Studies on the Biological Control of the Chestnut Gall Wasp, *Dryocosmus kuriphilus* Yasumatsu (Hymenoptera: Cynipidae), with Particular Reference to the Utilization of Its Indigenous Natural Enemies*

By Torizo TORII**

(*Appl. Entomological Laboratory, Fac. Agric., Shinshu Univ.*)
(*With 12 Text-figures and 5 Plates*)

Contents

Page

Introduction 73

(I) Interrelationship between the emergence of the chestnut gall wasp and that of its parasitic wasps in Ina district 75

(1) The object of research. (2) Materials and method of research. a) Method of collecting galls. b) Preservation of galls and enumeration of the emergents. (3) The species of indigenous parasitic wasps and their relative ratio in individual numbers. (4) Interrelationship between the emergence of the host gall wasp and that of its parasitic wasps. a) Biostatistical method of analysis. i) Probit method. ii) Logit method. (5) Analysis of data. a) Comparison of validity between the logit and the probit method. b) Biological considerations. (6) Yearly fluctuation in the trend of emergence. a) Constancy of yearly trend of emergence. b) Constancy of emergence period. (7) The determination and the prediction of the period fittest for the biological control of the chestnut gall wasp. a) How to determine the period fittest for the biological control. b) How to predict the period fittest for biological control. i) Logit 75% date estimated from the data in the early stage of emergence. ii) How to etimate the maximum value of the cumulative number of the emergents. iii) The results obtained from the outdoor emergence trap. iv) The method of prediction to be recommended. (8) Advantage of the logit transformation. a) Applicability to the analyses of the cumulative percentage curves in general. b) Generalized logit method. c) Short-cut method of the logit transformation.

(II) The experimentation with the liberation of a key parasitic wasp.... 113

(1) Materials and the environmental conditions of the area where

---

* (1) Aided by a part of the grants from the Ministry of Agriculture and Forestry (from 1955 to 1957) and by grants from the Ministry of Education (in 1958).

(2) Chapters I-II were presented at the annual meeting of the Japanese Society of Applied Entomology and Zoology held at Tokyo, March 30-31, 1957, and Chapter III at the same meeting held at Tokyo, April 5-6, 1958, respectively.

** Professor of applied entomology.

Page

liberations were conducted. (2) Evaluation of release effectiveness. (3) Discussion.

(III) Host preference by the P^{32}-labeled parasitic wasps 119

(1) The object of research. (2) Method of research. a) Parasitic wasps employed. b) Method of tagging. i) Preparation of a tracer. ii) Release of P^{32}-tagged wasps. c) Preparation of specimens of dried leaves and galls. d) Preparation of macro-autoradiographs. (3) Results. i) Results obtained in 1958. a) Radioactivity of P^{32}-tagged parasitic wasps. b) Radioactivity of the plants caged with the P^{32}-labeled parasitic wasps. c) Analysis of macro-autoradiographical results. i) Descriptive features. ii) Results obtained in 1959. a) Radioactivity of P^{32}-tagged parasitic wasps. b) Radioactivity of the plants caged with the P^{32}-labeled parasitic wasps. c) Macro-autoradiographical results. (4) Discussion. a) From the view-point of parasite biology. b) Biological control combined with chemical control. 1) The present situation. 2) An experiment on dusting or spraying γ-BHC. i) Application to the chestnut galls collected in the early season. ii) Application to the chestnut galls collected at the maximum period of emergence of the chestnut gall wasp. iii) summary and conclusion. iv) Recomended control measures.

(IV) Summary and conclusions 142

(V) Literature 146

INTRODUCTION

A chestnut gall wasp, *Dryocosmus kuriphilus* Yasumatsu, is one of the most serious insect pests of chestnut trees in Japan. Once infested by this gall wasp, most of chestnut trees, cultivated or wild, cannot be free from its severe damage. A parasitized bud grows into a gall, somewhat ball-like in outline, about the size of half a kidney bean, occasionally into a monster gall as large as a strawberry in early summer. (phot. I, a) When severely attacked, the branches and twigs of the infested tree become deformed in the foliage, occasionally leading to its dying, if it is not killed outright. According to M. Yokoyama and M. Kinoshita (1951), this gall wasp, which is a native of Japan, was first reported from Kobe in 1945. Since then, it has spread with astonishing rapidity, throughout nearly all the prefectures except the northeast prefectures such as Hokkaido and Aomori, and the southeast prefecture, Kagoshima, probably due to its parthenogenetic propagation as well as its broad adaptability to different climates. The case is similar in Ina district. In Kamiina, the northern part of Ina district, no damage due to this gall wasp was reported in 1955, but in the beginning of summer of 1958, it already spread almost all over the localities therein. So far, the standard recommended control measures have been centred on plucking almost recklessly the galls from the infested trees and committing them to the flames. This control has, however, not always brought a statisfactory success as compared with much labour required, and the damage has been spreading widely year after year. The reason for this seems to lie in the following two: One is a serious drawback of this control measure; that is, by this all the parasites within the gall are destined to be burnt together with the gall, which in its turn leads up to a remarkable lowering of environmental resistance to the host in question; the other is that the infested wild chestnut trees, the galls of which escaped from being plucked, are responsible for the widespread of this gall wasp. To cope with this situation, the establishment of some more adequate, effective control measures has keenly be felt for years. A few reports have hitherto been published on this subject. As has been emphasized by Dr. K. Yasumatsu, however, there seems to be no other way for it but to invoke the aid of the natural enemies parasitic on this gall wasp to be controlled. Fortunately, about 11 species of effective Chalcidoid parasites have been discovered by Dr. K. Yasumatsu through his elaborate study. These are all native to Japan, being parasitic on the chestnut gall wasp. If some adequate measures should be taken to preserve these resident species at a higher level as much as possible, the future of the control of this gall wasp would be very bright. Hence, the question resolves itself down to how to preserve such indigenous parasitic wasps at the highest possible level. The answer to this question may be given by the elucidation of the parasite-host interrelationship concerning the period and/or trend of their emergence. If this is clarified, the way to satisfactory control, biological or insecticidal or both combined, may be opened. No reports in this rsepect have ever been published, so fas as I am aware.

Under the guidance of Dr. K. Yasumatsu, I have been investigating this problem these 5 years since 1955, with the data obtained from wild chestnut galls collected at various localities in Ina District, the southern part of Nagano prefecture. As the result, I could formulate the logit method available for predicting what I call the "period fittest for biological control", viz., the period most suitable for checking the host wasp at its minimum level and preserving its native parasitic wasps at their maximum level. If this method of prediction is effectual, and a subsequent control measure is adequate, it will not be impossible to increase the percentage of parasitism for the parasites to the extent of about 7~8-fold. As has been pointed out by Dr. Yasumatsu, almost no endeavours have hitherto been made in our country to augment the role played by resident natural enemies, in so far as the insect pests native to Japan are concerned, and the rational measure fit for it has long been waited for. The present paper will throw some light on it, I believe.

Another problem dealt with in this paper is the liberation experimentation with an effective parasitic wasp not inhabiting here. The result has been satisfactory, though the problem of permanent establishment remains to be seen. Furthermore, by using radioactive phosphorus P^{32} as a tracer, attempts have been made to make an experiment in the elucidation of host preference by the parasitic wasps under study. In addition to this, by applying γ-BHC and γ-BHC-1-C^{14}, an experiment* has been undertaken to clarify whether the combined use of natural enemies and insecticides is really possible or not. Although farther investigations are necessary, all the results have been encouraging and highly suggestive.

This work owes much to the kind guidance given by Dr. K. Yasumatsu, Professor of entomology at Kyushu University, under whose direction the greater part of this study has been performed; Dr. T. Esaki, late Professor of entomology at the same university, and Dr. T. Kitagawa, Professor of statistics at the same university, had the kindness to suggest to me biostatistical, applied ecological studies on parasitic wasps in relation to biological control. To all of them, my most grateful acknowledgement must be paid. To Dr. S. Ishii, Chief of the 1st Section of Insect Pests Control at the National Institute of Agricultural Sciences, I am indebted for much information and technique concerning the radioisotope experiment. To Dr. M. Fukaya, Chief of the Entomological Bureau at the same institute, I am much indebted for many valuable suggestions relating to the aspects of occurrence prediction. It is with my sincere gratitude that the influence of unremitting encouragement by Dr. N. Yagi, Professor of entomology at the Faculty of Textile and Sericulture, Shinshu University, is acknowledged heartily by me. I heartily appreciate the kindness shown by Mr. K. Nobara, Member of the Entomological Laboratory at the Kyushu University, who cooperated with me in identifying the specimens of parasitic wasps concerned. I am particularly indebted to Miss. M. Kamiya, Member of our laboratory, for her much assistance in all phases of this investigation. For this work I was subsidized with a part of the "Grant-in-aid for applied study" from the Ministry of Agriculture and Forestry (from 1955 till 1957) and with the "Grant-in-aid for personal study" from

* The experiment with γ-BHC-1-C^{14} is now in progress. The result is expected to be made public in Japanese Jour. Appl. Ent. Zool. in the near future.

the Ministry of Education (in 1958), respectively. Towards the authorities concerned go my deepest thanks. Finally, but not least, I have to express my gratitude sincerely for the kindness and cooperation shown by many officials concerned at the Forestry Section, the Shimoina Local Office, during the whole course of this investigation. The publication of this monograph has been made possible through the special good offices by Dr. N. Shirakura, Head of the Detached Library of our faculty, and by Dr. H. Mimura, Dean of the Faculty of Agriculture of our University. I take this opportunity to express my sincere gratitude to them.

(I) INTERRELATIONSHIP BETWEEN THE EMERGENCE OF THE CHESTNUT GALL WASP AND THAT OF ITS PARASITIC WASPS IN INA DISTRICT

(1) The Object of Research

As touched in the introductory remarks, the key to the solution of the problem concerning the biological control of the chestnut gall wasp is to clarify the interrelationship between its emergence and that of its parasitic wasps. The result obtained from the data in 1955 indicated that every wasp parasitic on the chestnut gall has a tendency to emerge about 10 days earlier than its host gall wasp does and to complete its emergence about two weeks ealier. It was furthermore found that the cumulative percentage emergence curve for the chestnut gall wasp and/or for the composite parasitic wasps has a pictorial appearance of the logistic curve or of the integrated normal curve, respectively. To determine whether or not these results imply the general interrelationship between the emergence of the two is of great importance in establishing the most reasonable control measure of the pest in question. From this angle, I made a detailed analysis of the data obtained during the past 5 years from 1955 till 1959, and found that the above finding in 1955 does not by any means indicate an exceptional case, but the general interrelationship between the two, so far as Ina district is concerned.

(2) Materials and Method of Research

a) Method of Collecting Galls

All the galls were taken from wild chestnut trees which were grown in forests or in scrub forests in Kamiina District (the northern part of Ina district) and/or in Shimoina District (the southern part of Ina district). The amount of galls collected at one location varies according to the degree of infestation of the trees, ranging from 50-100 grams (at Kamiina in 1956) to 500-1000 grams (at Shimoina in 1955-1957, and at Kamiina in 1957-1959). The census sampling consisted of the following procedure: As the representatives of each district, several localities where collections were made were selected. The sampling at each location was in principle performed according to the rule of random line sampling. First, the direction along which a collection is to be made was determined at random; secondly, 10-20 trees were selected at intervals of about 10 steps in the direction determined; and finally, one twig about 1 cm. thick was taken from each of the selected trees, all the galls grown on it being plucked. A twig unit per tree was selected randomly from the branches spreading in a definite direction. The numerical abundance of the galls thus collected gives a very reliable sample for the area concerned, and a twig sample for gall number may be a highly reproducible index of the damage at that area. In every sampling, attention was paid to the maintenance of its randomness, since the chestnut gall shows more or less tendency to colonize in distribution in area and even within one tree, as is usual with the spacial distribution of insect pests, and as aptly pointed out by Dr. K. Yasumatsu (1956). When the selected trees are tall, the clippers originated with me, as illustrated in phot. I-b, were employed. Gall gathering was as a rule carried out once in a period of ten days from the middle of June till the end of the same month. Although it was made in the latter half of May in some cases, it was found unfavorable, because the chestnut gall wasp tended to become unable to emerge normally from the young galls clipped in such an early season, while the parasitic wasps were able to emerge from them equally as they did from the older galls. The relative abundance of the composite parasitic wasps in Kamiina in 1956 may be ascribed to this cause. The details pertaining to gall collectin are shown in table I.

b) Preservation of Galls and Enumeration of the Emergents

The collected galls, which were sorted by localities and dates, were put into a small paper bag at the rate of about 100 grams per bag, and were stored on the rack as illustrated in phot. II-a. In 1959, an emergence trap was used to enumerate all the emergents from the collected chestnut galls. (phot. II-b). This trap proved itself to be very convenient and serviceable for saving labor. After the collection was over, the number of galls to 100 grams was counted by way of comparison of the degree of infestation. The emergents were caught with a suction pipe at intervals of one day as a rule, and afterwards were allowed to feed on honey soaked up in decapitated matchsticks which were placed inside a glass test tube 2.5 cm. in diameter and 18 cm. long. (See phot. III). At every census counting, counts were taken of

TABLE I. Number of localities where the chestnut galls were collected, the date on which they were collected, their weight in grams, and number of both the chestnut gall wasps and their parasitic wasps that issued from the galls.

