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Environmental factors and rearing techniques affecting the rearing of silkworm and cocoon production of *Bombyx mori* Linn

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Abstract
Silkworms have been domesticated or semi-domesticated insects for many centuries, as silkworms play an important role in the economy of men. The silkworm is by nature very delicate and sensitive to environmental conditions. Silkworm rearing is therefore aimed at producing very good quality cocoons of very high silk content utilizing the available optimum environmental conditions. The ecological factors, chiefly temperature, humidity, light and air during rearing show a significant effect in the growth of larvae and finally cocoon crop quality. However, other factors such as quality and quantity of the leaf supply and techniques of rearing adopted, such as feeding, cleaning, spacing etc. also influence the rearing activity. The silkworm behavior in relation to various environmental conditions varies with different stages of growth, and a bad environment affects the growth of worms.

Keywords: rearing techniques affecting, silkworm, cocoon *Bombyx mori* Linn

Introduction
Silkworms are poikilotherms. Thus, they change their body temperature according to the environmental temperature. Temperature has a direct effect on the various physiological activities of living organisms like other animals. The physiology of silkworm namely metabolic rate, activity of enzymes, nutrient conversion, digestion, assimilations, excretion, nervous stimulation, hormonal actions are influenced by environment temperature. The silkworm is capable of growing in temperatures ranging from 15 degree Celsius to 40 degrees Celsius but from the physiological point of view, ideal temperature ranges from 20 degree Celsius to 30 degrees Celsius. The silkworm rearing is aimed to produce good quality cocoons for which an ideal and desirable temperature ranges from 23-28 degree Celsius for getting maximum productivity. The effect of temperature on the physiology of silkworm during rearing can be divided into groups which are as follows: a) Temperature which is harmless to growth of silkworm b) Temperature which is favorable for the healthy growth of silkworms. c) Temperature favorable for making good quality cocoons. With the rise in temperature, metabolic activities of the worms are increased while they are slackened when temperature is low. Therefore, at high temperature the growth of larva is fast and consequently the duration of the larval period is shortened. On the other hand, growth becomes slow at low temperature with prolonged larval duration. The chowki worms (Young aged larvae) are comparatively stronger and more resistant to humidity and temperature than the late aged worms. Thus, the young ones feed very actively and grow vigorously under high temperatures in humid conditions. As mentioned above the temperature has a direct correlation with the growth of silkworm, wide fluctuations are avoided as they are harmful. The role of rearing humidity upon the growth and health of silkworm is similar to that of rearing temperature. Humidity helps to decrease the length of growing period of silkworm by accelerating the activity of physiological function. The pH value of blood is remarkably lower at high humidity 80-90% than at low humidity 60% condition. Expiration of carbon dioxide increases with rise in humidity. On the contrary, low humidity prolongs the length of the growing period of larva. The combined effect of both temperature and humidity largely determines the satisfactory growth of the silkworms and production of quality cocoons. In this way both temperature and humidity are complementary to each other. The role of humidity affects both directly and indirectly. Directly involves to influence the physiology of silkworm larva when indirectly influences the rate of withering of leaves in larval beds.

Therefore, suitable rearing must be determined while considering the following points. 1. The optimum humidity for the growth of silkworm is about 75% of relative humidity. The early instars are resistant to high humidity with relatively little or no effect of change in the moisture. On the contrary, the late age instars are weak against high humidity suffering badly from the damage of humidity. 2. In an extremely desiccated rearing room silkworms cannot eat Mulberry leaves owing to the withering of leaves. Then they become malnourished especially this problem occurs in early instars. 3. If the air of the rearing room is too moist, it becomes favorable to grow the pathogens and microbes thus silkworms are apt to suffer from disease considering above factors, different humidity conditions are there for different instars.

