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Recent Advances of Nutritional Physiology and Artificial Diet of the Silkworm, *Bombyx mori*, in Japan

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Abstract

Recently, the cocoon production is reducing very steeply in Japan. The main reason for this cocoon reduction is due to high labor cost and the production expense is apt to rise year by year. Accordingly, the improvement of cocoon productivity by retrenchment of the production expense is intensively required in my country. Therefore, the silkworm rearing on the artificial diet is largely expected as the innovation techniques of sericulture. Since rearing of the silkworm on artificial diets has first been achieved in 1960, and the silkworm larvae were reared throughout all the instars on artificial diets(Fukuda *et al.*, 1960, Ito & Tanaka, 1960). The artificial diets used at this time contained 50% or more of dried, pulverized mulberry leaves as one of the main ingredients, and the composition was rather simple. When reared on these diets, larval growth and development were retarded, small cocoons appeared, and the adults laid only a small number of eggs. At first, we would like to study about nutritional requirement of the silkworm, using the chemically defined diet which prepared artificially. Subsequently, attempts were made to improve the diets by replacement of crude ingredients with possibly purer compounds, as mentioned later. Before giving the details on individual nutrient, I would like to summarize about the outline of utilization flow of various nutrients through the larval-pupal development and these daily quantitative requirements per body weight in the silkworm when they were reared on mulberry leaves.

UTILIZATION OF VARIOUS NUTRIENTS

The amounts of dry matter and energy utilized by a single larva reared on mulberry leaves through the 4th and 5th instars have been measured. About 54 and 59% of the digested dry matter were stored in the mature larval body of male and female, while 47 and 42% were consumed during 4th and 5th instars by male and female, respectively.

Approximate one fourth of digested dry matter was distributed into cocooon layer and 11% into eggs.

The ratio of the metabolizable energy to the ingested one was approximately 45% in both sexes. The gross energy stored in the mature larva was found to be 64 and 66% of the gross metabolizable energy. Approximate 26 and 34% of the metabolizable energy were stored in the pupal bodies of male and female, and over 12% was utilized for egg for mation. The gross energy consumed in 4th and 5th instars was 37 and 35% of the metabolizable energy for male and female larvae, respectively.

The ratio of metabolizable carbon was approximately 40% in both sexes. Of the metabolizable carbon, 73 and 77% were stored in male and female mature larvae. The higher recoveries of both energy and carbon in the mature larvae than that of dry matter seems to suggests the accumulation of high-energy and high-carbon substances such as lipids in the mature larvae. Approximate 27 and 24% of metabolizable carbon were consumed during 4th and 5th instars. These consumptions of carbon were in fair agreement with the cumulative amount of respiratory CO_2^- output during the same period. The recovery of metabolizable carbon to cocoon layer was 28% in both sexes.

The nitrogen utilization is much more effective than those of others, in particular, the extreme high recovery of nitrogen to cocoon layer is remarkable characteristic in the silkworm nutrition. The nitrogen amounts ingested by male and female larvae in 4th and 5th instars were 12 and 13 mMoles, and apparent digestibility was 62～64% of total nitrogen and 70% of amino nitrogen in both sexes, respectively. It is noteworthy that about 70 and 66% of absorbed nitrogen were recovered as the silk proteins in male and female, respectively.

The daily ingestions of various kinds of nutrients per body weight of the larva are shown in Table 1. The digestibility of protein and amino acids was relatively high and it was extremely high in N_3-free extracts fraction including oligosaccharide such as sucrose which was known to be the main sugar in mulberry leaves. Whereas, cellulose was unable to be digested by the silkworm at all. The rate of absorption of vitamins was varied according to the kinds of vitamins. These daily requirements of nutrients by the silkworm are comparable with those in other species of animal and these values are seem to be useful from the viewpoint of animal comparative physiology. In general the daily ingestion of dry matter per body weight was much larger in the silkworm than those in other animals.

