

Experiments concerning the Utilization of the
Bean Basic Semisynthetic Media for Rearing
Two Species of Cutworm, *Agrotis ipsilon*
HUFNAGEL and *A. fucosa* BUTLER
(Lepidoptera, Noctuidae)*

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INTRODUCTION

The black cutworm, *Agrotis ipsilon* HUFNAGEL, and the turnip moth, *A. fucosa* BUTLER, are two main species of the Lepidopterous cutworm, of which the extensive damage is widespread throughout Okayama prefecture where tobacco is grown (KOIZUMI · KIYOKU 1959). Whereas, comparatively little work has been done on the detailed experimental studies on ecology, population dynamics, behaviour and habit which are related to the control of these insect pests, because of the difficulties for rearing large numbers from generations to generations. It is necessary, therefore, to devise first the artificial mass rearing method for such experimental studies as described above. Of course, there have been several investigations dealing with the laboratory mass rearing of the insects belonging to the Genus *Agrotis*. For instances, *Agrotis segetum* was reared by GÖRNITZ (1951) on the potato tuber etc., *A. ipsilon*, by HARRIS et al. (1958) on the red clover and by GEORGE et al. (1960) on the artificial media containing a ground lucerne, *A. orthogonia*, by JACOBSON and KLAKELEY (1957) on the wheat sprouts, and by MCGINNIS and KASTING (1959) on the wheat sprouts of thatcher, and *A. exclamationis*, by KOWALSKA (1962). However, there have been few investigations with the rearing of *Agrotis* on the semisynthetic or totally synthetic media.

The writers were able to rear successfully the tobacco cutworm, *Prodenia litura* FABRICIUS, by using the bean base semisynthetic media (KIYOKU 1966, KIYOKU · TSUKUDA 1966). Moreover, several improved methods were also used in the artificial rearing (KIYOKU · TSUKUDA 1966, KIYOKU · TSUKUDA 1967). Now, the present writers have conducted several experiments to determine whether the bean base semisynthetic media and improved rearing method of the tobacco cutworm can be adapted for the artificial rearing of both black cutworm and turnip moth. This report deals with the artificial culture of the two species and their life historical data obtained

* This work was presented at the Annual Meeting of Japanese Society of Applied Entomology and Zoology, Tokyo, April 3, 1967.

from such experimental cultures.

MATERIALS AND METHODS

The cultures of both black cutworm and turnip moth were started with larvae collected from the cabbage in a field. These larvae were reared on the leaves of cabbage in the laboratory until pupation. When adult moths emerged, they were allowed to mate and oviposit in the glass cylinders. Since these insects did not lay eggs on paper that lined on the inner surfaces of the glass cylinders, the egg plant sprouts were used for oviposition sites. When eggs hatched on the plant, the larvae were removed to a petridish, reared on the leaves of sweet potato until they reached the second instar. These second instar larvae were used in these experiments on the artificial rearing. The basic ingredient in the semisynthetic media was also kidney, soy or azuki bean seeds. These semisynthetic media were called the kidney bean media, soy bean media and azuki bean media, respectively. The media composed of two kinds of beans were called the kidney-soy bean medium and soy-azuki bean one. The composition of ingredients in the kidney bean media and soy bean ones, and procedure for preparing the media were almost similar to those already reported for rearing the tobacco-cutworm on semisynthetic media (KIYOKU 1966). Ratios of mixing two beans were 3:7 for the kidney-soy bean media and 5:5 for the soy-azuki bean media. Rearing vessels and the procedure for rearing larvae, and handling of insects also were not appreciably different from the procedure published in the previous paper, except the procedure taken for ovipositions.

RESULTS

1. Rearing the black cutworm on various semisynthetic media through one complete generation.

Data for the life history of black cutworms reared on four kinds of semisynthetic media are summarized in Table 1.

According to Table 1, the duration of growing period of the larvae reared on each semisynthetic medium tended to become somewhat longer than that of the control. Especially, that of larvae reared on the soy bean media was the longest of all. The duration of prepupal period did not differ much from that of the control, except in the case of the soy bean media in which it was longer than in the control. Percentage of pupation was not always high although many larvae grew well on each medium. Many larvae died just before reaching prepupa or deformities appeared frequently. Consequently, a small number of normal pupae was obtained and therefore the percentage of pupation was not large. Such a tendency was also seen in the control. Only the percentage of pupation in the kidney bean media, however, was considerably higher, being 66 percent. The weight of pupae varied to some extent according to the kinds of larval diets. Generally speaking, however, the diffe-

Table 1. Life history data of the black cutworms reared on some semisynthetic media through one complete generation.