Silkworms breathe through 18 spiracles on both sides of the body supplying the blood with oxygen through the trachea. This trachea distributes the oxygen to the body and at the same time unnecessary substances like carbon dioxide or water produced in the body are expelled through trachea. Therefore, it is needless to say that fresh air is necessary for the life of a silkworm. The air of the rearing room is contaminated by carbon dioxide, Sulfuric acid gas, carbon monoxide, ammonia, formaldehyde etc. produced by the breath of man, silkworm or Mulberry leaves fermented by litter, burning of coal. These gases are injurious to silkworms. When the gases increase in the beds and rearing room beyond the tolerance limits of worms become sluggish and do not feed. The safe limit for silkworm rearing is 1-2% of co2, 1% of formaldehyde, 0.02% of SO2 & 0.1% of ammonia in the era of rearing room. Young worms are less resistance to toxic gases though comparatively resistance to carbon dioxide. The productions of these gases are far less in early instars than in the late instars. The effect of SO2 causes prothetely in cocoon & makes cocoon quality worse and worsening the reliability of cocoon filaments. Ammonia (0.1%) in the room makes sericin insoluble while reeling. There is no correlation between light and survival of silkworm, however the silkworms are photosensitive and generally tend to crawl towards dim light. They do not like either strong light or complete darkness to produce heavier cocoons than those of silkworms reared in dark condition. The appetite of silkworms is more in a light place than in a dark place. The larva comes up to the upper surface of the bed more quickly under light conditions than in a dark condition. However, it is not advisable to rear in complete light or complete darkness. Silkworms fond of dim light of 15 to 20 lux avoid strong light and darkness. The late age worms thrive better in 16 hours light and 8 hours dark periods. Light helps in uniform distribution of Larvae in rearing beds. They are crowded and distributed in several layers in dark conditions compared to thin and even distribution in light conditions. Photoperiod influences on the early instars on the type of egg produced by the resulting moth.

Materials and Methods

Silkworm rearing involves the following steps

1. Disinfection and sterilization of rearing station.
2. Incubation and Brushing
3. Rearing of silkworms
4. Early age rearing
5. Late age rearing.

Result and Discussion

1. Disinfection and sterilization of rearing station

Chemical Disinfectants available for use in Sericulture

1) **Formalin:** It is commercially available as 36% formaldehyde in solution form. A mixture of 2% formalin + 0.05% detergent is an effective solution that can be used for disinfection purpose as spray. Formalin is effective only in rearing houses, which can be made airtight and it is faster and more pronounced at temperature above 25 ^{0}C and humidity more than 70%.
2) **Bleaching powder:** It is white amorphous powder, with a pungent smell of chlorine. For effective disinfection, a high-grade bleaching powder with an active chlorine content of 30% must be used. It should be stored in sealed bags, away from moisture, failing which it will be rendered ineffective. The action of bleaching powder is optimal under wet and contact conditions and therefore the surfaces of equipment and walls should be drenched with this solution. A 2% bleaching powder in 0.3% slaked lime solution is used for disinfection as spray.
3) **Resham Keet Oushadh (RKO):** It is a bed disinfectant which can be applied on the silkworm rearing bed to inactivate pathogenic microbes responsible for muscardine, Grasserie and nuclear polyhedrosis diseases in silkworm. RKO is economical and its usage increases the cocoon yield on an average of 7.00 kg per 100 diseases free laying (dfls). It is easy to use and has no adverse effect on silkworm health, human beings and domestic animals. The quantity of RKO required for treating 100 dfls is 3.25 kg. RKO is produced from locally available chemicals and the shelf life of RKO is six months from the date of manufacture.
4) **Slaked lime:** A very useful bed disinfectant in sericulture. Especially against viruses. It absorbs moisture and can be used to regulate bed humidity and maintain hygiene. Application of lime dust in combination with bleaching powder in and around rearing houses and premises improves hygiene in the environment.
5) **Chlorine dioxide:** Chlorine dioxide marketed as Sanitec is an ideal disinfectant available at 20,000 ppm concentration is a strong oxidizing agent, effective at broader pH range and at 2.5% concentration in combination with 0.5% slaked lime is effective against all silkworm pathogens. It is stable and may be activated at the time of its use. it possesses tolerable odor and is least corrosive at the suggested concentration.

2. Incubation and Brushing

Early age rearing/ Chawki rearing

The life cycle of silkworm consists of egg, larva, pupa (cocoon) and adult stages. Among these four stages, larval stage is the only feeding and active stage. The duration of larval period form hatching to spinning is about 26 days. During this long duration the larvae grow in size and enter cocoon (pupal) stage. To accommodate the larval body growth the larvae, undergo four moults and thereby the complete larval duration can be clearly differentiated into five instars or stadia. The first three instars (till the third moults) are known as young age or chawki and the last two instars are called as late age worms.