Table 1. *Daily amounts of nutrients ingested and utilized (Amount/g body wt./day)*

Nutrients	Ingested (a)	Utilized (b)	Ratio (b/a×100)
Dry matter(g)	0.2235	0.101	43.0%
Energy(cal)	1.005	0.466	46.4
Crude protein(mg)	56.9	38.1	67.0
Crude lipids(mg)	20.2	12.2	60.4
Crude cellulose(mg)	19.2	0.0	0.0
N_2-ree extracts(mg)	29.6	28.2	95.3
Crude minerals(mg)	24.0	2.6	10.8
Total amino acid(mg)	61.6	48.0	77.9
Arginine	3.8	3.0	78.9
Histidine	1.8	1.1	61.1
Isoleucine	2.7	2.1	77.8
Leucine	5.1	4.0	78.4
Lysine	4.3	3.5	81.4
Methionine	0.7	0.6	85.7
Phenylalanine	3.1	2.2	71.0
Threonine	3.8	3.0	78.9
Tryptophan	–	–	–
Valine	2.4	1.6	66.7
Thiamine(μg)	0.8	0.5	63.0
Riboflavin(μg)	5.0	2.9	58.0
Pantothenate(μg)	4.6	0.86	19.0
Nicotinate(μg)	18.0	3.1	17.0
Pyridoxine(μg)	3.3	1.1	33.0
Biotin(μg)	0.18	0.01	6.0
Folic acid(μg)	0.05	–	–
α-Tocopherol(μg)	59.0	27.0	46.0
Choline(mg)	0.38	0.23	61.0
Inositol(mg)	2.1	1.9	91.0

QUANTITATIVE AND QUALITATIVE NUTRITIONAL REQUIREMENTS

As is generally known, the completion of the chemically defined diet is a prerequisite for the analysis of nutritional requirements of the silkworm. Namely, the defatted soybean powder added as protein source was replaced with more pure protein such as milk casein. Milk casein was further replaced with amino acid mixture. Furthermore, mulberry leaves powder which added to the diet for acceleration of feeding, was replaced with one of feeding stimulant, morin. The typical composition of the chemically defined diets in shown in Table 2.

By using the chemically defined diet, much information has been accumulating on nutritional requirements for growth and survival of the silkworm. For the determination of the effect of nutrient on growth and survival, the single omission method used to be available. As a result, we have determined kinds of the essential nutrients and their necessary amounts. In Table 3 the kinds and the minimal optimal levels of nutrients essential are summarized. The levels of nutrients are calculated in a logarithmic unit of μ moles per g of dry diet and this unit is proportional to the number of molecule of various nutrients.

Ten amino acids, nine vitamins, sterol and at least four minerals were found to be essential for good growth and development of the silkworm. It is noticed that the order of dose requirement of nutrients is closely correlated with the content of respective nutrients in mulberry leaves, and the order of content is specific according to the kinds

Table 2. *Composition of the synthetic diet*

Substances (% of dry diet)		Substances(μg/g of dry diet)		Substances(% of dry dirt)	
Starch, potatto	7.50	(1) Vitamin B mixture		(2) Amino acid mixture	
Sucrose	12.50	Choline-Cl	2,000	Arginine-HCl	1.2
Wesson's salt mix.	4.00	myo-Inositol	2,000	Histidine-HCl	0.6
Ascorbic acid	2.00	Pantothenate-Ca	150	Isoleucine	1.0
Citric acid	0.50	Nicotinic acid	100	Leucine	1.6
Agar	15.00	Pyridoxine-HCl	30	Lysine-HCl	1.4
Morin	0.30	Riboflavin	20	Methionine	0.6
β-Sitosterol	0.50	Thiamine-HCl	20	Methionine	0.6
Soybean oil, refined	3.00	Biotin	2	Phenylalanine	1.2
Cellulose posder	33.27	Folic acid	2	Theronine	1.0
Vitamin B mixture(1)	0.43	(3) Antiseptics consisted of		Tryptophan	0.6
Amino acid mixture(2)	21.00	.015% of chloramphenicol		Valine	1.2
Antiseptics(3)	Added	and 0.75% of propionic acid.		Proline	0.8
Distilled water	3 ml/g			Aspartate-K	2.6
				Glutamate-K	2.4
				Alanine	1.4
				Cystine	0.4
				Glycine	1.0
				Serine	1.4
				Tyrosine	0.6

of nutrient. Thus, these dose specificity among various kinds of nutrient seems to suggest their specificity of metabolic function in the silkworm.