Charatcers examined	Media	Kidney bean media	Soy bean media	Kidney-Soy bean media	Soy-Azuki bean media	Control (Sweet potato leaves)
Number of 2nd instar larvae		50	50	30	60	50
Duration of larval period (days)		18.5	23.0	20.5	21.0	13.5
Duration of prepupal period (days)		2.2	3.5	2.5	2.0	2.0
Percentage of pupation		66.0	20.0	46.6	26.7	48.0
pupal weight (mg.)		♂ 291.0 ♀ 303.0	307.5 343.0	289.5 292.5	349.9 386.6	311.5 330.7
Duration of pupal period (days)		11.0	13.0	13.5	12.0	10.5
Percentage of emergence from pupae		30.3	70.0	57.1	69.1	87.5
Number of normal adults emerged		♂ 5 } 10 ♀ 5 }	3 } 7 4 }	4 } 8 4 }	6 } 11 5 }	9 } 17 8 }
Percentage of yield of adults from initial larvae		20.0	14.0	26.7	18.3	42.0
Number of eggs per female		234.0	203.0	211.0	452.0	947.0
Hatchability (%)		50.2	18.5	21.3	26.5	88.6

rence was not significant. The duration of pupal period showed a tendency to vary according to the differences in semisynthetic media and seemed to be usually longer than that of the control. It was frequently observed that when the percentage of pupation was high (or low), that of emergence from the pupae became inversely low (or high). Owing to such a relationship, the percentage of emerged adults from initial second instar larvae could not attain to a high value and represented always rather small values. Even the highest percentage obtained from the kidney-soy bean media did not exceed 26.7 %. Utilizing the semisynthetic media developed in the previous experiment with the tobacco cutworms for the present rearing of the black-cutworms, it was found that some normal adults could be obtained although the percentage in emergence of adults was low. These adult females laid eggs scatteringly on the leaves of plants provided for their oviposition sites. Moreover, numbers of eggs laid varied widely due to differences in the environmental factors. On account of such circumstances, it was difficult to determine the accurate numbers of eggs deposited by females. Roughly speaking, however, it was usually possible to obtain

above 200 eggs per female on this artificial rearing. In spite of being the low hatchability of these eggs, a considerable number of young larvae was obtained for rearing the second generation. The deformities of pupae and / or adults appeared frequently throughout the artificial rearing, which were not included in Table 1. The pupa deformities had often a weakend area between the abdomen and thorax, and most of the adult deformities had unexpanded wings.

From the present experiments, we may state that the artificial rearing of the black cutworms is successful to some extent by utilizing the semisynthetic media developed in the rearing of tobacco cutworms which was conducted in the previous experiments. Although it is difficult to decide which is more suitable for the present cultures among several media, the kidney bean media seem to be more suitable than the other media as the artificial diets of these insects.

2. *The successive laboratory rearing of black cutworms on the kidney bean media.*

Three successive generations of the black cutworms have been reared on the kidney bean media. The results are given in condensed form in Table 2.

Table 2. Life history data of the black cutworms reared on a kidney bean medium through three successive generations.

Characters examined	Number of initial 2nd instar larvae	Duration of larval period (days)	Duration of prepupal period (days)	Percentage of pupation	pupal weight (mg.)	Duration of pupal period (days)	Percentage of emergence from pupae	Number of normal adults emerged	Percentage of yield of adults from initial larvae	Number of eggs per female	Hatchability (%)	
Generation	I	50	18.5	2.2	66.0	$\begin{matrix} \uparrow 291.0 \\ \uparrow 303.0 \end{matrix}$	11.0	30.3	$\begin{matrix} \uparrow 5 \\ \uparrow 3 \\ \uparrow 10 \end{matrix}$	20.0	234	50.2
	II	30	18.0	1.8	53.3	—	11.5	18.3	$\begin{matrix} \uparrow 1 \\ \uparrow 2 \end{matrix}$ } 3	10.0	303	55.1
	III	30	16.8	2.8	36.6	$\begin{matrix} \uparrow 309.8 \\ \uparrow 320.5 \end{matrix}$	12.6	45.5	$\begin{matrix} \uparrow 2 \\ \uparrow 3 \end{matrix}$ } 5	16.6	201	31.2