Importance of Chawki rearing: young age or chawki rearing and late age rearing techniques are different. Both nutritional and ecological requirements during these two stages are different. The essential point in rearing of young silkworms is to get strong and sturdy silkworms. The success of sericulture depends to a large extent on the successful rearing of young worms. Young age worms are more resistance to high temperature and humidity and grow healthier, ensuring success of cocoon crop. The first character of young silkworms is that, they grow extremely fast. To support their fast growth, they must be given highly nutritious sufficient mulberry leaves. Undergrown mulberry makes silkworms susceptible to disease growth becomes uneven, reflects on rearing resulting in poor cocoon crop. Therefore, young silkworms are fed with tender and succulent mulberry leaves. The leaf eating time is shorter in earlier stages then later instars. However, the total leaf requirement of silkworm larva in only 6.33 percent, during chawki rearing up to III moults. But body weight increases by 400 times, while 300 times increase in body size and 500 times increases in silk gland weight are achieved during young stage provided the conditions and methods of rearing are ideal. Further the rate of increase in body weight of larva per given time is more in the first instar and it decreases with the age. The effects if insecticides disinfectants, injurious gases etc, on the larvae are more in the earlier instars than in the later instars. Keeping in view of above points the young worm rearing must be carried out with maximum care.

Environmental Conditions: Since silkworms have been domesticated for many centuries, they are by nature quite delicate and are very sensitive to environmental conditions. The ecological factors chiefly temperature, humidity, light and air during rearing have a significant in influence on the growth of larva and ultimately on cocoon crop quality. Of course, the other factors like quality and quantity of leaf supply and techniques of rearing adopted such as feeding, cleaning, spacing etc, are also to be considered. The influence of environmental conditions is not the same throughout the rearing period, but varies in different stages of larval growth depending upon the physiological condition and voltinism of the silkworm.

Temperature: Temperature plays a vital role on the growth of the silkworms. As silkworms are cold blooded animals, temperature will have a direct effect on various physiological activities. Rise in temperature increases various functions and with a fall the activities are decreases. Increased temperature accelerates larval growth and shortens the larval period. On the other hand, at low temperature the growth is slow and larval period is prolonged. The optimum temperature for normal growth of silkworms is between 20 °C and 28 °C and the desirable temperature for maximum productivity ranges from 23 °C to 28 °C. Temperature above 30 °C directly affects the health of the worm. If the temperature is below 20 °C all the physiological activities are retarded, especially in early instars, as a result worms become too weak and susceptible to diseases. The temperature requirements during the early instars (I, II, III) is high and the worms feed actively and grow very vigorously. Such vigorous worms, can stand better even at adverse conditions in later instars. Optimum rearing temperature for rearing is 24 – 28 °C. In general, the early instar larvae are resistant to high temperature and it also helps in improving survival rate and cocoon characters. The temperature has a direct correlation with the growth of silkworms and wide fluctuation of temperature is harmful and as for as possible it should be avoided. The optimum temperature required for rearing silkworms of different early instars are as follows:

Table 1: The optimum temperature required for rearing silkworms of different early instars are as follows:

S. N	Stage of silkworm Larvae	Optimum temperature of rearing (°C)
1	I st instar larvae	28 ± 2 °C
2	IInd instar larvae	27 ± 2 °C
3	IIIrd instar larvae	26 ± 2 °C
4	IVth instar larvae	25 ± 2 °C
5	Vth instar larvae	22 ± 2 °C

Regulation of Temperature: Generally, the room temperature is low during winter and rainy day which should be regulated by heating with electric heaters or charcoal fires. Electrical heaters are best since they do not emit any injurious gases. When electricity is costly and not available, properly dried charcoal and can be used. In this case however the live cinders should be covered with a layer of ash for more regulated room to raise the temperature should be avoided.