Year	District	Chestnut galls collected (Weight in grams)	No. of localities	Date	No. of chestnut gall wasps	No. of parasitic wasps
1955	Kamiina	Almost no infestation was recorded.				
	Shimoina	1807	4	June 22–28	2306 (127.6)	108 (6.0)
1956	Kamiina	318.6 (161*)	6	May 13–June 14	117 (36.7)	79 (24.8)
	Shimoina	14185 (116*)	12	May 18–July 4	7797 (55)	910 (6.4)
1956	Kamiina	2650 (200*)	2	May 30–June 3	1002 (37.9)	206 (7.8)
	Shimoina	2715 (161.4*)	7	June 3–24	1338 (49.3)	298 (11.0)
1958	Kamiina	1250 (197*)	2	June 24	848 (67.8)	338 (27.0)
1959	Kamiina	1160 (136.2*)	2	June 20	525 (45·3)	108 (9.3)

Note: Figures in parentheses indicate the average number of galls (*), that of chestnut gall wasps, and that of parasitic wasps per 100 grams, respectively.

the emergents of the parasitic and the host gall wasp, respectively, and the species of the former was identified. Each of the parasitic wasps and the host wasp could still emerge as good as even from the gall which was dried up to the extent at which it diminished to about 1/5 in weight, unless the galls are plucked in too early a season. Sometimes they bored an exit hole through a paper bag and flew away from it. The gall-racks were, therefore, confined to a small room with a door closed. When counts were taken, the exit-holes on a paper bag were papered and all the escapers were recovered. According to the classification by districts and by years, the total numbers of the emergents of the two were presented in table I.

(3) The Species of Indigenous Parasitic Wasps and their Relative Ratio in individual Numbers

The species of the parasites that issued from the chestnut galls collected at various localities in Ina district from 1956 till 1959 have been identified as follows:

Eupelmus urozonus Dalman, } : Family Eupelmidae,
Peleumus ferrierei Yasumatsu,* }
Torymus elegantulus Yasumatsu,*◎ }
Megastigmus spp.,*◎ } : Family Torymidae,
M. maculipennis Yasumatsu,* }
Ormyrus nigritibialis Yasumatsu,◎ } : Family Ormyridae,
O. flavitibialis Yasumatsu,* }
Amblymerus amoenus japonicus Yasumatsu, : Family Pteromalidae,
Eurytoma rosae Nees,◎ } : Family Eurytomidae,
Eudecatoma spp.* }
Diptera spp.

The 10 species other than Dipterous species are the already-recorded, belonging to what Dr. Yasumatsu calls the 11 effective parasitic wasps on the chestnut gall wasp. Of these, the 4 species, marked ◎, belong to the 5 most effective species, and the 6 species asterisked are new to Ina district. These 11 species are grouped into 5 families belonging to the superfamily Chalcidoidea and 1 Dipterous parasite whose specific name is not yet identified. Of the 11 effective parasitic wasps, the species that are not yet found in this district is only one species, viz., *Torymus beneficus* Yasumatsu.

The relative ratios in individual numbers for these species could not be compared for all the data obtained. The following results, presented in table II, will serve as a representative sufficient to give us the general picture in question.

As can be seen, the relative percentage for each species varies comparatively remarkably according to years and/or districts. But that for *Megastigmus* spp. is overwhelmingly large, ranking first in every case; *Ormyrus nigritibialis* ranks second, and the third place varies according to years, being occupied either by *Amblymerus amoenus japonicus, Eurytoma rosae,* or *Peleumus ferrierei,* respectively. *Torymus elegantulus* occupies the 3rd place in Kamiina in 1958. *O. flavitibialis,* though very few in number, was found for the first time at Kamiina in 1959. It is to be noticed that higher ranks are in most cases occupied by such species as *Megastigmus* spp., *Ormyrus nigritibialis*

TABLE II. Relative percentage of the parasitic wasps emerging from the chestnut galls collected in Ina district, the southern part of Nagano Prefecture.

Year	1957		1958	1959
District	Kamiina (A)	Shimoina	Kamiina (A)	Kamiina (B)
Megastigmus spp.	51.0	61.7	49.7	19.5
Ormyrus nigritibialis	15.8	9.1	16.9	32.3
Amblymerus amoenus japonicus	10.7	5.0	5.6	15.0
Eurytoma rosae	8.2	7.4	8.3	10.5
Diptera spp.	6.7	5.0	0.3	6.8
Peleumus ferrierei	3.6	8.1	7.4	6.8
Torymus elegantulus	2.0	3.0	8.9	0
Ormyrus flavitibialis	0	0	0	7.5
Eupelmus urozonus	2.0	0.7	2.9	1.6
Total	100.0	100.0	100.0	100.0

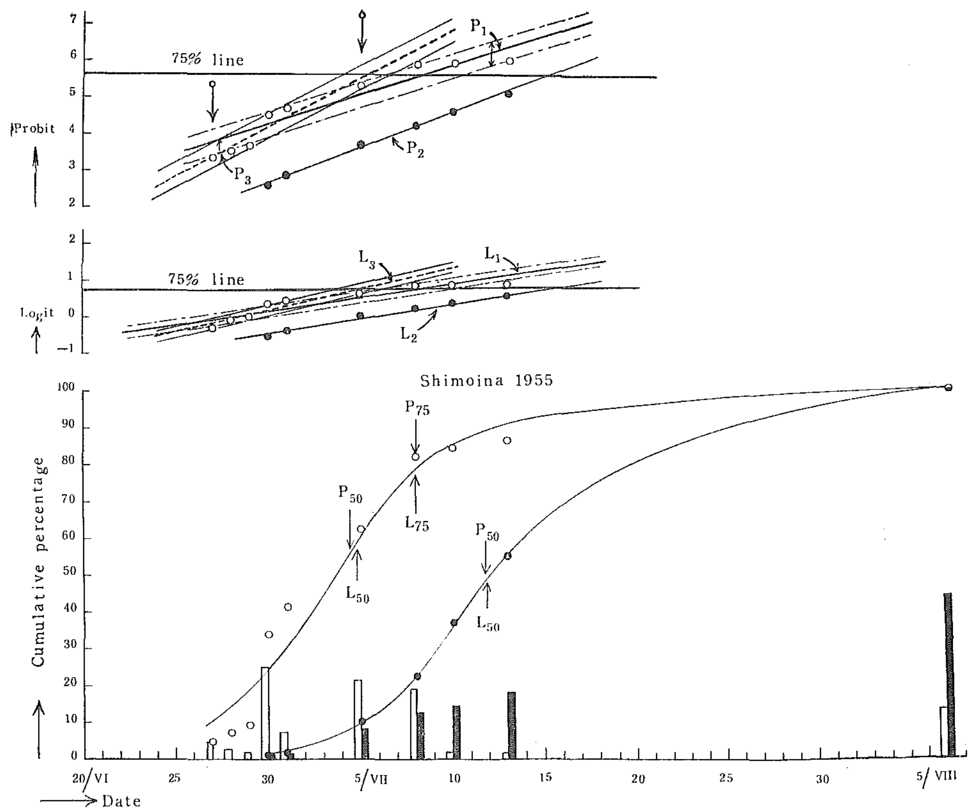
Note: These values can be regarded to represent roughly the relative population density of each species at each area under natural conditions.

and *Eurytoma rosae*. What arrests our attention particularly is that *Megastigmus* spp. and *O. nigritibialis* are in most cases overwhelmingly large in relative population density in Ina district. If these species are regarded as powerful natural enemies specially adapted to this district, the utilization and positive preservation of these species will make a great contribution to the biological control of the chestnut gall wasp in Ina district, as emphasized by Smith (1941).

(4) Interrelationship between the Eemergence of the Host Gall Wasp and that of its Parasitic Wasps

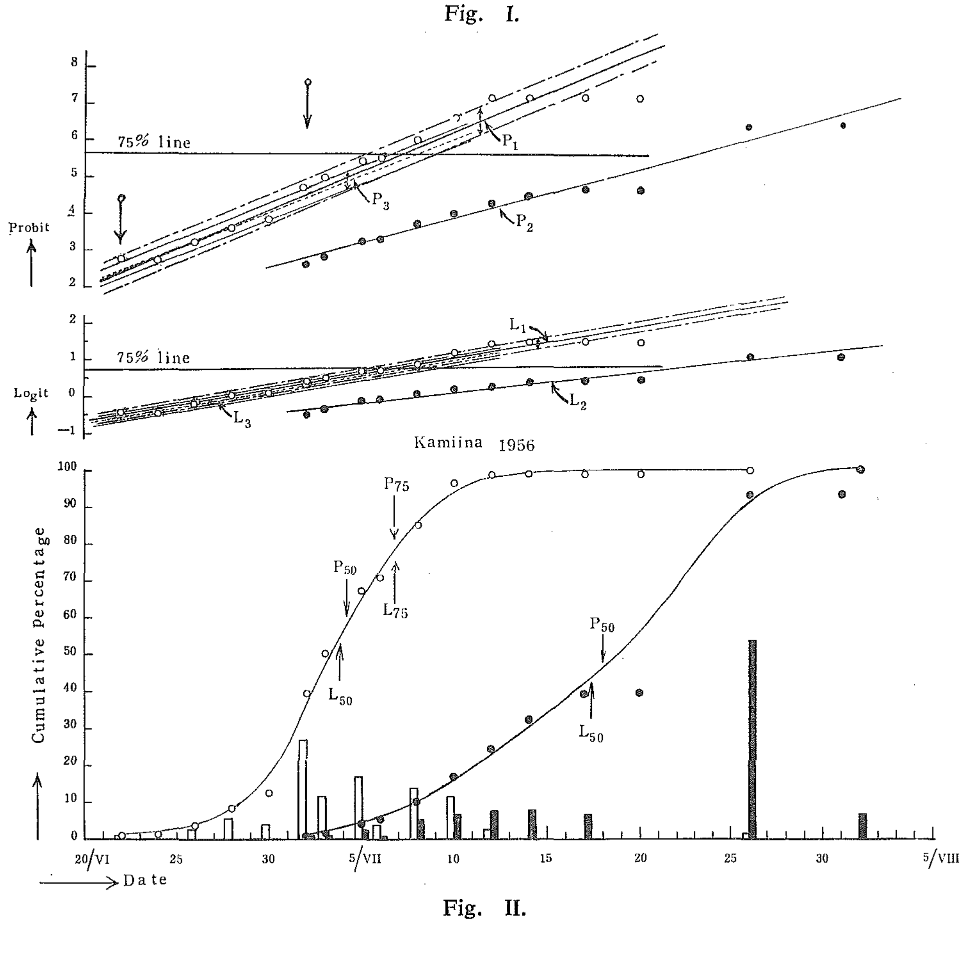
a) Biostatistical method of analysis

The emergent insects from the chestnut galls were divided into two groups, viz., the chestnut gall wasp and a composite group of its parasitic wasps. For these two groups, the cumulative percentage emergence was calculated separately. The marks ○ and ● in Figs. I-VII denote such percentages plotted against the census date; the circles corresponding to composite parasitic wasps and the black circles to the host gall wasp, respectively. Similarly, the percentage emergence for the two was indicated as histogrammatic diagrams □ and ■ in the same figures, respectively. As is obviously seen, the dots denoting the cumulative percentage emergence for each group tend to have a definite locus, which a free-hand sigmoidal curve drawn through their positions will approximately define. The latter diagram for each group shows also an approximately symmetrical polygon with the maximum percentage emergence as centre. These facts may be viewed in different ways. Let us suppose that the latter polygon stands for a a bell-shaped normal curve. Then the former sigmoidal curve is an integrated normal curve. If the former sigmoidal curve is assumed to be a logistic curve, then the latter polygon must be regarded as a curve given by the first-order derivative of the logistic curve, which is different in its mathematical characteristic from a normal curve, being more flat-topped and symmetrical in shape. Thus



Probit
75% line
P1
P2
P3
Logit
75% line
L1
L2
L3
Shimoina 1955
Cumulative percentage
P75
L75
P50
L50
20/VI
25
30
5/VII
10
15
20
25
30
5/VIII
Date

Fig. I.



Probit
75% line
P1
P2
P3
Logit
75% line
L1
L2
L3
Kamiina 1956
Cumulative Percentage
P75
L75
P50
L50
20/VI
25
30
5/VII
10
15
20
25
30
5/VIII
Date

Fig. II.

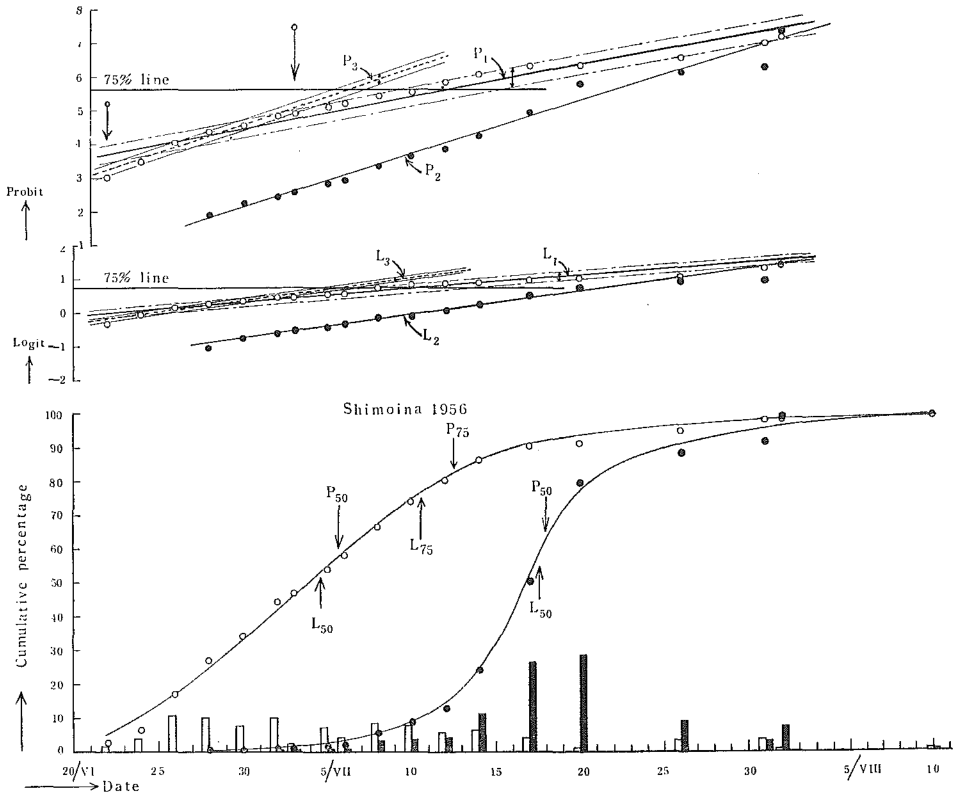


Fig. III.