Data on the kidney bean media shown already in the Table 1 were represented again in Table 2 as the results of the first generation, because of convenience for comparing with each result obtained from other successive generations. Durations of larval, prepupal and pupal periods in the second generation were almost similar to those in the first generation. However, both percentages of the pupation and of the yield of adults from the initial larvae decreased as compared with those of the first generation. Emerged adult moths were only a male and two females. When this male was mated with a female, the female laid fortunately 303 eggs. As the over

half of the egg mass hatched, the writers were able to begin the rearing of the third generation.

In the third generation, the percentage of pupation became smaller and the lowest of all generations. The number of eggs per female and their hatchability did not increase. However, the growth rate of larvae was fast, and the duration of larval period shortened, the percentage of emergence from the pupae increased and the pupal weight was never inferior or rather superior to that of the first generation. From these experiments, the artificial rearing of the black cutworms on the semisynthetic media seemed to be more difficult than the rearing of tobacco cutworms. However, the writers believe that we shall be able to rear the black cutworm more successfully if a suitable media are developed further and suitable modifications of rearing methods are made in the future.

3. *Observations on the growth and development of black cutworm reared individually on the kidney bean media.*

In another experiment, twelve second instar larvae of the black cutworm were reared individually on the kidney bean media, using the same procedure as that used in the previous experiments with the tobacco cutworm (KIYOKU・TSUKUDA 1967). The numbers of moulting and instars, duration of each instar period, width of head capsule of each instar larvae and durations of prepupal and pupal period were determined more exactly than those in the former mass rearing experiments. The results obtained are summarized in Table 3. The data obtained from the control (the rearing on the sweet-potato leaves) are shown on each right side in the table 3.

Table 3. Experimental results on the growth of black cutworms reared individually on the kidney bean media.

Media Instar or stage	Kidney bean media		Control (Sweet potato leaves)	
	Duration of growing period (days)	Width of head capsule (mm.)	Duration of growing period (days)	Width of head capsule (mm.)
2nd instar	2.2	0.42	2.3	0.43
3rd instar	2.3	0.75	1.8	0.66
4th instar	3.3	1.18	2.5	1.14
5th instar	3.8	2.06	3.1	2.06
6th instar	6.8	3.14	5.9	3.17
Larval stage	18.4	—	15.6	—
Prepupal stage	1.6	—	2.0	—
Pupal stage	♂ 11.5	—	9.5	—
	♀ 12.0	—	10.5	—

According to Table 3, the differences between data obtained with the kidney bean media and those of the control were not significantly large. The larvae moulted five times and passed six instars during their growing periods. Duration of instar period

became longer as the instar progressed. Duration of pupal period tended to be somewhat shorter for the male than for the female. The width of head capsule of larvae grew from 0.42 mm. for the second instar to 3.14 mm. for the sixth one.

In this individual rearing experiment, seven pupae was obtained from twelve larvae, one of which was a malformation pupa. Five normal moths emerged successfully from those seven pupae of which two were males, and the other three females. The pupal weights increased as compared with those obtained in the former experiments, they were 367.5 mg. for male and 480.5 mg. for female. The percentages of pupation, of emergences from the pupae and of yield of adults from the initial number of second instar larvae were found to be 58.3%, 71.4 % and 41.6%, respectively. When two males were mated with two females of all emerged adults, the two adult females laid 1260 eggs and 800 out of these eggs hatched normally. Therefore, the fecundity or number of eggs per female was 630 and hatchability of the eggs 55.5%, as a result of calculation. These results were the best ones of all the results obtained in the present experiments.

4. *The successive laboratory rearing of turnip moth on the kidney bean media.*

The successive rearing of turnip moths have been tried by using the same procedure and the same semisynthetic media as used in the successive rearing of the black cutworms. To date, three successive generations have been reared but the full data on the third generation can not be obtained by this time. Accordingly, data of rearing of the first two successive generations are briefly summarized in Table 4.

Table 4. Life history data of the turnip moths reared on a kidney bean medium through two successive generations.