Namely, amino acid group functions as source of structural proteins of body, whereas vitamins functions as the precursor of coenzymes and they play important catalytic roles in metabolism. Furthermore, the lipogenic substances such as choline, inositol and sterol seem to be utilized for the precursors of membrane structures of cells and tissues. These contents of the lipogenic substances in mulberry leaves are much larger than those of other vitamins, being in agreement with these minimal optimal levels for good growth and development of silkworm.

RELATIONSHIP BETWEEN NUTRITIONAL REQUIREMENT AND METABOLISM

It seems to be very important to know about the relationship between the requirements and metabolism of various kinds of nutrients in the silkworm for understanding the nutrition more deeply.

1. Carbohydrates

Carbohydrates are utilized by the silkworm for energy source and for synthesis of both lipids and amino acids. The digestibility and nutritive effect of carbohydrates are closely related to the activity of the respective hydrolases in the digestive system. Amylase is the most active carbohydrase found in the digestive fluid, whereas the epithelial tissue has several kinds of active glycosidases. The distribution of enzymes in the epithelial tissue suggests that they function an important role in oligosaccharides digestion. In particular, saccharase activity is the strongest among these enzymes in the gut, being good corresponding with large content of sucrose in mulberry leaves.

During starvation of 5th instar larvae, the larvae decreased their levels of fat body glycogen much more quickly than that of hemolymph trehalose. This suggests that some regulatory mechanism function for maintaining a relatively stable levels of trehalose. When a certain amount of each sugar solution was administered to the starved larvae, the fat body glycogen and hemolymph trehalose were recovered from the lowered levels. The reco-

Table 3. *Minimal optimal levels of essential nutrients for silkworm*

Nutrients	Content
1) Amino acid	(mg/g)
Arginine	8
Histidine	5
Isoleucine	8
Leucine	8
Lysine	8
Methionine	4
Phenylalamine	8
Threonine	7
Tryptophan	2
Valine	8
2) Sterol	(mg/g)
Choleserol	2.5
3) Vitamin	(μg/g)
Choline	750
Inositol	1,000
Nicotinic acid	20
Pantothenate	20
Pyridoxine	5
Riboflavin	5
Thiamine	0.5
Biotin	1
Folic acid	1
4) Mineral	(mg/g)
K	8
P	2
Mg	1
Zn	0.01

Horie(1978)

very grade was greatly dependent on the kinds of dietary sugar. Testing this reaction, we can analyze the nutritive value of each sugar for growth of the silkworm.

Using radiochemical method, we could determine the relative pathway participations of glycolysis and pentose phosphate cycle in glucose catabolism of the silkworm. The relative pathway participation of pentose phosphate cycle was calculated to be approximately 35% in the whole body of 5th instar larvae and it much higher in fat body tissues than others. The pentose phosphate cycle is known to contribute to reductive synthesis such as fatty acids synthesis through NADP redoxsystem in this cycle, and the linked enzyme activities were specially strong in fat body tissues.

2. Lipids and sterols

The silkworm larvae require polyunsaturated fatty acids such as linoleic and linolenic acids for their normal growth and development. The visible abnormalities occurred in adult wings on lack of polyunsaturated fatty acids in the diet. The nutritive efficiency was much higher with addition of fatty acid mixture to the diet than that of single one. The nutritive value of vegetable oils is dependent on the composition of fatty acids, and oils from soybean, corn, linseed and safflower are useful for promotion of growth. The fatty acids of mulberry leaves are consisted of palmitic, linoleic and linolenic acids, and there are enough amounts to maintain good growth of the silkworm in mulberry leaves.

The composition of fatty acids in the silkworm tissues altered according to the composition of dietary fatty acids. By radiochemical method, it was proved that palmitic, stearic and oleic acids are synthesized from glucose via pyruvate decarboxylation in the silkworm. The rate of fatty acid synthesis is remarkably influenced by the levels of fatty acids and carbohydrates in the diet, and the dietary fatty acids inhibited fatty acid synthesis in a manner of negative feedback regulation. The retardation of fatty acid synthesis was observed in the omission of biotin from the diet and oleic acid content was reduced conspicuously under this condition. This fact shows that biotin plays an important role through enzyme system in fatty acid synthesis.