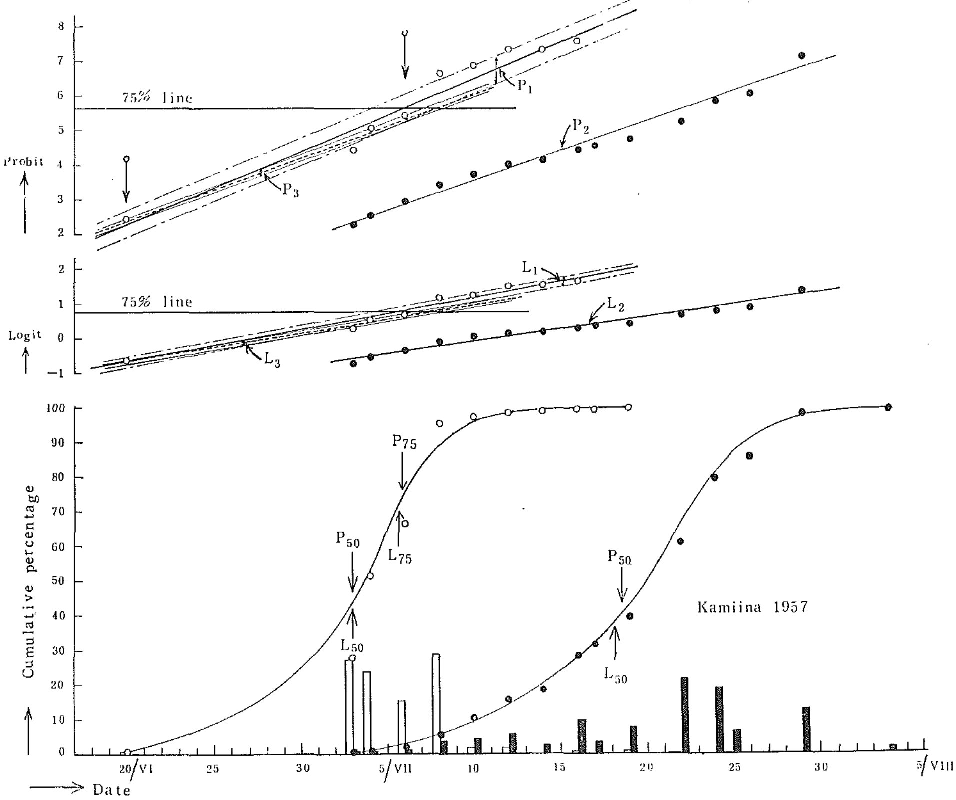
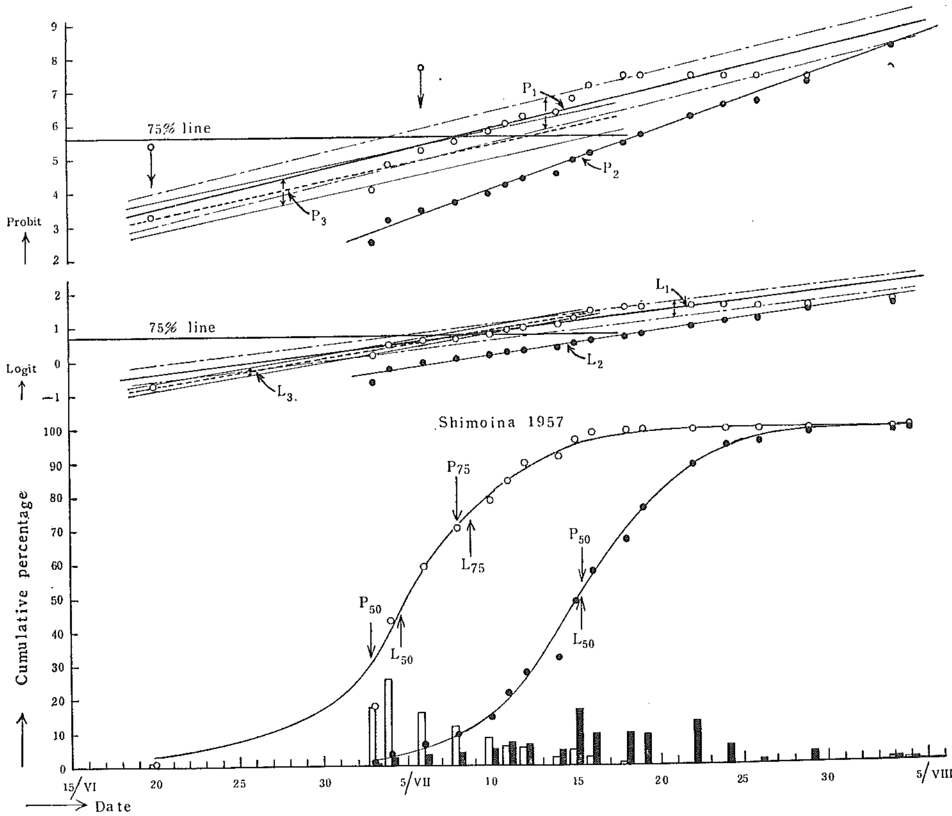
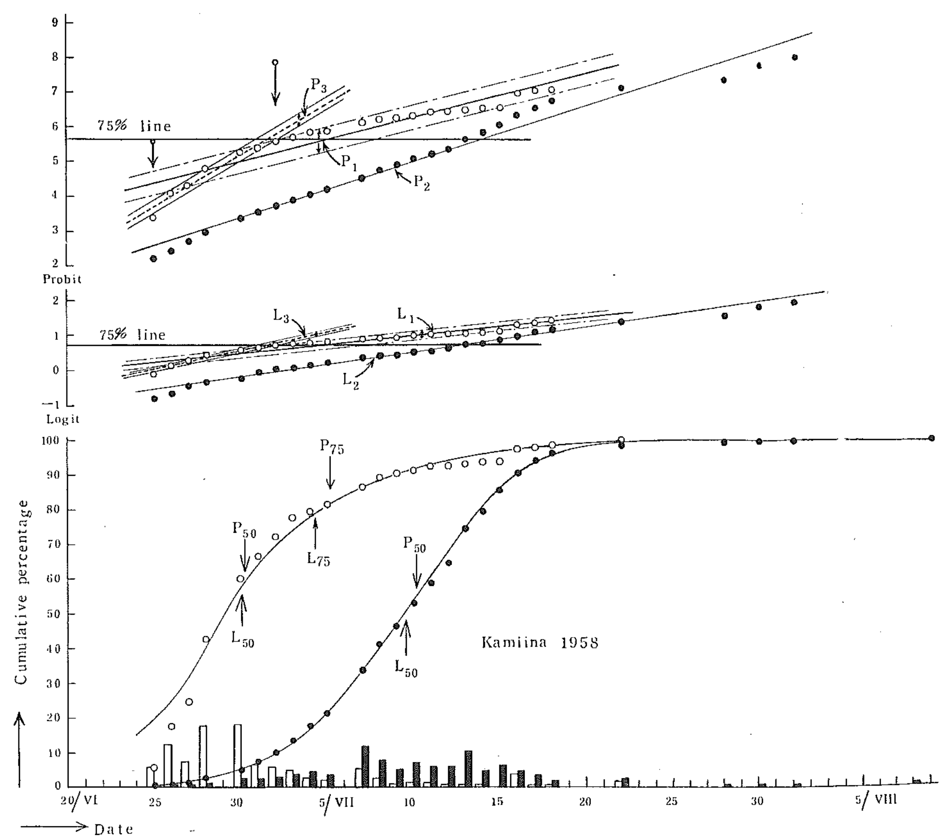


Fig. IV.



Probit
75% line
P_1
P_2
P_3
Logit
L_1
L_2
L_3
Shimoina 1957
Cumulative percentage
P_{75}
L_{75}
P_{50}
L_{50}
15/VI
20
25
30
5/VII
10
15
20
25
30
5/VIII
Date

Fig. V.



Probit
75% line
P_1
P_2
P_3
Logit
L_1
L_2
L_3
Kamiina 1958
Cumulative percentage
P_{75}
L_{75}
P_{50}
L_{50}
20/VI
25
30
5/VII
10
15
20
25
30
5/VIII
Date

Fig. VI.

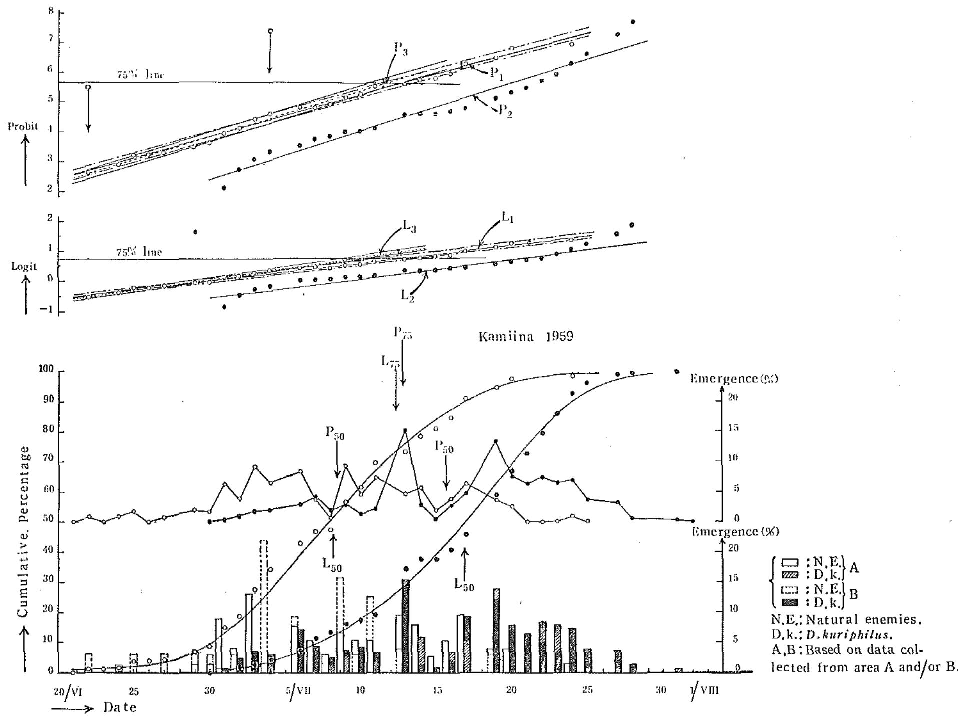


Fig. I–VII. The cumulative percentage emergence for the chestnut gall wasp and that for its composite parasitic wasps, and the logit as well as the probit line of least squares.

○ : Cumulative percentage emergence of the composite parasitic wasps.
● : Cumulative percentage emergence of the chestnut gall wasp.
▭ : Percentage emergence of the composite parasitic wasps.
▬ : Percentage emergence of the chestnut gall wasp.
○⟶ : Indicatiag the opening and the closing date employed for the calculation of the predictive logit and/or probit line of least squares.
(solid lines with arrows) : Showing the upper and the lower limit for the logit and/or the probit line of least squares.
(dashed lines with arrows) : Showing the upper and the lower limit for the predictive logit and/or probit line of least squares.
L_{50} and P_{50}, L_{75} and P_{75} : Indicating the relation between the percentage emergence given by the free-hand logistic curve and the 50% and/or 75% emergence estimated from the logit and/or the probit line of least squares.
L_1, L_2, L_3 and P_1, P_2, P_3 : See the text.

the further analysis is entirely conditioned on how we view the above two divergent versions.

i) *Probit method*

Now, let us hold the former version, that is, the version to the effect that the locus of the percentage emergence lies closely to a normal curve, and consequently that of the cumulative percentage emergence closely to an integrated normal curve.