Because the carbon dioxide and other gases emitted in this process are injurious to silkworms. Besides the above processes the doors and windows should be kept closed during nights to keep out the cold. Late in the day, as the outside temperature goes up doors and windows should be opened to allow warm air in to the room. In Rainy states except for a few days of winter and rainy days, the temperature is often above the optimum level. Thus, it is problem for the readers to lower the temperature rather than heating up of rearing room. This sort of temperature is averse to silkworms. This adverse effects to a certain extent be mitigated through proper designing of the rearing house and by ensuring adequate ventilation and free circulation of air. During summer season when the day temperature is high, all the windows should be kept open during night, to bring down the temperature. And early in the morning all the windows and doors should be opened so that the cool air from outside is allowed into the rearing room to bring down the temperature. When the sun rises and the temperature goes up, doors and windows should be closed. Besides these windows and doors are covered by wet gunny cloth on a hot day to reduce the temperature. Otherwise, air coolers can also be used for this purpose.

Humidity: It plays a vital role in silkworm rearing. The combined effect of both temperature and humidity largely determines the satisfactory growth of the silkworms and production of good quality cocoons. Its role is both direct and indirect. It directly influences the physiological functions of the silkworm. The young age silkworms can withstand to high humidity conditions better than later age worms and under such condition the growth is vigorous. The humidity conditions for different early age worms are as follows:

Table 2: The humidity conditions for different early age worms are as follows:

S. N	Stage of silkworm Larvae	Optimum Relative humidity of rearing (%)
1	I st instar larvae	85 ± 5%
2	IInd instar larvae	80± 5%
3	IIIrd instar larvae	75 ± 5%
4	IVth instar larvae	85 ± 5%
5	Vth instar larvae	80 ± 5%

Humidity indirectly influences the rate of withering of the leaves in the silkworms' beds. Under dry conditions the leaves wither very fast and become unsuitable for feeding. This effects growth of the larvae and also results in wastage of leaf fed. Retarded growth of young larvae makes them weak and susceptible to disease. At a humidity of 90 percent or higher, if temperature is kept at 26 – 28 °C, they can grow without being greatly affected. Therefore, the humidity is kept high to prevent mulberry leaf withering.

Regulation of humidity: Like temperature, humidity also fluctuates widely not only from season to season but also within the day itself during any season. Therefore, it is a must for the readers to regulate it. For this purpose, paraffin paper is used for rearing beds during chawki rearing to raise humidity. Otherwise, wet foam rubber pads or paper pads soaked in water can also be used to increase humidity in the beds. However, it is important to lower humidity to 70 per cent or below during the moulting in each instar to facilities uniform and good moulting. Otherwise, it results in a. silkworms remain under the net b. uneven growth c. become susceptible to disease d. bed cleaning requires much lab our e. missing worm number increases Therefore readers must remember the drying and disinfection of bed during moulting without fail. Removal of paraffin paper during moulting raises the drying effect.

Air: Like other animals' silkworms also require fresh air. By respiration of silkworm's carbon dioxide gas is released in the rearing bed. Besides this carbon monoxide, ammonia, Sulphur dioxide etc., are also released in the rearing room by burning of charcoal to raise temperature. These gases are injurious to silkworms. Therefore, care should be taken to allow fresh air through proper ventilation to keep the toxic gases at a low level. If CO2 exceeds to 2 per cent concentration, the growth of silkworm is retarded. Insecticides and disinfectants are also avoided in the rearing room. Air plays an important role in regulating room temperature and humidity. Artificial air circulation is useful for bringing down high temperature and humidity.

Light: Silkworms are photosensitive. They have a tendency to crawl towards dim light. They do not like either strong light or complete darkness. The larval moults is uniform when silkworms are reared in 16 hours light and 8 hours darkness**.**

Feeding of Larvae: The Mulberry leaf is the exclusive food of the silkworms (Bombyx mori). The growth of the silkworm very much depends on the quality of leaves fed to them. The leaf quality is influenced by various factors such as soil, pruning, fertilizer, rainfall, irrigation etc. with these conditions' mulberry grows luxuriously with rich contents of proteins and carbohydrates. Further the leaves are also succulent due to high nutrient content. This type of leaves is edible for silkworms for better growth and to produce good cocoons. Leaves of mulberry grown on loamy soil contain more water, protein and less carbohydrate and fiber. Further the leaves mature slowly. Mulberry leaves form trees grown in sandy or gravel soil mature quickly becoming rough and coarse. These leaves contain less moisture, protein and more carbohydrates and fiber. Application of balanced fertilizers with major elements required by the plants improves both physical and chemical properties of the leaves. In well distributed rainfall or irrigated conditions the mulberry growth is vigorous. Leaves of these plants are rich in nutritive value and are soft and succulent. In areas where temperature fluctuations during night and day are high the leaf quality becomes superior. The nutrients synthesized during the day are least utilized during the cool night hours. Therefore, the nutrients are better preserved in the leaves. Mulberry raised under ideal agronomic conditions are better for rearing silkworms. The conditions are as follows.