It has been shown that lipophorin in the hemolymph serves as the carrier for the transport of various kinds of lipids including diacylglycerol, cholesterol and hydrocarbons from the sites of absorption, storage and synthesis to the site of utilization.

The silkworm larvae require sterol essentially for growth and survival, indicating inability for *de novo* synthesis of sterol in the silkworm. β-sitosterol was found to be main sterol in mulberry leaves and it is of high nutritive value, whereas a great portion of sterol of the silkworm tissues is comprised by cholesterol. Using ^{3}H-β-sitosterol, cholesterol formation was demonstrated to occur in the silkworm. It is quite certain that the conversion of phytosterol to cholesterol by removing the C-24 alkyl group is a prerequisite to the utilization of these sterols. The possible intermediates of β-sitosterol dealkylation such as fucosterol and desmosterol were very effective for good growth.

When the antimetabolite(24, 28-iminofucosterol) for β-sitosterol dealkylation was added to the diet, cholesterol formation from β-sitosterol was strongly inhibited and larval growth was retarded very sharply. The possible metabolic pathway of cholesterol formation ; cholesterol seems to be formed from β-sitosterol via fucosterol, fucosterol 24, 28-epoxide and desmosterol.

3. Proteins and amino acids

Nutrition of proteins and amino acids is particularly of importance for the silkworm larvae because of their active synthesis of silk proteins. The optimal levels of dietary protein in the diet for good growth is 22 to 26% which is in agreement with protein content in mulberry leaves. The protein ingested in the larvae seems to be digested stepwise through digestive cavity and midgut tissue ; proteins seem to be hydrolyzed into low-molecular peptides in digestive fluid at strong alkaline condition and the peptides are further hydrolyzed into amino acids by the midgut peptidases after absorption.

The nutritive value of various kinds of protein are greatly dependent on their compositions of amino acids. The value is very low in wheat gluten and corn zein. The addition of these low-value proteins to the diet results in the retardation of growth, in particular, that of the silkglands and in decrease of hemolymph protein. Under these condition, the excretion of uric acid in the feces is elevated conspicuously. Even in these low-nutritive proteins, when the limiting amino acids were supplymented, the nutritive values of gluten and zein were markedly improved. Consequently, the silkgland growth was accelerated and uric acid excretion was reduced.

The ten essential amino acids for the silkworm are arginine, histidine, isoleucine, leucine, lysine, methionine, phenylalanine, threonine, tryptophan and valine. The each amount of amino acids in mulberry leaves satisfy the requirements for silkworm growth. When any one of the ten essential amino acids is omitted from the diet, protein synthesis in the larval body sharply reduced and the uric acid excretion is steeply elevated. Phenylalanine and methionine requirements can be partly replaced with tyrosine and cystine, respectively. The conversion of phenylalanine to tyrosine and of methionine to cystine via cystathionine has been demonstrated by radiochemical method. Proline is formed from arginine via ornithine, and thus proline requirement can be replaced by either arginine or ornithine in the diet.

Furthermore, the proline synthesis is inhibited by the addition of proline to the diet by the mode of negative feed-back regulation. The omission of both glutamic and aspartic acids resulted in marked retardation of growth. The requirements of these acidic amino acids can be partly replaced with non-essential amino acids, such as alanine, glycine and serine. Certain quantitative ratio of acidic amino acids to non-essential amino acids was necessary for the good growth, and this optimal ratio was varied according to larval development and the sexes.

There are several reports on the composition of free amino acids in hemolymph of the silkworm. Large amount of cystathionine and lanthionine were found in the hemolymph. The quantity of these sulfur amino acids were much greater in the female than in the male. This sexual difference was associated with formation of egg in which a lot of lanthionine was contained. Some mutant(sku) of silkworm accumulated conspicuous amounts of the special amino acids, such as leucine, isoleucine and valine in their hemolymph. This mutant seems to be deficient the enzymes which are responsible for the conversions of the amino acids to the respective fatty acids. This example suggests that the application of special mutant is very useful for study on amino acid metabolism of the silkworm.

The amino acid composition in the hemolymph changes drastically according to the dietary conditions including the kinds of protein, amino acid supplementation to the proteins and vitamin deficiency. The aminotransferase activities in various tissues were remarkably dependent on the dietary pyrudoxine levels, being accompanied with abnormal changes in amino acid composition of hemolymph.