Characters examined	Number of initial 2nd instar larvae	Duration of larval period (days)	Duration of prepupal period (days)	Percentage of pupation	Pupal weight (mg.)	Duration of pupal period (days)	Percentage of emergence from pupae	Number of normal adults emerged	Percentage of yield of adults from initial larvae	Number of eggs per female	Hatchability (%)
Generation I	54	28.6	2.9	77.8	448.7	15.6	83.3	$\begin{matrix} \uparrow \\ + \\ \circ \\ \circ \end{matrix} \left. \begin{matrix} 12 \\ 23 \end{matrix} \right\} 35$	64.8	543.8	76.1
Generation II	30	$\begin{matrix} \uparrow \\ \circ \\ \circ \end{matrix} \left. \begin{matrix} 27.0 \\ 28.2 \end{matrix} \right\}$	3.0	60.0	$\begin{matrix} \uparrow \\ \circ \\ \circ \end{matrix} \left. \begin{matrix} 438.8 \\ 488.8 \end{matrix} \right\}$	$\begin{matrix} 15.2 \\ 17.6 \end{matrix}$	$\begin{matrix} 41.7 \\ 50.0 \end{matrix}$	$\begin{matrix} 5 \\ 3 \end{matrix} \left. \right\} 8$	26.7	134.7	35.8

As shown in Table 4, twelve normal adult males and twenty-three normal adult females were obtained from the fifty-four initial second instar larvae, the percentage of yield of adults attained to 64.8 %, and the resultant adult females could lay 543.8 viable eggs per female. The duration of larval period upto prepupae was distinctly longer than that of the black cutworms in the former cultures, being on an average

28.6 days. The duration of pupal period was also relatively longer, being 15.6 days on the average. Since the individual cultures were not conducted, exact data could not be obtained. From the present mass rearing, however, it was presumed that the numbers of moulting and of instars passed were five and six, respectively. The average width of head capsules of the larvae of successive instar were presumed to be 0.75 mm. for the second, 1.15mm. for the third, 2.01mm. for the fourth, 2.28mm. for the fifth and 2.98mm. for the sixth instar.

In the rearing of the second generation, both percentages of pupation and emergence decreased, the egg production was less as compared with those in the first generation. On the other hand, there were no great differences in the durations of larval, prepupal and pupal period between two generations. Generally speaking, the results of the second generation seemed to be more or less inferior to those of the first generation. Whereas the appearances of deformities in both pupae and adults were small in numbers.

Since over one-third of the eggs deposited in the second generation hatched, the consecutive rearing for the third generation could be conducted. As stated above, the culture of third generation has not been completed yet, but the growth and development of larvae seemed to be better than in the former generation. Thus, it is expected that the successive generations of turnip moth also can be reared successfully on the semisynthetic media composed of the kidney bean as a basic ingredient.

DISCUSSION

From the present experiments, it has been expected that the successive generations of two species of cutworms can be reared successfully on the bean basic semisynthetic media. For the purpose of giving better results, of course, the rearing procedure leaves room for improvement. Such improvement may concern the environmental conditions of rearing as well as the physical conditions of the larval diet.

Sometimes a marked reduction of the larvae of the black cutworm occurred at the beginning in larval period. Such a reduction influenced greatly the percentages of pupation and emergence. One of the causes was presumed to be the physical conditions of the media such as water contents or solidity of the media. If solidity of the media was inadequate, it was difficult for larvae to take diets, consequently, they grew slowly and some larvae died by starvation, or died being caught by the diets when they came upon contact with diets. After the fourth instar, whereas, many larvae grew normally and there was no tendency to cannibalism among the larvae. Consequently, no remarkable reduction of the individuals during the larval period occurred. During the prepupal and pupal stage, however, numerous individuals became abnormal, usually dark in color and compressed in shape or deformities were frequently seen among the prepupae and pupae obtained. The causes for these phenomena seemed to be the desiccation as well as the composition of the media.

In the rearing plots in which the percentage of pupation was low, the high percentage of emergence was obtained on account of the facts that most of the pupae could emerge to adults. On the other hand, in the rearing plots in which the abnormal pupae often appeared, the percentage of emergence decreased in spite of high percentage of pupation, by the reason that considerable numbers of these pupae were abnormal, and consequently failed to emerge as the normal adults.