1. Good soil, neither two clayey nor too sandy, but not acidic.
2. Application of optimum and balanced fertilizers
3. Suitable cultural operations
4. Assured irrigations or rainfall.

On the above said conditions the leaves are rich in protein, and carbohydrates besides high leaf moisture.

Preparation of leaves for feeding young worms: Depending on the size of the worms' complete leaves can't be used for chawki worms. Further leaf quality can also be influenced by the process of chopping. However, the cut surfaces of leaf lead to loss of moisture. Therefore, it is essential to adjust the chopping of leaf so as to protect the quality of leaf. The withering of leaf in rearing bed can be prevented using paraffin paper and foam rubber or paper soaked in water. This induces to raise humidity the main advantage of chopped leaf is to facilitate even distribution of feed to the worms. In cold conditions chopped leaves prevent the silkworm bed from dampness. Leaves do not curl up when the air is not and dry. However, a greater number of leaves are wasted besides lab our expenses.

Spacing: The silkworms grow very rapidly from age to age and increase many times their weight and size from the previous instar. The total increase in weight from hatching to the end of Vth instar is about 7,000 to 10,000 times. Overcrowding of silkworm larvae means insufficient space for movement and free feeding of the worms and so the larvae crawl over one another. Crowded situation in rearing trays results in an increase in the accumulation of gases, increased humidity, heat, and fermentation of litter /fecal matter particularly during the early stage when temperature and humidity in rearing room are high. This in turn causes under development/unhealthy growth of larvae, wastage of feeding leaf and unhygienic condition. If there is space beyond the optimum required, there is a considerable wastage of mulberry leaves and also the lab our costs for feeding increases. The rearing space would vary in different stages of the silkworm larvae as is evident from the table. To provide more and adequate space for the growing worm, the rearing space has to be extended at each stage and this is called spacing. Spacing is usually done along with bed cleaning and is given once a day.

Table 3: Trays for spacing the silkworm

Leaf requirement	(In kg) for 50 Dfls Or about	2000 eggs for different races
Larval instar	Uni/Bivoltine	Multivoltine
I st instar larvae	1-2	1-2
IInd instar larvae	5-6	2-3
IIIrd instar larvae	20-55	15-20
IVth instar larvae	80-90	35-55
Vth instar larvae	450-475	301-350

Bed cleaning: Silkworms are fed with large quantity of mulberry leaves than their eating capacity. Thus, unconsumed leaves which are unfit for food remain in the tray at the end of each feed. Besides these excreta of worms forms a thick bed. Out of the total weight of leaf taken as food, three fifth is excreted and only two-fifth is being assimilated by the silkworm. The pilling Leaf size (cm2) instar peak eating stage preparation for moults I, I, III 2.0 4.0 full leaf cut in to four pieces 1.0, 1.5, 2.0 to start with 0.5, 2.0, 4.0, Silkworm rearing technology: of litter makes the beds moist. This releases process of fermentation liberating injurious gases and also favors multiplication of pathogens. All these above factors are harmful to the worms. Therefore, removal of old (unused) mulberry leaves, fecal matter of silkworms, exuviae, dead or unhealthy worms etc., from the rearing bed is called bed cleaning. Frequency of Cleaning involves lab our and frequent cleaning is not advisable as it cuts the economics of reading. While cleaning loss of worms is inevitable especially in chawki rearing. The frequency of cleaning for young worms are as follows. I instar — once II instar — twice i.e., once just after the I moults and again before setting for II Moults III instar — thrice i.e., once after moults, once in the middle of III age and once just before setting for IV moults.