4. Vitamins and minerals

The nine essential vitamins for growth and survival of the larvae are choline, inositol, nicotinate, pantothenate, pyrudoxine, riboflavin, thiamine, biotin and folic acid. Biotin deletion from the diet resulted in alteration of fatty acid composition of the larval tissues as metioned above. The deficiency of pyrudoxine caused a serious disturbance in free amino acid composition of hemolymph.

The minerals essential for growth and survival of the silkworm are, at least, potassium, phosphorus, magnesium and zinc. A lot of minerals accumulated in larval body were excreted at the end of the last instar all at once and this excretion is seemed to be useful for avoidance of excess concentration of minerals in pupal body. In particular, it is noteworthy that zinc is accumulated in the female pupa and is distributed to the egg content. However, the physiological meaning of zinc in the eggs has not been known yet.

ARTIFICIAL DIET AND THE PRACTICAL REARING ON THE DIET

1. Development of practical application

I would like to change the subject to the artificial diet. According to the improvement of diet composition on the basis of fundamental informations, the application of artificial diet technique was going to plan for use of practical sericulture. At present there is little difficulty in rearing the silkworm on artificial diets and in obtaining cocoons of good quality.

The practical use of artificial diets in sericulture of Japan has been carried out since 1977, for the rearing of young larvae of the silkworm in cooperative rearing houses. The practical application of artificial diets in sericulture which has enabled to save a great deal of labor for the rearing and to rear young healthy larvae, has rapidly expanded(Fig 1). As a matter of fact, young larvae hatched from 170 thousand boxes of eggs have been reared on artificial diets in the cooperative rearing houses in 1993.

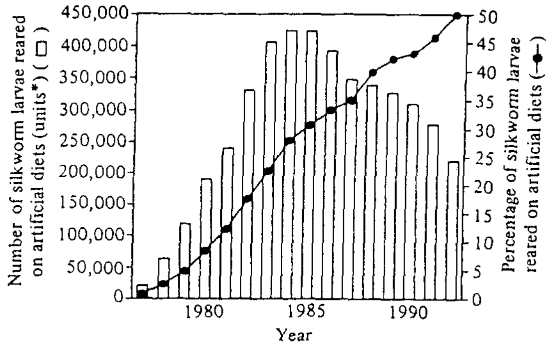

Fig. 1. *Changes with time in silkworm rearing on artificial diets in the cooperative rearing houses.*
**One unit contains 20 thousand larvae.*

One box is a unit of selling eggs and contains 20 thousand eggs or more. Thus, 170 thousand boxes amount to 3.4 billion larvae, which account for about 54% of the total number of larvae reared in that year in the whole Japan. Although the use of artificial diets for the rearing of silkworm from 1st to 3rd instars is desirable for the reasons mentioned above, this practice is restricted to the rearing of larvae from 1st to 2nd instars because of the high cost of diet.

2. Formulation of artificial diets

As generally pointed out, there are at least four main requirements in the formulation of artificial diets for the silkworm ;1) to satisfy both qualitative and quantitative nutritional requirements.

2) to posses the suitable physical properties3) to be free from the pathogenic microbial contamination4) not to contain possible deterrent or repellent for feeding The dietary composition is necessary to be modified according to the larval development. For example, the level of dietary protein should be increased for the 5th instar larvae, and water content should be reduced with larval growth. Physical factors are also very important in the maintenance of good quality of diet. Water content of approximate 75% is favorable for good growth and agar addition is required for keeping good gelation of diet. The pH value of the diet also affects the property of diets including gelation and antiseptic acti-

vity.

The putrefaction of the diet preceeds very quickly, unless controlled by the addition of the suitable antimicrobial agents. Although the aseptic rearing is the most favorable method to avoid microbial contamination, this method is not practical in a large scale rearing in sericulture and it requires a large capital investment. The silkworm, *Bombyx mori*, has strict feeding habit, and they prefer mulberry leaves far better than other plants.

3. Price down of artificial diet

The typical composition of the artificial diet for the practical purpose is shown in Table 4. Generally, commercial diets for the silkworm contain 20 to 25% of mulberry leaf powder on a dry weight basis. Over 5% of agar is added as gelation agent in the diets to keep the water content at a level of 70~75%. However, leaf powder and agar should be reduced in order to lower the cost of diets. The application of a linear programming method is very useful for price down of the diets. To design the composition of artificial diets using a linear programming method, the following necessary conditions must be determined ;

1) Contents of various kinds of nutrients in feed ingredients

2) Amounts of various kinds of nutrients which must be incorporated into the diet.