In the case of the turnip moth the young larvae grew faster and a reduction of individuals also was smaller as compared with the black cutworm. However, the mortality of older larvae was often high, especially higher under a dry condition. It is necessary to control the moisture in the media as well as in the rearing environment. Regarding the condition of the media, it seems to be better to rear the larvae on somewhat solid media.

SUMMARY

The present writers have tried several experiments to determine whether the bean basic semisynthetic media and the improved rearing method for rearing the tobacco cutworm, *Prodenia litura* FABRICIUS, in the previous experiments can be adapted for the present artificial cultures of both the black cutworm, *Agrotis ipsilon* HUFNAGEL, and the turnip moth, *A. fucosa* BUTLER.

1. Both normal adult males and females which could lay viable eggs could be reared successfully from the second instar larvae of black cutworms on four kinds of media—the kidney bean media, soy bean media, kidney-soy bean media and soy-azuki bean media, though the duration of larval growing period became longer and the percentages of pupation, emergence and the yield of adults obtained from the larvae initially used were lower than those from the control which was fed on the leaves of sweet potato. The kidney bean media seemed to be more suitable for the artificial rearing of this insect as compared with three other bean basic media.

At 26°C., the durations of larval, prepupal and pupal period were 18.5, 2.2 and 11.0 days, respectively. The percentages of pupation, emergence and yield of emerged adults from the initial second instar larvae were 66.0, 30.3 and 20.0 %, respectively. The pupal weights were 291.0 mg., for males and 303.0 mg. for female. The number of eggs laid per female was 234 and the hatchability of these eggs, 50.2 %.

2. Using the kidney bean media, which seemed to be the most promising medium, three successive generations of the black cutworms were reared successfully as shown in Table 2. However, the results were slightly inferior to those in the previous experiments of *Prodenia litura* FABRICIUS.

3. In the experiments in which the larvae were reared individually using one larva per glass tube, the numbers of moulting and larval instars passed, duration of each instar period, width of head capsule of each instar were observed more exactly than in the case of the above culture. The percentages of pupation, emergence and

yield of adults were also examined accurately in this experiment. These results were considerably superior to those obtained in the above ones except that the duration of larval period was longer.

4. Three successive generations of turnip moths have been reared using the same procedure and kidney bean media as used in the successive rearing of the black cutworms. Although full data on the third generation could not be obtained by this time, it was expected that these successive generations could be reared successfully as in the rearing of the black cutworms. The life history data of the turnip moth reared on the kidney bean media at 26°C. were as follow. The durations of larval, prepupal and pupal period were 28.6, 2.9 and 15.6 days, respectively. The percentages of pupation, emergence and yield of adults were 77.8, 83.3 and 64.8 %, respectively. The pupal weights were 438.8 mg. for males and 488.8 mg. for females. The number of eggs laid per female was 543.8 and the hatchability of these eggs, 76.1 %.

ACKNOWLEDGEMENTS

The authors wish to express their grateful appreciations to Dr. C. HARUKAWA, emeritus professor of Okayama University for reading the manuscript. Many thanks for assistance are due to Messrs. T. YOSHII, AKIYAMA and Mrs. S. YOSHIDA of our laboratory.

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2種のネキリムシの飼育へ豆を基礎とする 半合成飼料を利用することに関する実験

清久正夫・佃律子

摘 要

筆者らが前にハスモンヨトウの人工飼育において確立した豆を主体とする半合成飼料と飼育法を今回は2種のネキリムシ——タマナヤガとカブラヤガの人工飼育に応用してみた。その結果カブラヤガは勿論タマナヤガにおいても前のハスモンヨトウの場合より幾分飼育成績が劣ったが、金時や大豆など1種のみよりなる半合成飼料もそれらの2種混合の飼料においても比較的容易にこれらの虫を飼育することができた。使用した飼料のうちで比較的によいと思われた金時飼料によって両種の累代飼育を試みたところ3世代以上の連続飼育の可能なことがわかった。しかし昆虫の種類によって用いる飼料の条件を多少変化させる必要を感じた。飼料の成分についてはあまり問題はないように思ったが、その含有水分と環境湿度および飼育中の虫の管理に改善を要する点が認められた。なお飼育容器当たり1頭ずつ飼育するいわゆる個体飼育をも実施してその發育状態をこまかに観察した。