Then we can transform the values of the cumulative percentage emergence into probits, as is usual with a dosage-mortality curve in bioassey. In this case, the equation

$$p=\frac{1}{\sqrt{2\pi}\,\sigma}\int_{-\infty}^{x} e^{-(x-m)^2/2\sigma^2}\,dx \quad \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots(1)$$

is the basis for the analysis; p expresses the cumulative percentage emergence, x the time in days measured from any date as origin, and m the abscissa corresponding to the peak of the curve $(\sqrt{2\pi}\,\sigma)^{-1}\exp\{-(x-m)\}^2/2\sigma^2$. Let the variable x of the above formula be transformed as follows:

$$y=(x-m)/\sigma^2+5. \quad \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots(2)$$

Then we have

$$p=\frac{1}{\sqrt{2\pi}\,\sigma}\int_{-\infty}^{y-5} e^{-\frac{1}{2}y^2}\,dy, \quad \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots(3)$$

which gives us probits y corresponding to the observed values of p. As is shown by the formula (2), the relation between the probits y and the variable x is linear. Hence, it is clear that this functional relation gives us a clue as to whether or not the cumulative percentage emergence curve takes substantially the shape of an integrated normal curve.

ii) *Logit method*

Let the cumulative percentage emergence p be expressed by the logistic curve

$$p=[1+\exp\{-r(x-m)\}]^{-1} \quad \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots(4)$$

where m is the value of x corresponding to $p=\frac{1}{2}$, or to the point of inflexion of the curve. Then we have its derivative of the first order

$$dp/dx=[1+\exp\{-r(x-m)\}]^{-2}r\exp\{-r(x-m)\}, \quad \ldots\ldots\ldots\ldots(5)$$

as a curve corresponding to the percentage emergence curve. The value of m denotes the abscissa against the peak of the curve (5). Now, let us put

$$l=\frac{1}{2}r(x-m), \quad \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots(6)$$

and then from the formula (4), we have easily

$$l=\frac{1}{2}ln\,p/(1-p), \quad \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots(7)$$

where l is termed logit in the sense of a logistic unit. Obviously, there is a linear relation between logits l and the variable x, as is indicated in the formula (6).

Thus, by this functional relation as a clue, we can determine whether or not the cumulative percentage emergence curve conforms to the logistic curve. This is the basis for the logit analysis. It is noted in this connection that there is no essential change in the theory, when common logarithm is employed in the formula (7) instead of natural logarithm. In this report, the formula $l'=\frac{1}{2}\{\log p/(1-p)+1\}$ is adopted as logit transformation according to J. Berkson (1950), which is essentially the same as that given in (7).

TABLE III. Line of least squares for the cumulative percentage emergence transformed into probits and/or logits.

Year	1955	1956	
District	Shimoina	Kamiina (A)	Shimoina
P_1	$y=0.1620x+3.6018\pm0.3744$ (N=9)	$y=0.1949x+2.4943\pm0.4174$ (N=15)	$y=0.0965x+3.6042\pm0.2857$ (N=18)
P_2	$y=0.1932x+2.4971\pm0.2139$ (N=9)	$y=0.1332x+2.6918\pm0.2736$ (N=12)	$y=0.1565x+1.6738\pm0.2577$ (N=15)
P_3	$y=0.2675x+3.1400\pm0.3151$ (N=6)	$y=0.1920x+2.3480\pm0.2223$ (N= 6)	$y=0.1733x+2.9941\pm0.1575$ (N= 7)
L_1	$l=0.0730x-0.1400\pm0.1498$ (N=9)	$l=0.0808x-0.5358\pm0.2327$ (N=15)	$l=0.0387x-0.0597\pm0.1612$ (N=18)
L_2	$l=0.0822x-0.5395\pm0.1378$ (N=9)	$l=0.0533x-0.4222\pm0.1157$ (N=12)	$l=0.0684x-0.9658\pm0.1297$ (N=15)
L_3	$l=0.1139x-0.2827\pm0.1553$ (N=6)	$l=0.0861x-0.6184\pm0.1437$ (N= 6)	$l=0.0696x-0.2888\pm0.0798$ (N= 7)
Starting date of reckoning	June 26	June 21 for P_1, P_3, L_1, L_3; July 1 for P_2, L_2.	June 21 for P_1, P_3, L_1, L_3; June 27 for P_2, L_2.

Year	1957		1958
District	Kamiina (A)	Shimoina	Kamiina (A)
P_1	$y=0.2096x+2.0854\pm0.4045$ (N=18)	$y=0.1163x+3.3642\pm0.4903$ (N=10)	$y=0.1263x+4.2354\pm0.3658$ (N=18)
P_2	$y=0.1669x+2.1939\pm0.1884$ (N=14)	$y=0.1772x+2.5980\pm0.1387$ (N=14)	$y=0.1614x+2.4192\pm0.2735$ (N=17)
P_3	$y=0.1814x+2.1938\pm0.0852$ (N= 4)	$y=0.1074x+3.0903\pm0.4453$ (N= 4)	$y=0.2953x+3.3892\pm0.1904$ (N=10)
L_1	$l=0.0925x-0.8042\pm0.1437$ (N=10)	$l=0.0578x-0.4374\pm0.2222$ (N=10)	$l=0.0571x-0.1702\pm0.1152$ (N=18)
L_2	$l=0.0698x-0.6701\pm0.1127$ (N= 4)	$l=0.0687x-0.4388\pm0.0743$ (N=14)	$l=0.0697x-0.6095\pm0.1362$ (N=17)
L_3	$l=0.0804x-0.7447\pm0.0441$ (N= 4)	$l=0.0801x-0.8291\pm0.1156$ (N= 4)	$l=0.1089x-0.0985\pm0.0785$ (N= 7)
Starting date of reckoning	June 19 for P_1, P_3, L_1, L_3; July 2 for P_2, L_2.		June 24

Year	1959
District	Kamiina (B)
P_1	$y=0.1442x+2.5201\pm0.1255$ (N=24)
P_2	$y=0.1714x+2.3289\pm0.3066$ (N=24)
P_3	$y=0.1517x+2.4441\pm0.1396$ (N=10)
L_1	$l=0.0571x-0.4845\pm0.0580$ (N=24)
L_2	$l=0.0622x-0.5318\pm0.1500$ (N=24)
L_3	$l=0.0680x-0.5681\pm0.0516$ (N=10)
Starting date of reckoning	June 22

Note: P_1; L_1: Probit and/or logit line of least squares for parasitic wasps.

P_2; L_2: Probit and/or logit line of least squares for the chestnut gall wasp (Host).

P_3; L_3: Probit and/or logit line of least squares for prediction use. (for parasitic wasps).

N: Number of dots plotted against the x-axis.

b) Problem of testing goodness of fit

In the foregoing analyses, it should be born in mind that the linear relation between x and probit y and/or logit l does not signify the usual regression of y and/or l on x and *vice versa*, but merely a mathematically functional relation, since x, the time in days measured from any date as origin, cannot be assumed to be a normal variate, while both y and l are assumed to be such. Consequently, it would be quite meaningless in this case to test goodness of fit to line by the usual method of testing hypothesis, even if such a test were made formally. The case is similar in the other kinds of tests of significance, which are applicable to the sampling variation of the parameters pertaining to the relation between two normal variates. This is, of course, limited to the present case alone.

(5) **Analysis of Data**

a) Comparison of validity between the logit and the probit method

The linear relation between x and probit y and/or logit l can be expressed by the line of least squares. The formulae for such lines are tabulated in table III, the graphs founded upon such formulae being drawn in Figs. I-VII, respectively.
In these graphs, the lines labeled "P" and/or "L" denote the line of least squares corresponding to probits and/or logits, respectively; the small figures 1 and 2 affixed to "P" and/or "L" denote the line for composite parasitic wasps and/or the chestnut gall wasp, respectively. With regard to another figure 3, an explanation will be given later.

As presented in graphs, every dot for probit and/or logit lies approximately close to the assigned line of least squares to which it belongs. This holds, of course, good for every case of composite parasitic wasps and/or the chestnut gall wasp. From this, it may be said that, from the standpoint of eye observation alone, each of the cumulative percentage emergence curves for the two can be fitted substantially well by the integrated normal curve and likewise by the logistic curve; in other words, that the percentage emergence curve for the two can be expressed well by the normal curve and likewise by the curve given by the first-order derivative of the logistic curve. Scrutinous inspection reveals, however, the fact that in every case the logit line of least squares shows a better fit to the observed dots. To demonstrate this, the total sum of squares of deviations from the line, viz., $S(Y-y)^2$ and/or $S(L-l)^2$, was calculated, respectively. The results are indicated in table IV.
As expected, all the values of $S(L-l)^2$ are remarkably smaller than those of $S(Y-y)^2$, irrespective of years and districts. This warrants the propriety of the above graphical impression.

The values of $S(L-l)^2$ for the composite parasitic wasps do not, in most cases, amount to about 1/6 of those of $S(Y-y)^2$. The same values for the chestnut gall wasp also constitute about 1/2.4 or less of those of $S(Y-y)^2$. This implies that the cumulative percentage emergence curves are better fitted by the logistic curve in every case. Accordingly, it comes to this conclusion that in every case the percentage emergence curve is better expressed by the curve given by the first-order derivative of the logistic curve than done by the normal curve.

TABLE IV. Comparison among the sums of squares of errors of estimate by the probit and/or logit line of least squares.

Classification	Year	District	Probit line of least squares: $S(Y-y)^2$	Probit: Coefficient of line of least squares (b_i)	Probit: Correlation coefficient (r_i)	Logit line of least squares: $S(L-l)^2$	Logit: Coefficient of line of least squares (b_i)	Logit: Correlation coefficient (r_i)	$\frac{S(Y-y)^2}{S(L-l)^2}$
Composite parasitic wasps	1955	Shimoina	0.9817	0.1620 (0.2675)	0.948 (0.937)	0.1570	0.0730 (0.1139)	0.949 (0.918)	6.25
	1956	Kamiina	2.2644	0.1949 (0.1920)	0.971 (0.964)	0.3377	0.0808 (0.0861)	0.975 (0.929)	6.71
		Shimoina	1.3061	0.0965 (0.1733)	0.949 (0.980)	0.1827	0.0387 (0.0696)	0.977 (0.969)	7.15
	1957	Kamiina	1.3088	0.2096 (0.1814)	0.975 (0.986)	0.1651	0.0925 (0.0804)	0.984 (0.994)	7.93
		Shimoina	3.8463	0.1163 (0.1074)	0.933 (0.907)	0.7899	0.0578 (0.0801)	0.943 (0.987)	4.87
	1958	Kamiina	2.6759	0.1263 (0.2953)	0.931 (0.976)	0.2654	0.0517 (0.1089)	0.957 (0.970)	10.08
	1959	Kamiina	0.3466	0.1442 (0.1559)	0.995 (0.978)	0.0739	0.0571 (0.0213)	0.994 (0.985)	4.69
Chestnut gall wasp	1955	Shimoina	0.1830	0.1932	0.998	0.0760	0.0822	0.957	2.41
	1956	Kamiina	0.7485	0.1332	0.978	0.1338	0.0533	0.976	5.59
		Shimoina	0.8633	0.1565	0.990	0.2185	0.0684	0.986	3.95
	1957	Kamiina	0.4263	0.1669	0.991	0.1525	0.0698	0.983	2.80
		Shimoina	0.2888	0.1772	0.996	0.0828	0.0687	0.993	3.49
	1958	Kamiina	1.7959	0.1614	0.987	0.4449	0.0697	0.983	4.04
	1959	Kamiina	2.0688	0.1714	0.979	0.5016	0.0622	0.961	4.12

Note: Figures in parentheses indicate the values concerning the predictive probit and/or logit line of least squares.

Another noteworthy fact is that almost all the values of $S(Y-y)^2$ for composite parasitic wasps are far larger than those for the chestnut gall wasp. The former is about 1.5 to 6 times as large as the latter even in the same year, its ratio to the latter sometimes coming up to more than 10, for instance, to about 13 in 1957 at Shimoina. This fact implies that the integrated normal curve fits, if anything, well the cumulative percentage emergence curve for the host gall wasp rather than for its composite parasitic wasps. Needless to say, somewhat similar tendency can be seen in some cases of the logit line of least squares. In this case, however, the values of $S(L-l)^2$ for the composite parasitic wasps are nearly the same as, or smaller than, those for the chestnut gall wasp, excepting a very few unusual results. This is a positive evidence that the cumulative percentage emergence curves for the two are substantially better fitted by the logistic curves.

b) Biological considerations

It was found that the probit transformation yielded a better fit to the cumulative percentage emergence for the host gall wasp in almost all the cases tested. How can this fact be explained biologically? As previously mentioned, the premise for probit transformation lies in the assumption that the percentage emergence curve follows a normal curve. Generally speaking, it would not be illogical to assume that, when a population of the same species of insects is treated, the time variation in their percentage emergence depends principally upon individual fluctuations in their emergence, or in their growth rate. If that is the case, it will also be accepted that the percentage emergence curve concerned takes a form of a normal curve with the period of maximum emergence as centre, since the host gall wasp under investigation is undoubtedly assumed to constitute the same monospecific population. This is a biological answer to the above question. On the contrary, when a group of insects are composed of different species, just as in the case of the present composite parasitic wasps, it would be almost impossible to express the composite percentage emergence curve for the group in one normal curve, because it is quite natural that each species of component parts has its own emergence curve characterized by its unique peak and the range of variation peculiar to it. In such a case, the peak of the composite curve will be determined by that of the principal curve for a numerically predominant species. This principal curve may, of course, be normal in its shape. The other normal curves for the remaining component species with a small population will be overlapped with one another before and behind the peak of the principal curve, according to their emergence period. Naturally, such an integrated curve tends to have some symmetrical form other than a normal curve. The curve most suitable for such an integrated symmetrical curve may be the first-order derivative of a logistic curve which is generally accepted to be fit for diversified biological phenomena. The reason why the logit line of least squares shows a better fit to the observed logits for the composite parasitic wasps may thus be explained clearly from both the statistical and the biological standpoint. Quite similarly, an explanation will be given of the reason why the values of $S(Y-y)^2$ for the composite parasitic wasps are far larger than those for their host wasp. For the sake of better understanding of this version postulated, the schematic illustration is given analytically in Fig. VIII, and the actual result in 1957 is shown in Fig. IX. As can be seen in Fig. IX, the position of the peak of the period of emergence for each component species of the group of parasitic wasps in 1957 differs apparently from one another. This cannot be taken to have resulted from chance variation; it should be regarded as *prima facie* inductive evidence for the propriety of the deductive illustration given in Fig. VIII.