3) Limiting factors and optimum contents of feed ingredient which affect the feeding behavior as well as larval growth and physical properties

4) Price of feed-ingredient

The optimal formulation of diet composition with the lowest cost was calculated by linear programming method using electronic computer(Table 5). However, this low-cost diet was unpalatable to the normal races of the silkworm. Thus, it began to have a serious desire to breed the silkworms which fed readily on the low-cost diets. The new polyphagous races of the silkworm were bred successfully by Japanese breeder groups after a few years. These races possess a high adaptability to the diet, and were recognized and authorized in 1991~93 by the Japanese Government as a distinctive races. These races possess a capacity for silk productivity almost similar to that of the commercial races(Table

Table 4. *Composition of an artificial diet of the silkworm and cost ratio of feed ingredients*

Feed Ingredients	Amount added (%)	Cost ratio (%)
Mulberry leaf powder	30.0	36.4
Defatted soybean meal	28.0	5.1
Cellulose powder	15.0	6.4
Corn starch	6.1	0.9
Citrate	3.7	1.8
Salt mixture	4.0	4.9
Sucorse	4.0	1.5
Agar	7.0	26.3
Ascorbic acid	0.5	1.3
Vitamin B mixture	0.4	7.4
Phytosterol	0.3	2.2
Soybean oil refined	1.3	0.5
Antiseptics	1.0	5.3
Total	10.13	100.0

Table 5. *Composition of the low-cost diet for the silkworm designed using a linear programming method (Yanagawa et al., 1992)*

Feed ingredients	LPY-501 diet for		
	1st to 2nd instars	3rd instar	4th instar
Mulberry leaf powder	4.000	4.000	–
Defatted soybean meal	35.000	38.000	38.000
Corn meal	40.702	46.506	46.608
Rape bran	–	–	4.000
KCl	0.664	0.194	0.349
K_2HPO_4	1.085	1.483	1.317
$MgHPO_4 \cdot 3H_2O$	0.460	–	–
$MgSO_4$	0.318	0.909	0.844
$CaCO_3$	2.114	2.098	2.221
$FePO_4 \cdot 4H_2O$	0.094	0.069	0.096
Vitamin mix. for chick	0.174	0.128	0.182
Vitamin mix. for eel	0.016	0.200	–
Choline·Cl	0.043	0.035	0.009
Inositol	0.078	0.071	0.092
Ca-pantothenate	0.014	0.013	0.014
Niacin	0.008	0.006	0.007
Pyridoxine-HCl	0.002	0.002	0.002
Ascorbic acid	1.000	1.000	1.000
Citrate	3.000	3.000	3.000
Soybean oil refined	1.153	1.209	1.177
Phytosterol	0.165	0.167	0.172
Antiseptics	0.910	0.910	0.910
Total	100.000	100.000	100.000

Table 6. *Quality of cocoon produced by the polyphagous silkworm race, N601×C601, reared on low-cost diet.*

	LPY-501 diet
Rate of survival of pupae(%)	95.3
Weight of cocoon(g)	1.82
Weight of cocoon layer(g)	0.42
Rate of cocoon layer(%)	23.1
Length of cocoon filament(m)	1,034
Size of filament(d)	3.36
Rate of reelability(%)	86
Rate of raw silk(%)	20.0

Larvae were reared on the diet during the 1st to the end of the 4th instar, and the newly ecdysed 5th instar larvae were transferred to mulberry leaves. (Yanagawa *et al.*, 1992)

6).

5) Improvement of diet processing

At present a few companies are permitted to prepare and to sell artificial diets for practical rearing. The cooperative rearing house used to be supplied with the diets from the companies in the forms of either prepared wet diet or pre-cooked dry powder diet. In the latter case the diets are further prepared by cooking in the local diet-preparation centers which are equipped with various preparation apparatus.

