaves due to enough mulberry leaves in the rearing seat. In the case of 1st to 3rd instar, the mulberry leaves are cut into small pieces, so there is a possibility that the mulberry leaves dry much faster and become a problem. Therefore, this experiment was supplied in the form of mulberry shoots without cutting mulberry leaves, and conducted by using from 4th instar silkworms. However, in case of 1st to 3rd instar silkworms that need to supply chopped mulberry leaves, it is not easy to apply the HIT system because drying of mulberry leaves is likely to occur rapidly. Methods to improve this will need to be studied next. The new HIT system costs additional expenses to install heater tubes under the rearing tray and new oil boilers. However, it was calculated as earning more than the cost (data not shown). In conclusion, the newly introduced HIT system was proven to be a useful method of greatly improving the previous heating system for silkworm rearing and eventually maximizing the income of silkworm rearing farmers.

Acknowledgements

This work was supported by a grant from Agriculture Science & Technology Development (PJ010018022022) from Rural Development Administration of the Republic of Korea.

References

Asaoka K (2000) Deficiency of gustatory sensitivity to some deterrent compounds in “polyphagous” mutant strains of the silkworm, *Bombyx mori*. J Comp Physiol A 186, 1011-1018.

Herold HM, Scheibel T (2017) Applicability of biotechnologically produced insect silks. Z Naturforsch C J Biosci 72, 365-385.

Iizuka T, Tamura T, Sezutsu H, Mase K, Okada E, Asaoka K (2012) Genetic analysis of the electrophysiological response to salicin, a bitter substance, in a polyphagous strain of the silkworm *Bombyx mori*. PLoS One 7, e37549.

Jo YY, Kim S. Lee JH, Kweon HY, Ju W, Kim H, *et al.* (2020) Characteristics of the 3rd day of 5th instar silkworm powder: effect of preparation method. Int J Indust Enomol 40, 22-27.

Kim SH, Jo YY, Kweon HY, Lee JH (2021) Effect of extraction conditions on radical scavenging and cholesterol metabolism regulating capacity of silkworm larvae. Int J Indust Entomol 43, 45-51.

Kim TK, Yong HI, Kim YB, Kim HW, Choi YS (2019) Edible insects as a protein source: a review of public perception, processing technology, and research trends. Food Sci Anim Resour 39, 521-540.

Kumari SS, Subbarao SV, Misra S, Murty US (2011) Screening strains of the mulberry silkworm, *Bombyx mori*, for thermotolerance. J Insect Sci 11, 1-14.

Kurin R (2002) The Silk Road: Connecting Cultures, Creating Trust. Talk Story 21, 1-11.

Liang W (1985) Low temperature damage and defense strategies in oak silkworm production. Can Ye Ke Xue 11, 61-63.

Okano F, Satoh M, Ido T, Okamoto N, Yamada K (2000) Production of canine IFN-gamma in silkworm by recombinant baculovirus and characterization of the product. J Interferon Cytokine Res 20, 1015-1022.

Osanai M (1978) Longevity and body weight loss of silkworm moth, *Bombyx mori*, varied by different temperature treatments. Exp Gerontol 13, 375-388.

Rahmathulla VK, Suresh HM (2012) Seasonal variation in food consumption, assimilation, and conversion efficiency of Indian bivoltine hybrid silkworm, *Bombyx mori*. J Insect Sci 12, 82.

Tabunoki H, Bono H, Ito K, Yokoyama T (2016) Can the silkworm (*Bombyx mori*) be used as a human disease model? Drug Discov Ther 10, 3-8.

Ueda Y, Sakurai T, Yanai A (1993) Homogeneous production of feline interferon in silkworm by replacing single amino acid code in signal peptide region in recombinant baculovirus and characterization of the product. J Vet Med Sci 55, 251-258.

Vijayan K, Nair CV, Urs SR (2010) Assessment of genetic diversity in the tropical mulberry silkworm (*Bombyx mori* L.) with mtDNA-SSCP and SSR markers. Emirates J Food Agric 22, 71-83.

Wang X, Zhang Y, Kong H, Cheng J, Zhang M, Sun Z (2020) Novel mulberry silkworm cocoon-derived carbon dots and their anti-inflammatory properties. Artif Cells Nanomed Biotechnol 48, 68-76.

Zhang HJ, Faucher CP, Anderson A, Berna AZ, Trowell S, Chen QM, *et al.* (2013) Comparisons of contact chemoreception and food

acceptance by larvae of polyphagous *Helicoverpa armigera* and oligophagous *Bombyx mori*. J Chem Ecol 39, 1070-1080.

Zhang L, Zhang L, Li Y, Guo X, Liu X (2016) Biotransformation effect of *Bombyx Mori* L. may play an important role in treating diabetic nephropathy. J Integr Med 22, 872-879.

Zhou HM, Wang ZB (1982) Effect of egg duration on spring oak silkworm. Can Ye Ke Xue 8, 229-231.