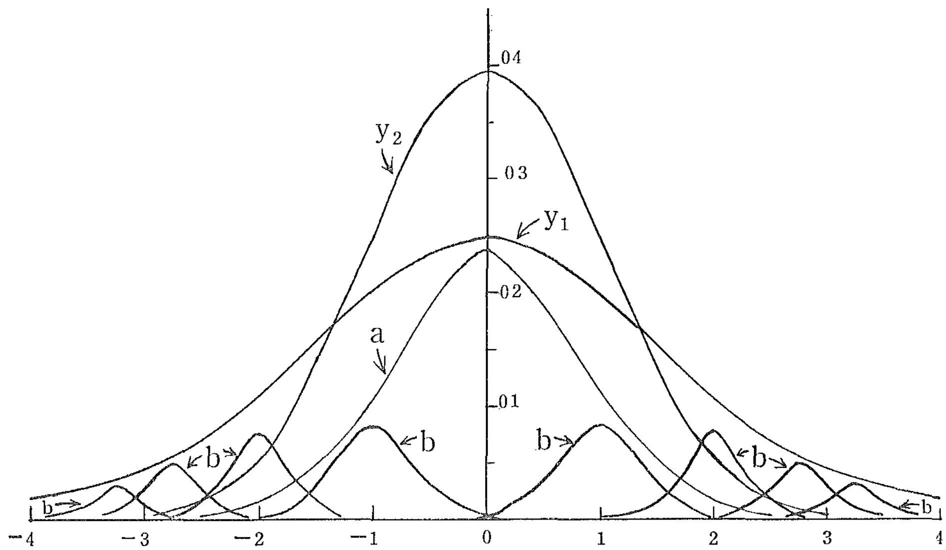


Fig. VIII. Schematic representation of how the symmetrical curve dy/dx, where y is a symmetrical logistic curve, is constructed with its component normal curves.
$y=[1+\exp(-x)]^{-1}$: A standard type of a symmetrical logistic curve.
$y_1=dy/dx=\exp(-x)[1+\exp(-x)]^{-2}$.
$y'_2=(\sqrt{2\pi})^{-1}\exp(-x^2/2)$: A standard type of a normal curve.
b: Component normal curves, the resultant of which constitutes the curve dy/dx.

Another question to be raised is how the following, rather contradictory fact should be interpreted biologically; namely, the fact that all the values of $S(L-l)^2$ for the chestnut gall wasp are considerably small as compared with those of $S(Y-y)^2$ for that. This fact implies that the percentage emergence curve for the chestnut gall wasp is also fitted better by the curve given by the first-order derivative of a logistic curve than is done by the normal curve assigned; in other words, that its emergence curve *should be considered to be the integration of some component normal curves* with different peaks and ranges of variation. By way of illustration on this point, two sets of emergence curves for the chestnut gall wasp are depicted. Each set consists of two curves concerning two areas which are fairly wide apart. One set of curves represent the data obtained at Kumashiro area and Tomikusa area in Shimoina in 1957, the distance between the two areas being about 25 km. The other set of curves stand for the data obtained at two areas in Kamiina, namely, at Tagiri area and at Suwagata area, respectively, the distance between the two areas being

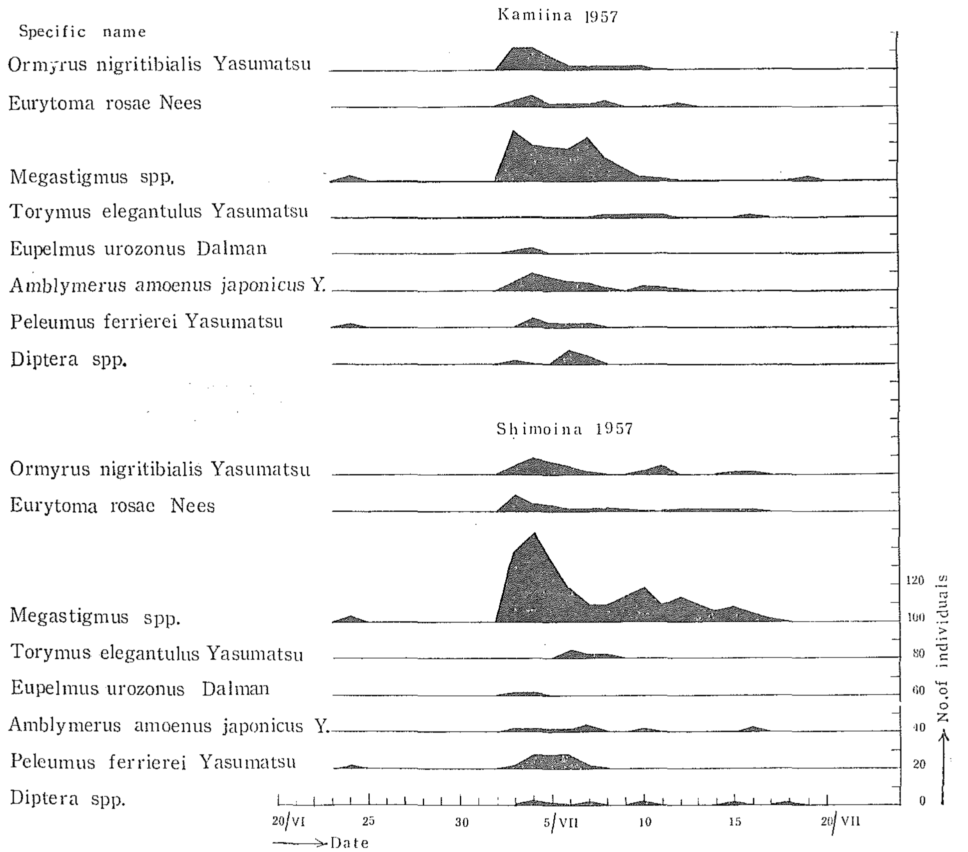


Fig. IX. Difference in the position of the peak of the emergence curve for each component species of the composite group of the parasitic wasps that issued from the chestnut galls collected at 2 different localities in Kamiina District and/or 7 localities in Shimoina District, respectively, in 1957.

about 13 km. The results are shown in Fig. X. As expected, the interval between the two peaks of the emergence curves for the two areas belonging to the same district is, roughly speaking, about 4-5 days in time. There seems to be no substantial difference in the range of variation between the two curves in a set. In the case of the chestnut gall wasp, too, there is undoubtedly a considerable difference in the maximum period of emergence between the two groups inhabiting different localities. Probably, the growth rate of a gall insect, such as a chestnut gall wasp, is liable to be influenced considerably by the conditions of the microhabitat concerned, that is to say, by the state of growth of its host plant as well as the other microenvironmental factors thereat. Then, it is quite natural that the state of its emergence varies considerably as the microhabitat concerned shifts. Some positive evidences for this biological interpretation may be produced. One is the number and size of galls formed on one twig; the other is the number of individuals of larvae found within one gall. With regard to the former, table V will offer an adequate evidence. The demonstration of the latter will be given in table VI (A, B, C, and D). As can be seen in table V,

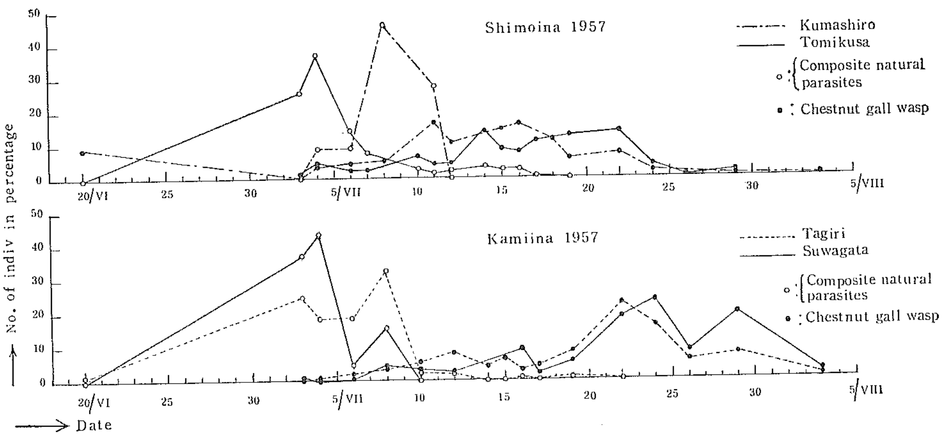


Fig. X. Showing the interval between the two peaks of emergence curves for the chestnut gall wasp as well as that for its natural parasites issuing from the chestnut galls collected at 2 different areas in the same district, viz., Tagiri and Suwagata in Kamiina District, or Kumashiro and Tomikusa in Shimoina District, respectively, in 1957.

TABLE V. Average number of the chestnut galls formed on a unit twig about 1 cm. thick.

Year		1955	1956	1957	1958	1959
District	Localities					
Shimoina	Neba	50.0*	—			
	Chisato (A)	35.0*	1.5	20.2 (120)		
	Seinaiji	32.8	—	—		
	Chisato (B)	29.9	20.6	35.9 (135)		
	Namiai (A)	19.4*	—	—		
	Namiai (B)	16.4	—	—		
	Tomikusa	—	46.3 (180)	—		
	Yamamoto	—	59.0	—		
	Shimojo	—	73.6*	—		
	Iida (A)	5.1	23.0 (203)	54.5 (120)		
	Goka	5.1	—	—		
	Iida (B)	—	59.5*	—		
	Kamisato (A)	—	53.0 (217)	25.5 (130)		
	Oojima	—	7.0	—		
Kamiina	Kamikatagiri	—	41.0 (218)	—		—
	Tagiri	—	11.3 (201)	38.6 (220)	79.0 (193)	—
	Suwagata (A)	—	25.3 (216)	43.2 (180)	—	—
	Suwagata (B)	—	9.5 (187)	—	—	—
	Inokubo	—	15.0 (213)	—	—	—
	Habiro (A)	—	20.0 (219)	—	—	—
	Habiro (B)	—	13.0 (173)	—	—	—
	Minamiminowa (A)	—		—	—	37.3 (136)
	Minamiminowa (B)	—		—	—	27.8 (137)

Note: 1) 20–30 unit twigs were sampled at each locality.
2) Sampling dates: May 17–June 24.
3) Figures in parentheses indicate the number of galls per 100 grams.
4) Asterisks denote the locality where some parasitic natnral enemies were liberated.

TABLE VI. A. Number of the chestnut gall wasp larvae per gall in the galls collected at diversified localities (From the results secured in 1956).

District	Kamiina (Coll. on June 10–13)					Shimoina (Coll. on June 4–11)					
Area	Suwagata (A)	Habiro	Matsukawa	Kami-katagiri	Suwagata (B)	Iida (A)	Kamisato (A)	Tomikusa	Kamisato (B)	Goka	Toyooka
No. of larvae (x)	Frequency					Frequency					
1	10	8	4	5	1	9	15	7	7	3	0
2	8	5	9	3	1	10	77	5	4	2	5
3	1	7	5	7	5	1	9	4	4	4	3
4	3		1	3	10		2	1	2	6	2
5			1	2	1			3	0	2	3
6					0				2	2	2
7					2				1	1	1
8											4
Total	22	20	20	20	20	20	103	20	20	20	20
Mean	1.86	1.95	2.30	2.70	3.85	1.60	1.98	2.40	2.70	3.60	4.65
No. of galls per 100 grams	216	201	211	218	187	203	217	180	189	120	134
Sum of x (locality total)	41	39	46	54	77	32	204	48	54	72	93

Note: It can be seen that the greater the number of galls per 100 grams becomes, or the smaller in size the galls become, the smaller the number of larvae per gall tends to become. This tendency can be confirmed by the analyses of variance shown in tables VI. B, C, and D.

TABLE VI. B. Analysis of variance of the numbers of the chestnut gall wasp larvae per gall in the galls collected at 5 and/or 6 different localities.

a) Kamiina District

Source of variation	D. F.	Sum of squares	Mean square
Between means	4	52.9699	13.2425**
Within localities	97	126.4909	1.3040
Total	101	179.4608	

$F_{0\,97}^{\,4}=13.2425/1.3040=10.155>F_{97}^{4}(\alpha=0.01)\doteqdot3.52$

b) Shimoina District

Source of variation	D. F.	Sum of squares	Mean square
Between means	5	161.5392	32.3078**
Within localities	197	297.1111	1.5082
Total	202	458.6503	

$F_{0\,197}^{\,5}=32.3078/1.5082=21.421\gg F_{197}^{5}(\alpha=0.01)\doteqdot3.11$

TABLE VI. C. Analysis of variance of the chestnut gall wasp larvae per gall in the galls collected at 4 and/or 5 different localities.

a′) Kamiina District

Source of variation	D. F.	Sum of squares	Mean square
Between means	3	40.9	13.6333**
Within localities	76	103.9	1.3671
Total	79	144.8	

$F_{0\,76}^{\,3}=13.6333/1.3671=9.972>F_{76}^{3}(\alpha=0.01)\doteqdot4.06$

b′) Shimoina District

Source of variation	D. F.	Sum of squares	Mean square
Between means	4	109.84	27.46**
Within localities	95	265.15	2.7911
Total	99	374.99	

$F_{0\,95}^{\,4}=27.46/2.7911=9.838>F_{95}^{4}(\alpha=0.01)\doteqdot3.52$

TABLE VI. D. Significant difierence D between two-locality totals at the 5 per cent. level of significance.