Recently, the new convenient processing method for the diet is developed. In this processing the diet is possible to be prepared very efficiently in the Twin Screw Extruder under a high temperature and pressure in a single process. In the course of this processing, this diet in the shape of pellet(pellet-diet) is formed. By dipping the dried pellet-diet in a appropriate amounts of water, the wet artificial diet can be prepared with ease. The rearing tests on this diet have been done repeatedly with satisfactory results. The pellet-diet will be expected to be useful for convenient preparation, low-priced transportation and long term storage of the diet.

6) Practical mass rearing

At present, the practical mass rearing are carried out in the cooperative rearing house where the automatic rearing machine and feeding machine are installed. The application of this technique is extending in the form of the rearing of young larvae and the half-grown larvae are delivered to each farmer, where they are continuously reared on fresh mulberry leaves for the rest of larval period until cocoon-spinning. As a results, not only farmers can save a lot of labor but also the larvae can avoid the cause of a disease during young larval instarsl

7) Prospects of rearing on artificial diet

In closing. I would like to consider briefly about the prospects of silkworm rearing on the artificial diet in sericulture of Japan. Through both the breeding of polyphagous silkworm races and the development of low-cost artificial diets, it is possible to extend the period of rearing on artificial diets until the end of 3rd instar. Furthermore, when this period of rearing on artificial diets can be extended until the end of the 4th instar practically, farmers may rear only the 5th instar larvae on fresh mulberry leaves only for a week.

This system was designated as "One-Week Silkworm Rearing System". When this system could be applied in practical sericulture, farmers would make possible to rear silkworms about ten times each year, instead of usually three to four times at present. As a result, it may be possible to expansion of scale of sericulture business, without more capital investment and equipment.

On the other hand, the rearing on artificial diet throughout the whole instars will greatly contribute to the safety production of silkworm eggs from the point of pebrine elimination.

REFERENCES

Oukuda, T., Suto, M. & Higuchi, Y. (1960) Silkworm raising on the artificial food. J. Seric. Sci. Japan 29, 1-3,(In Japanese)

Horie, Y. (1978) Quantitative requirement of nutrients for growth of the silkworm, *Bombyx mori* L. JARQ 12, 211-217.

Horie, Y. & Watanabe, K. (1983) Design of the composition of the artificial diets for the silkworm, *Bombyx mori*, by linear programming method: application of the ingredients of feeds for domesticated animals and fowls. Bull. Seric. Exp. Stn., 29, 259~283, (In Japanese with English summary).

Ito, T. (1983) Nutrition and artificial diets of the silkworm, *Bombyx mori*. 1~292, Nihon-Sanshi-Shinbun

Press, Tokyo.

Ito, T. & Tanaka, M. (1960) Rearing of the silkworm on an artificial diet and the segregation of pentamolters. J. Seric. Sci. Japan 29, 191~196,(In Japanese with English summary).

Kanda, T. (1992) Genetical study of feeding habits of the silkworm, *Bombyx mori,* to new low-cost diet designed by linear programming method and application to the breeding. Bull. Natl. Inst. Seric. Entomol. Sci., 5, 1~89,(In Japanese with English summary).

Mano, Y. *et al.* (1991) Breeding and evaluation of adaptability of the silkworm, *Bombyx mori,* to the new low-cost artificial diet, LPY lacking mulberry leaf powder. Bull. Natl. Inst. Seric. Entomol. Sci., **3**, 31~56, (In Japanese with English summary).

Miyazawa, F. (1993) The present situation and prospects in the study on artificial diets of the silkworm. Sanshi-Gijutsu(Sericultural Techniques), 145, 6~11 (In Japanese).

Shinbo, H. & Yanagawa, H. (1994) Low-cost artificial diets for poly-phagous silkworms, JARQ 28, 262~267.

Yanagawa, H., Suzuki, K. & Watanabe, K. (1992) Exclusion of agar in an artificial diet for polyphagous strains of the silkworm, *Bombyx mori.* J. Seric. Sci. Jpn., 61, 158~164 (In Japanese with English summary).

Yanagawa, H., Watanabe, K. & Suzuki, K. (1991) Development of a low-cost artificial diet-development of an artificial diet by applying a linear programming method for polyphagous strains of the silkworm, *Bombyx mori.* Bull. Natl. Inst. Seric. Entomol. Sci., 3, 57~75, (In Japanese with English summary).