Moulting: The silkworm larval life has five instars and four moults. The larvae cast off its skin to accommodate the body growth. This is called moulting. The silkworm larvae attain their maxi-mum body growth of particular instar and as a result body becomes stout, and shiny and amber colored. These two characters are seen in a larva at the approach of moulting. In relation to the size of the body, the head of the worm appears small and dark. This is the time for bed cleaning and wide spacing. After them worms are about to settle for moults are given one or two feeds which helps to reduce the humidity and favors uniform moulting. In high humid conditions a thin layer of time powder is dusted. This prevents early molted larva from eating, favoring uniform growth. Feeding is stopped when all larvae settle for moults. Under proper rearing conditions all the larvae settle uniformly for moults and come out of moults uniformly. The moulting time for first age is 20 hrs. Second and third age larvae requires one day. Moulting is a very sensitive process in the life cycle of silkworms. After moulting fresh larvae of next instar comes our casting their old skin. The worm's head is bigger in relation to the body size. It is rusty in color, less shiny because of loose skin. The first feeding of the new instar starts only after almost all worms passes moults. Newly molted worms are dusted with ceresin lime prior to first feed to prevent muscardine. Any irregularity in settling for moults is noticed, all such late larvae are segregated by net feeding and reared a second batch. Care should be taken to keep the bed dry during moults. These facilities the larvae to wriggle out of the old skin.

Rearing methods: There are three methods or rearing but in all methods importance is given to, the maintenance of leaf quality, humidity, temperature so as to ensure vigorous and healthy development. The rearing methods are; 1. Paraffin Paper Rearing a good quality paraffin paper is used in this method. It should be devoid of petroleum smell, folds, and turns. It is spread as a bottom layer and as a cover for rearing beds. In between the sheets on all four sides of rearing bed, strips of wet foam rubber or newspaper are placed to maintain the required humidity. Light weights are kept on the top paraffin sheet to seal the edges for better maintenance of rearing bed humidity. While feeding the worms, the top paraffin paper sheet must be removed 30 minutes prior to feeding. This allows supply of fresh air to the silkworms and eliminates toxic gases accumulated in the bed. When the worms settle for moults, paraffin paper is not necessary. Further the bed must be dry during moults. A thin layer of lime powder is sprinkled over the bed which helps to keep the bed dry.

Box Rearing: In this method specially made boxes are used for rearing. The boxes may be with or without lids.

a) **Rearing in boxes with lids:** It completely resembles the paraffin paper method. After preparation of bed a lid is placed on the box and later arranged in the shelves. In third instar lids are not necessary. When the larvae settle for moults, the paraffin paper, wet foams and the lids are removed to keep the bed dry.

b) Rearing in Boxes without lids: This rearing again resembles paraffin method. The wooden boxes of uniform size with 10-15 cm deep are used. After preparing the rearing bed the boxes are piled one over the other for rearing first instar. For rearing second and third instar larvae, a space of 2-3 cm between the boxes is made for ventilation. The boxes are kept open for at least 30 minutes prior to each feeding. It must be completely open when larvae start settling for moults. Care must be taken to disinfect the worms to prevent muscardine.

c) Co-operative Rearing: Co-operative rearing of silkworms requires technical skills. These skills are lacking in most of rural farmers. If the silkworms are not reared properly in the young stages, they are prone to diseases in later instars, resulting in crop failures. Besides this rearers are not able to afford the necessary equipment for silkworm rearing under ideal conditions. In order to overcome all these problems co-operative rearing have been organized to provide technical assistance, idea conditions etc. the rearing is conducted up to second or third moults. These are also called as chawki rearing centers. These centers are provided with ideal rearing houses with all the necessary equipment. The total rearing is supervised by technical experts. Mulberry leaf for rearing is provided form a single garden which ensures uniform quality of leaf.

Conclusion

Especially rearing technology and environmental factors affects the cocoon morphology as well as its stiffness and strength, which we attribute to altered spinning behavior and sericin curing time. Environmental & rearing technology affect cocoon coloration, perhaps due to tanning agents. The leaves are soft and succulent. This type of leaves is easily digested and best utilized. From the quality point of view the requirements of young worms are completely different from those of late age worms. The chawki worms require tender, soft and succulent leaf having higher contents of moisture, protein, sugars and less starch and fiber. However too soft leaves are not suitable for the worms. Our findings demonstrate environmentally & rearing technology induced quality parameters that must not be ignored when analyzing and deploying silk cocoon, silkworm growth, silk filaments or silk derived bio-polymers.

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