For a′ (Kamiina District).

$$|D|\geqq\{2k\times V_E\times F_{76}^{3}(\alpha=0.05)\}^{1/2}=(40\times1.3671\times2.728)^{1/2}\doteqdot12.213,$$

where $k=20$, $V_E=1.3671$, and $F_{16}^{3}(\alpha=0.05)\doteqdot2.728$.

For b′ (Shimoina District).

$$|D|\geqq\{2k\times V_E\times F_{95}^{4}(\alpha=0.05)\}^{1/2}=(40\times2.7911\times2.465)^{1/2}\doteqdot16.588,$$

where $k=20$, $V_E=2.7911$, and $F^{4}{}_{5}\doteqdot2.465$.

the number of galls per twig (about 1 cm. thick) differs apparently from one another according to localities, irrespective of years and districts. Generally speaking, the areas where numerically abundant galls are recorded tend to be characterized by a relatively small number of galls per 100 grams, and naturally by a comparatively large gall in size. On the other hand, the newly infested areas such as the localities in the northern part of Ina District, viz., Kamiina, are generally characterized both by a small number of galls per twig and by a small gall in size as compared with the old infested areas such as those in the southern part of Ina District, viz., Shimoina.

The number of larvae within a gall varies remarkably according to localities, in general, as shown in table VI. A. The tendency to this is particularly evinced in the results at neighbouring localities in the same area. The results at locality A and B in the same area are the instance in point. Analyses of variance (Table VI. B, & C) and the test of significance for the difference between two locality totals (Table VI. D) clearly verify the generality of the above findings.

From every point of view, it may safely be concluded that the growth of a chestnut gall wasp is much affected by the microhabitat conditions concerned, as the natural result of which the period of its emergence undergoes more or less change in the position of its peak as well as in the range of its variation. The second problem with which we are confronted has thus come to a satisfactory solution.

It should be noted in this connection that the tendency similar to this can be seen in the emergence periods for the composite parasitic wasps which issued respectively from the galls collected at two different localities adjacent to each other, as illustrated in Fig. X. For this the same version as in the case of their host gall wasp may hold true, because the life of such parasitic wasps is considered to be entirely dependent upon that of their host gall wasp.

In summary, it may be stated that the cumulative percentage emergence curve is fitted better by the logistic curve than by the integrated normal curve for both the chestnut gall wasp and the composite group of its parasitic wasps, and that the background for it can be adduced from both the biostatistical and purely biological standpoint. It has also been pointed out that whether this version holds true or not can be determined by the extent of the value of $S(L-l)^2$, viz., the sum of squares of deviations from the logit line of least squares assigned to the data.

On the basis of this version, a smooth curve, which is presented in Figs. I-VII, respectively, has been drawn by eye through the dots for each cumulative percentage emergence, in due consideration of the trend in their deviations from the logit line of least squares concerned. Naturally, the shape of each curve is characteristic of logistic curves. In the present case, however, the integrated normal curve, if drawn, would also have the similar pictorial appearance as the logistic curve, having been so close as to be almost superposable.

(6) Yearly Fluctuations in the Trend of Emergence

a) Constancy of yearly trend of emergence

The general trend of emergence is evinced in the shape of a cumulative percentage emergence curve. The yearly change in it, if any, can naturally be grasped by comparison of the shape of each curve. To be more accurate, it can be compared by the gradient of the logit line of least squares; in other words, by the value of the coefficient of x in the formula for the logit line. As is indicated in table IV, such coefficients are very uniform in value. The coefficient, b_i, of the logit line, the "logit coefficient" for short, is all very small and substantially very uniform in value which converges for the most part toward the range $+0.07\sim+0.08$, irrespective of years, localities, the host gall wasp or the composite parasitic wasps. Needless to say, the tendency to this is clearly indicated in the gradient of the logit line shown in Figs. I-VII. The similar relation to this holds true for the values of the probit coefficient, b_i, though they are about twice as large as those of the former, ranging from $+0.10$ to $+0.21$. It is true that such coefficients are all very small in value, each being less than $+0.21$, but undoubtedly they differ substantially from 0. The reason is that each of the logit lines or of the probit lines can never be regarded as parallel to the x axis. As discussed previously, all the tests of significance applicable to the relation between two normal variates, such as the test of uniformity of regression coefficients and the tests of significance of regression coefficients, would be all meaningless in this case, even if such a test were made formally. Nevertheless, such two sorts of tests were made for trial, but in every case the level of significance concerned came up to 0.999 or over, the illogical values far beyond the expected range. This is the very fact indicative of irrationality of making such a kind of test in this case. It is to be noted in this connection, however, that the value of the correlation coefficient, r_i, between x and l and/or y, which has been calculated quite formally, is very close to unity in every case, suggesting that the change in the value of l and/or y is substantially in direct proportion to that in the value of x.

Thus the upshot is as follows: The values of r and/or $1/\sigma^2$, which are the coefficient of x in the formula (5) and/or (2), respectively, are positive numbers significantly greater than zero, and substantialy equal to one another, respectively, in every case. In other words, it follows that all the logistic curves shown in Figs. I-VII (or all the integrated normal curves, though not drawn), are substantially uniform in shape with one another. From this, it may be concluded more concretely that, when taken as a whole, fluctuations in the trend of yearly emergence for the chestnut gall wasp and/or the composite group of its parasitic wasps are substantially uniform in type, respectively, regardless of years and districts.

b) Constancy of Emergence Period

From the form of the free-hand logistic curve as well as the gradient of the logit line in Figs. I-VII, the theoretical starting point of each curve may be inferred by eye observation, since there is almost no sharp break in the region near the actual starting point in each curve, though the utmost precaution should always be exercised in such extrapolation. For the composite parasitic wasps, the date of the first appearance of the emergents has been estimated at about 20th June every year. For the chestnut gall wasp, it has been estimated at about 1st July, about 10-13 days later than that for the former. These are common both to Kamiina and

Shimoina District. About 99 per cent of the component species of the parasitic wasps complete their emergence by about 20th July. Of them, a very few species such as *Megastigmus* spp., *Ormyrus flavitibialis* and *Eurytoma rosae* continue to emerge until the middle of August rather intermittently, though such emergents are very few in individual number. In this respect, too, almost no difference can be seen in substance between Kamiina and Shimoina District. A large majority of chestnut gall wasps terminate their emergence by about 5th August at the latest, about 10-15 days later than the date of the final emergence of the parasite group. These interrelationships are kept constant, irrespective of years and district. From this it follows that in Ina district the period of emergence for the chestnut gall wasp is characterized by about ten-day or so time lag when compared with that for the composite group of its parasitic wasps.

As will be discussed in Chap. III, the parasitic wasps on this host wasp seem to have at least two generations a year. Moreover, the period of their emergence covers about one month ranging from about 20th June till about 20th July. It will, therefore, be favorable a great deal for them to complete their emergence about ten days or so earlier than their single-brooded host completes its emergence. Because, possibly such a mode of parasitism will afford them ample chances for adjusting themselves to the growth season of the plants on which their second-generation host galls are to be formed. Judging from the fact that the parasitic wasps are native to this district and had presumably been parasitic on various kinds of insect galls before the chestnut gall wasp invaded here, such a mode of parasitism may originally be fitted for them, and accordingly it may probably not be an adaptation newly acquired by them as a result of modification of their hosts. No reports on the similar observation to this have ever been published, so far as I am aware.

(7) The Determination and the Prediction of the Period fittest for the biological Control of the Chestnut Gall Wasp

a) How to determine the period fittest for the biological control

From the angle of biological control, it is hoped to keep the population density of the native natural parasitic enemies (wasps) at the maximum level, and at the same time to check the population density of the host pest-wasp (chestnut gall wasp) at the minimum level by some adequate economic measures. Whether such an economic measure can be adopted or not depends entirely upon whether the period fittest for such a step can be found out or not. Such a period, if any, may be called the "period most suited for biological control in the wide sense". The term "biological control" here employed is understood, according to DeBach, P. (1958), as the following: that is, as "the utilization of organism for the economic control of populations of animals and weeds". In the present paper, importance is particularly attached to the preservation and propagation of resident natural enemies rather than to those of introduced natural enemies. This is contrasted with the usual definition that "biological control invokes the aid of the natural enemies, either parasite or predator, which are bred artificially in large number and introduced into the country or district of the pest to be controlled" (Smith, K. S., 1951). This may be called the biological control in the narrow sense. Herein lies the reason why the biological control here considered may be called so in the wide sense.

Now, what I call the period fittest for biological control may roughly be inferred by inspection into the interrelation between the cumulative percentage emergence curve for the host gall wasp and that for its composite parasitic wasps. The procedure is as follows: Let's slide a rule perpendicularly along the x axis to and fro. Then, find out the position where the rule cuts the cumulative percentage emergence curve for the two at the same time at such a ratio as to cut the curve for the host wasp at its starting slope corresponding to about 10 per cent or less of the whole and the curve for composite parasitic wasps at the part corresponding to about 70-80 per cent of the whole. This method may not be applied, however, to the prediction of such a period in general, because each cumulative curve can not be drawn completely until the emergence is over.

I have attempted to formulate the above cursory inference, and succeeded in prescribing the method of prediction of the period in question. The necessary procedure for this formulation is as follows: (1) To transform the values, p, of the cumulative percentage emergence for both the composite parasitic wasps and the host gall wasp into logits, respectively. (2) To find graphically the date corresponding to any aimed-at cumulative percentage emergence, say, 75%, for the composite parasitic wasps with the logit ordinate as clue. For the necessary values of logits, see the column attached to the bottom of table VII. The date is of course fixed on the nearest integer. The date thus determined is called the logit 75% date and so forth. (3) To find the value of the cumulative percentage emergence for the host gall wasp on such a logit date determined. (4) To compare the cumulative number of the composite parasitic emergents with that of the emergents of the host gall wasps on such a fixed logit date.

With regard to the procedure necessary for prediction, details will be entered in the next section.

Now, to such a logit date, two sorts of values of the cumulative percentage emergence will correspond; one is that inferred from the curve postulated, and the other the actual value attained on that date. Needless to say, these two kinds of values are, in most cases, somewhat discrepant from the aimed-at values, e.g., 70%, 75% etc. The value inferable from the curve will be called the value attained, and the actual value attained on that date the observed value, respectively, for the sake of contrast with the aimed-at value. The above steps are, of course, applicable quite in like manner to the probit date.

In regard to the logit and/or probit date, several attained values are tabulated in table VII in comparison with those aimed-at. Whether or not the aimed-at date is suited for the period most fit for biological control can firstly be judged by both the relative relation between the cumulative percentage for the composite parasitic wasps and that for the host gall wasp on that date, and secondly by the goodness of agreement between the aimed-at percentage and that attained. As can be seen in the table, the higher the aimed-at percentage becomes, the better the agreement between it and the attained percentage tends to become, while the inequalities in the percentages

TABLE VII. Logit and/or probit date estimated from the cumulative number of the emergent natural parasites that issued from the chestnut galls collected in Ina district.

(a) Logit date

Year	1955	1956		1957		1958	1959
District	Shimoina	Kamiina	Shimoina	Kamiina	Shimoina	Kamiina	Kamiina
70% date attained	July 7	July 6	July 9	July 5	July 8	July 3	July 12
Attained % of natural parasites	73	72	70	63	70	73	72
Attained % of chestnut gall wasp	18	5	7	2	9	12	25
75% date attained	July 8	July 7	July 11	July 6	July 9	July 4	July 12
Attained % of natural parasites	79	79	77	74	75	77	72
Attained % of chestnut gall wasp	21	7	12	2	12	16	25
80% date attained	July 9	July 7	July 12	July 6	July 10	July 6	July 14
Attained % of natural parasites	83	79	80	74	30	83	80
Attained % of chestnut gall wasp	28	7	14	2	15	26	34
85% date attained	July 10	July 8	July 14	July 7	July 11	July 7	July 15
Attained % of natural parasites	86	85	86	83	84	86	84
Attained % of chestnut gall wasp	37	10	25	3	20	34	39
90% date attained	July 11	July 9	July 16	July 8	July 13	July 10	July 17
Attained % of natural parasites	88	90	89.5	90	90	91	90
Attained % of chestuut gall wasp	44	13	41	45	32	53	50
95% date attained	July 13	July 11	July 20	July 10	July 15	July 13	July 19
Attained % of natural parasites	91	96	93	96	95	94	43
Attained % of chestnut gall wasp	55	20	78	8	49	73	56

(b) Probit date

Year	1955	1956		1957		1958	1959
District	Shimoina	Kamiina	Shimoina	Kamiina	Shimoina	Kamiina	Kamiina
70% date attained	July 8	July 7	July 11	July 5	July 7	July 4	July 12
Attained % of natural parasites	79	79	77	63	65	77	72
Attained % of chestnut gall wasp	21	7	12	2	7	16	25
75% date attained	July 8	July 7	July 13	July 6	July 8	July 5	July 13
Attained % of natural parasites	79	79	84	74	71	81	76
Attained % of chestnut gall wasp	21	7	19	2	9	21	29
80% date attained	July 9	July 8	July 14	July 7	July 10	July 7	July 14
Attained % of natural parasites	83	85	86	83	80	86	80
Attained % of chestnut gall wasp	28	10	24	3	15	33	34
85% date attained	July 11	July 9	July 17	July 8	July 11	July 8	July 15
Attained % of natural parasites	88	90	90	90	84	89	84
Attained % of chestnut gall wasp	44	13	51	4.5	20	40	39
90% date attained	July 12	July 10	July 20	July 9	July 13	July 10	July 17
Attained % of natural parasites	90	94	93	94	90	91	90
Attained % ofchestnut gall wasp	50	17	78	6	32	53	50
95% date attained	July 14	July 12	July 23	July 11	July 16	July 13	July 20
Attained % of natural parasites	92	97	95	97	96	94	96
Attained % of chestnut gall wasp	61	23.5	86	11	56	73	69

Note: Values of logits and of probits.

Per cent	Logit	Probit
50	0.50	5.00
70	0.68	5.52
75	0.74	5.67
80	0.80	5.84
85	0.88	6.04
90	0.98	6.28
95	1.14	6.65

for the host gall wasp become more conspicuous as the aimed-at percentage becomes higher. Taking account of these facts together with the parasite-host interrelationship in individual number, the ligt 75% date will be sufficient for our purpose. By the way, the cumulative percentage emergence on any probit date is in general larger by 5-6 per cent than the aimed-at percentage. From the angle of the conservation of the composite parasitic wasps alone, therefore, the inference drawn from the probit date may be said to be more useful for our purpose. But, if it is taken into consideration the fact that the percentages for the host wasp on any date are far higher, such a view should be abandoned. Especially, it must be taken into account that the emergent host wasp is remarkably large in number as compared with the parasitic wasps emerging from its gall. Thus, if one should choose such a date as adequate time for biological control, one would be too late to expect satisfactory results. In this respect, too, the inferiority in validity of inference drawn from the probit transformation has clearly been revealed.

Another prerequisite for securing the reliability of the inference concerned is that the attained percentages are to be kept substantially constant, irrespective of years and districts. On inspection of the table, the average value of the said percentage for th host gall wasp may roughly be estimated at about 10%, and that for the composite parasitic wasps at about 77%. Whether these values are substantially constant or not, however, needs to be tested stochastically. To carry out this test, the following operations were performed according to M. Masuyama (1953, p. 196): Put

$$r=2p-1\,, \qquad (1)$$

where p is the attained percentage in question. And we get

$$\frac{1}{2}ln\frac{p}{1-p}=\frac{1}{2}ln\frac{1+r}{1-r}=Z\,, \qquad (2)$$

where Z is what is called R. A. Fisher's Z-transformation, which follows approximately the normal distribution. This is the basis for the test. Now, let $\bar{p}$ be the population mean or the hypothetical estimate of p_i. Let Z be the value of $\hat{Z}$ corresponding to $\bar{p}$. Then, the unbiased estimate of variance of Z_i will be given by

$$u^2=\Sigma(Z_i-\bar{Z})^2/(\mathrm{N}-1)\,, \qquad (3)$$

where $\bar{Z}$ is $\Sigma Z_i/\mathrm{N}$, and N is the number of p_i, i.e., 6, in this case. Thus, the significance of the disparity in value between p_i and $\bar{p}$ can be tested by F-test, viz.,

$$\left.\begin{array}{l} F=\mathrm{N}(\bar{Z}-\hat{Z})^2/u^2 \\ df_1=1, \quad df_2=\mathrm{N}-1 \end{array}\right\} \qquad (4)$$

Now, the value of $\bar{p}$ is assumed as 0.77 (77%) for the composite parasitic wasps, and as 0.1 (10%) for the host wasp. The result is given in table VIII, which warrants the

TABLE VIII. Test of uniformity of the attained percentages for the composite parasitic wasps and/or the chestnut gall wasp on the logit 75% date.

	$\bar{p}$	$\hat{z}$	$\bar{z}$	F	$d.f.$	α
Composite parasitic wasp	0.77	0.6044	0.5827	10.7838	5,1	$0.20<\alpha<0.25$
Chestnut gall wasp	0.10	−1.0983	−1.0316	7.3140	5,1	$0.25<\alpha<0.50$

adequacy of the hypothesis under test. From this, it may safely be said that on the logit 75% date we shall be able to expect the average value of the cumulative percentage emergence to be 77% for the composite parasitic wasps, and to be 10% for the host gall wasp, regardless of years and districts, so far as the present data are concerned. Actual numbers of individuals of the cumulative emergents on the logit 75% date were calculated from these hypothetical values, 77% and 10%, of the cumulative percentage emergence, and they were given in table IX, a. It should be noted here that the logit 75% dates converge on the days in the beginning of July every year, ranging from 4th to 12th July with 8th July as centre, although the dates show more or less a tendency to move to the latter half of the term at the old infested areas.

The relative relation between the attained and the observed cumulative percentages of the emergents on the logit 75% date can be grasped from the results given in table IX, b. Each value in a pair (A_1, A_2 ; B_1, B_2) agrees practically well with each other. This also is another evidence that the logit transformation is very reliable. It is worthy of note in this connection that the majority of the values both of the attained and the observed for the host gall wasp are comparatively larger than the aimed-at values indicated in table IX, a, while those for the composite parasitic wasps approximate practically to the aimed-at percentages. Such a relation seems to result from the numerical abundance of the host gall wasp. If that is the case, it will be better to fix the period fittest for biological control earlier by 1 or 2 days than that estimated in such a region where the population density of the host gall wasp is comparatively high.

The percentage of the number of the composite parasitic wasps (A_0) to that of the host gall wasp (B_0) needs to be noticed. If some adequate control measures be applied to the host gall wasp on and after the logit 75% date, and if the remainder of the emergents, viz., 90%, of the host gall wasp, which is expected to emerge after that date, be killed entirely, the ratio of A_0 to B_0 will become as large as that presented in column 6 of table IX, a. When compared with the ratio $A_1 : B_1$ for the attained percentage and with the ratio $A_2 : B_2$ for the observed percentage, both of which are presented in table IX, b, the ratio $A_0 : B_0$ nearly equals in general trend to both of them, though the same ratio for Kamiina in 1959 differs remarkably from those for the former two, which is ascribable mainly to an unknown sudden decrease in the number of emergent parasitic wasps on 6-7th July (cf. Fig. VII.).

Now, let e_1, and e_2 be average numbers of eggs laid by a female of the composite parasitic wasps and/or the host wasp, respectively. Then biotic potential for a female of the former and/or the latter will be $e_1/2$ and e_2, respectively, on the assumption of the sex ratio for the former to be 50 to 50, since the host gall wasp is parthenogenetic. As will be discussed later, the parasitic wasps, if not all, seem undoubtedly to be double-brooded at least. The number of eggs deposited by their adult females at the end of the second brood, therefore, will become

$$N_1 e_1^2 (1 - W/100)/4 , \qquad \ldots\ldots (1)$$

where N_1 is the total number of their adults at the end of the first brood, W being the total environmental resistance during the second brood. The number of eggs laid by the host gall wasp at that time will be

TABLE IX. Cumulative number of the emergents on the logit 75% date.

(a) Cumulative number of the emergents calculated from the hypothetical cumulative percentage emergence, viz., 77% for composite parasitic wasps and 10% for the chestnut gall wasp.

Year	1955	1956		1957		1958	1959
District	Shimoina	Kamiina	Shimoina	Kamiina	Shiomina	Kamiina	Kamiina
Logit 75% date	July 8	July 7	July 11	July 6	July 9	July 4	July 12
Composite natural parasites ($A_0=T_p\times 0.77$)	84	61	701	159	230	260	83
Chestnut gall wasp ($B_0=T_w\times 0.10$)	231	12	780	100	134	85	53
$A_0 : B_0$ (%)	36.4	508.3	89.9	159.0	171·6	305.9	156.6

Note: T_p and T_w denote the total number of the composite natural parasites and that of the chestnut gall wasp, respectively.

(b) The attained and the observed cumulative number of the emergents.

Year		1955	1956		1957		1958	1959
District		Shimoina	Kamiina	Shimoina	Kamiina	Shiomina	Kamiina	Kamiina
Composite parasitic wasp	Attained (A_1)	85	62	701	152	224	260	78
	Observed (A_2)	89	67	730	137	235	269	81
Chestnut gall wasp	Attained (B_1)	484	8	936	20	161	136	132
	Observed (B_2)	526	12	993	19	192	151	148
Composite parasitic wasps : Chestnut gall wasp (%)	Attained ($A_1 : B_1$)	17.6	787.5	74.9	760.1	139·1	191.2	59.1
	Observed ($A_2 : B_2$)	16.9	558.3	70.6	721.1	122.4	178.1	54.8

Note: Figures underlined denote the values observed one day after the logit 75% date.

$$N_2e_2, \quad \ldots\ldots (2)$$

where N_2 is the total number of its adults at the end of its emergence. Then, on the assumption of perfect monoparasitism, the percentage of parasitism of the composite parasitic wasps to the host gall wasp will be

$$N_1e_1^2(1-W/100)/(4N_2e_2). \quad \ldots\ldots (3)$$

The ratio $N_1 : N_2$ will be given by the results shown in table X.

TABLE X. Total number of individuals emerging from sample galls and the ratio of composite parasitic wasps to chestnut gall wasp as viewed from the total.

Year	1955	1956		1957		1958	1959
District	Shimoina	Kamiina	Shimoina	Kamiina	Shimoina	Kamiina	Kamiina
Composite parasitic wasps (c)	108	79	910	206	298	338	108
Chestnut gall wasp (d)	2306	117	7797	1002	1338	848	525
c : d (%)	4.7	67.5	11.7	20.6	22.3	39.9	20.6
c : (c+d) (%) (Assumed percentage of parasitism)	4.5	40.3	10.5	17.1	18.2	28.5	17.1

Note: As regards the ratio c:(c+d) see the text.

Namely, $N_1 : N_2$ 12 : 100 for the minimum, roughly,
and $N_1 : N_2$ 40 : 100 for the maximum, roughly.

In this estimation, such rather unusual data as those in 1955 and in Kamiina in 1956 were excluded.

Now, the assumed percentage of parasitism at the time when the two groups completed their emergence will be given, from (2), by

$$3e_1^2(1-W/100)/(100e_2) \quad \text{for the minimum,} \ldots\ldots (4)$$

and

$$e_1^2(1-W/100)/(10e_2) \quad \text{for the maximum,} \ldots\ldots (5)$$

respectively. From table IX, a, the ratio $N_1 : N_2$ on the logit 75% date will roughly be estimated at 90 : 100 for the minimum and at 306 : 100 for the maximum, respectively. Consequently, on the assumption that the control made on and after this logit date is perfect, the assumed percentage of parasitism at the end of the second brood for the composite parasitic wasps will be

$$9e_1^2(1-W/100)/(40e_2) \quad \text{for the minimum,} \ldots\ldots (6)$$

and

$$163e_1^2(1-W/100)/(200e_2) \quad \text{for the maximum,} \ldots\ldots (7)$$

respectively. Hence, the ratio of (6) to (4) and/or (7) to (5) will be

$$9/40 : 3/100 = 7.5 : 1$$

and

$$153/200 : 1/10 = 7.7 : 1,$$

respectively. From this, it follows that, in case of success in such control, the assumed percentage of parasitism in question will be increased up to 7.5-fold at the minimum and 7.7-fold at the maximum.

By the way, if the parasite-host interrelationship concerned is assumed to be perfectly monoparasitic, the percentage of parasitism in question may be inferred from the ratio of the total number (c) of the composite parasitic wasps to the sum total (c+d) of the total number of the composite parasitic wasps plus the total number (d) of the host gall wasp. These assumed percentages, presented in table X, may be taken to be indicative of the possibility that the percentage of parasitism concerned varies rather remarkably from 10.5% to 28.5% under natural conditions. From these assumed percentages multiplied by 7.5 and/or 7.7, the hypothetical percentages of parasitism in case of success in the control made on and after the logit 75% date have been calculated and are given in the following table.

Year	1955	1956		1957		1958	1959
District	Shimoina	Kamiina	Shimoina	Kamiina	Shimoina	Kamiina	Kamiina
Min.	(33.8)	(302.3)	78.8	128.3	136.5	213.8	128.3
Max.	(34.7)	(310.3)	80.9	131.7	140.1	219.5	131.7

(Note: Figures in parentheses denote rather unusual results.)

From this it may be suggested that it is not necessarily impossible to increase the percentage of parasitism concerned up to nearly 100 per cent or more in the very year when the control based upon the logit 75% date is successfully performed. It goes without saying that this suggestion is based on purely hypothetical ground. Nevertheless, it will safely be stated that there is an ample hope for success in checking the emergence of the host gall wasp at the lowest possible level, if only the control made on and after the logit 75% date produces satisfactory results. Probably, if utmost efforts are made at the control of the chestnut gall wasp in this way for two or three years consecutively, the damage by this pest will surely be checked to such an extent as no further control measures are needed.

To sum up, the period fittest for the biological control of the chestnut gall wasp in Ina district may be determined by the logit 75% date for the composite group of its parasitic wasps, the actual dates of which ranging from 4th July to 12th July with 8th July as centre. It is not necessarily impossible to increase the percentage of parasitism of the composite parastic wasps to the host gall wasp up to about 7.5 times as much as that under natural conditions, and to such an extent as high as 100 per cent or more in the very year when the control based on the logit 75% date is performed in an effective way in substance.

b) How to predict the period fittest for biological control

i) *Logit 75% date estimated from the data in the early stage of emergence*

If the logit 75% date is already known as a result of 1 or 2 years' observations made acording to the project above-mentioned, the date empirically induced may surely be available for the prediction of the period fittest for biological control in the year yet to come. But there is much fear that such an empirically fixed date tends to undergo more or less fluctuations according as the macro- and the micro-environmental conditions in that year vary, since it is nothing but a precedent founded upon past results. It is desired, therefore, to find out a more reliable method of prediction, which is not solely based upon the data secured in the preceding year, but upon the data obtained in that year. Such a method, if any, will naturally lead to a semi-empirical formulation. Hence, it has been tested how the logit 75% data will be useful when it is estimated from a few initial data in the early stage of emergence of the composite parasitic wasps. The procedure of operation is essentially the same as that previously performed with total data. What is to be added is only the estimation of the maximum number of the cumulative emergents, viz., the total number of the emergents of the composite parasitic wasps, to the details of which I shall refer in the next section.

For the sake of avoiding complexity, the principle of test being essentially the same, the test which follows will be made on the assumption that the cumulative percentage emergence in question does not mean the original record, but the estimate calculated with the estimated total number of the emergents as base.

The formulae, L_3, given in table III, represent the logit line of least squares calculated from the data in the early stage of emergence of the composite parasitic wasps. Together with these, the formulae, P_3, for the probit line of least squares concerned are given in the same table, for comparison. These lines will be called the predictive logit, or probit, lines of least squares, otherwise called the predictive logit, or probit, lines for short; and so will quite similarly the dates concerned be called the predictive logit, or probit, 75% dates, etc. The graphical representation of these lines is given in Figs. I-VII. In fixing the term for prediction use, the standard has been settled not to pass 3rd July, viz., the date 5 days prior to the logit, or probit, 75% date already fixed as the time fittest for the biological control, though the term has been extended in some cases up to 6th July on account of scarcity of data obtained in the early stage of emergence concerned. The size, n, of the sample involved in each term varies according to years and districts, counting 4, 6, 7 and 10, as presented in both the figures and table.

As can be seen in Figs. I-VII, predictive logit, or probit, lines, L_3 or P_3, lie in most cases practically close to the original logit, or probit, lines, L_1 or P_1. If the measure of deviation, $S_E=\pm\sqrt{S(E-y)/(N-2)}$, where E denotes the logits, or the probits, determined by the predictive lines, y representing both determined by the whole data, is taken into account, the band $L_1 \pm S_{E1}$, or $P_1 \pm S_{E1}$, can be substantially superposed by the band $L_3 \pm S_{E3}$, or $P_3 \pm S_{E3}$, with regard to the result for Kamiina in 1956 and 1959 and for both Kamiina and Shimoina in 1957, respectively. This must be an indication that the prediction founded upon the band $L_3 \pm S_{E3}$ is substantially equal in precision to that based on the band $L_1 \pm S_{E1}$. In the other cases, the gradient of the band $L_3 \pm S_{E3}$ is slightly steeper than that of the band $L_1 \pm S_{E1}$. As the result, the junction of the band $L_3 \pm S_{E3}$ and the horizontal line falling on the logit ordinate, 0.74, corresponding to logit 75%, is situated at the portion a bit nearer to the origin as compared with the junction of the same horizontal line and the band $L_1 \pm S_{E1}$. Quite a similar relation to this can also be seen between the band $P_3 \pm S_{E3}$ and that $P_1 \pm S_{E1}$. This tendency is rather favorable for the object of prediction, because there is almost no fear of being too late to carry out necessary control, in such a case as the date thus predicted is utilized.

TABLE XI. Predictive logit 75% date estimated from the composite natural parasites that issued from the chestnut galls.

Year	District	Limit	Logit line of least squares: Date	Logit: Cumulative percentage emergence: Composite natural parasites	Logit: Cumulative percentage emergence: Chestnut gall wasp	Probit line of least squares: Date	Probit: Cumulative percentage emergence: Composite natural parasites	Probit: Cumulative percentage emergence: Chestnut gall wasp	Term and no. of plots employed as base
1955	Shimoina	Upper limit	July 4	54	8	July 4	54	8	June 27–July 4
			(July 8)	(79)	(21)	(July 8)	(79)	(21)	
		Lower limit	July 7	73	17	July 7	73	17	6
1956	Kamiina	Upper limit	July 6	72	5	July 7	79	7	June 22–July 2
			(July 7)	(79)	(7)	(July 7)	(79)	(7)	
		Lower limit	July 9	90	13	July 9	90	13	7
	Shimoina	Upper limit	July 5	55	2	July 6	58	3	June 22–July 3
			(July 11)	(77)	(12)	(July 13)	(84)	(19)	
		Lower limit	July 7	63	4	July 7	63	4	7
1957	Kamiina	Upper limit	July 7	84	3	July 7	83	3	July 20–July 6
			(July 6)	(74)	(2)	(July 6)	(74)	(2)	
		Lower limit	July 8	89	5	July 8	89	5	4
	Shimoina	Upper limit	July 7	65	7	July 8	71	9	June 20–July 6
			(July 9)	(75)	(12)	(July 8)	(71)	(9)	
		Lower limit	July 10	80	15	July 16	91	57	4
1958	Kamiina	Upper limit	July 1	63.5	7	July 1	63.5	7	June 25–July 2
			(July 4)	(77)	(16)	(July 5)	(81)	(21)	
		Lower limit	July 3	73	12	July 2	68.5	9	
1959	Kamiina	Upper limit	July 9	56	12	July 11	67	21	June 22–July 4
			(July 12)	(72)	(25)	(July 13)	(76)	(29)	
		Lower limit	July 11	67	21	July 13	76	29	10

Note: Figures in parentheses indicate the original logit and/or probit 75% date and the attained percentage on each date.

The numerical data predicted on the basis of the band $L_3 \pm S_{E3}$, or the band $P_3 \pm S_{E3}$, are given in table XI. When compared with those founded on the band $L_1 \pm S_{E1}$ and/or the band $P_1 \pm S_{E1}$, respectively, no wide discrepancy can be seen between the two predictive informations on the whole. If examined more minutely, however, it will be found out that the predictive logit 75% dates approach more closely, in every case, to the original logit 75% dates than the predictive probit 75% dates do. This tendency can be seen also in the fact that the values of the coefficients of x in the formulae of the predictive logit lines approximate more closely to those in the formulae of the original logit lines than those in the formulae of the predictive probit lines do. Thus, in the predictive informations, too, evidence is clear that the logit transformation is superior to the probit transformation.

According to the numerical informations here presented, it will be found that the period fittest for the biological control of the chestnut gall wasp lies on the whole close either to the dates corresponding to the upper limit $L_3 + S_{E3}$, and/or the lower limit $L_3 - S_{E3}$, of the predictive logit 75% date, or to the period put between the two limits.

ii) *How to estimate the maximum value of the cumulative number of the emergents*

The difficulty encountered in the actual application of the above predictive logit method lies in the fact that the number of the emergents should be transformed into percentage. This resolves itself down to the question of how the maximum value of the cumulative number of the emergents shall be estimated. The case is, of course, quite alike in the application of the probit analysis. The difficulty which we face can never be solved, so far as the probit analysis is employed. But, if the logit method is utilized, it will easily be solved. This is one of the terms decisively advantageous to the application of the logit analysis.

Let the maximum value in question be G. And then the logistic curve concerned will be

$$p(x) = G/[1 + \exp\{-r(x-m)\}.$$

From this, the following linear difference equation with a definite difference h is given:

$$1/p(x-h) = \exp(-rh)/p(x) + \{1 - \exp(-rh)\}/G\,. \quad \ldots\ldots\ldots\ldots (1)$$

Accordingly, plotting the finite differences diagram $[1/p(x),\ 1/p(x+h)]$, following M. Masuyama (1948, 1950. See also T. Torii, 1956, 1959.), we can estimate the value of $1/G$ at the ordinate of the point of intersection of the line (1) and a bisector of the coordinate angle at the origin.

As an example of this operation, the data for the composite parasitic wasps in Shimoina in 1956 and in Kamiina in 1958 have been utilized. The initial 8 values of the cumulative number of the emergents in the former case and likewise the 7 values in the latter have been employed, respectively. From the finite differences diagram, shown in Fig. XI, the value of G for the former was estimated at 893, and that for the latter at 333, respectively. These values approximate closely to the actual maximum values, i.e., 910 and 338 in substances. With these values of G as base, the necessary percentages were calculated, and further they were transformed into logits, and finally

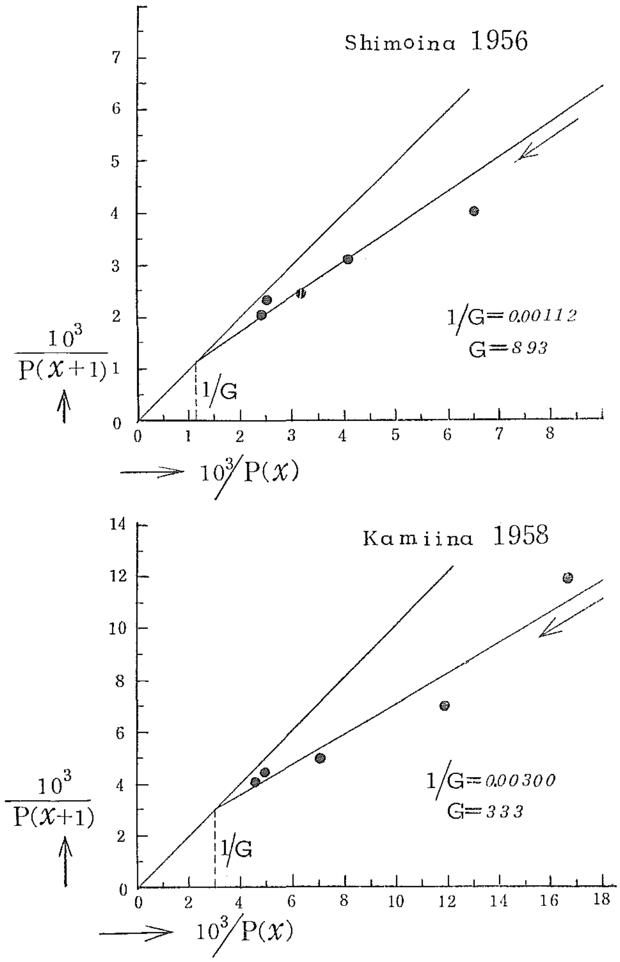


Fig. XI. The finite differences diagram for estimating the maximum value, G, of the cumulative emergence curve. An arrow on the graph indicates the direction from the initial to the latter stage of emergence.

the formulae for the predictive logit lines of least squares, in th true sense of the words, were calculated. In table XII are presented all these estimates together with the values calculated from the observed maximum values. Apparently, all the estimates approximate very closely to the actual, showing that they are fit for prediction use to the full.

The utmost care should be taken to avoid the errors involved in drawing the line corresponding to the linear equation (1), because whether the line is drawn accurately or not affects very sensitively the precision in the estimation of the value of G. In order to ensure the accuracy of the estimation, the best use of the trend shown in the arrangements of the dots which converge closer to the origin should be made without too much adherence to a few dots scattering at the portions far

TABLE XII. Estimated values of logits calculated on the basis of the assumed maximum value, G, of the cumulative number of the emergents for the composite natural parasites, and the logit line of least squares concerned.

Shimoina, 1956	June 22	24	26	28	30	July 2	3	5	Assumed G	Observed G
$10^3/P$	47.62	17.86	6.49	4.07	3.18	2.47	2.34	2.03	893	910
Estimated logits	−0.309	−0.078	0.160	0.290	0.367	0.460	0.482	0.544	(Calculated with G 893 as base.)	
Observed logits	−0.313	−0.091	0.154	0.284	0.361	0.452	0.474		(Calculated with G 910 as base.)	

Logit line of least squares, calculated from the estimated logits: $l=0.0655x-0.2678\pm0.0239$ (n=8)

Logit line of least squares, calculated from the observed logits: $l=0.0696x-0.2888\pm0.0798$ (n=7)

Kamiina, 1958	June 25	26	27	28	30	July 1	2	Assumed G	Observed G
$10^3/P$	52.63	16.67	11.91	6.99	4.93	4.46	4.10	333	338
Estimated logits	−0.104	0.166	0.264	0.438	0.597	0.656	0.719	(Calculated with G 333 as base.)	
Observed logits	−0.112	0.167	0.259	0.432	0.588	0.646	0.707	(Calculated with G 338 as base.)	

Logit line of least squares, calculated from the estimated logits: $l=0.1098x-0.0955\pm0.0788$ (n=7)

Logit line of least sqnares, calculated from the observed logits: $l=0.1089x-0.0985\pm0.0785$ (n=7)

Note: Figures at four places of decimals in the third and the fourth line in each table were omitted.

distant from the origin, since the latter is usually subject to sharp fluctuations in position because of their expressing the cumulative number of the emergents in the initial, rather unstable, stage of emergence.

iii) *The results obtained from the outdoor emergence trap*

The results hitherto analyzed rest on the data acquired from the galls kept inside the room. In order to examine the artificial effect, if any, the result obtained from the outdoor emergence trap was investigated. As can be seen in phot. I. c. the trap was settled on a bundle of branches of a wild chestnut tree on 17th June, 1959. The day was just at the time when an indoor observation was started. The emergents which came up into the attached glass tubes were counted and indentified every day as a rule.

The cumulative percentage emergence curves concerned (Fig. XII) are considerably irregular and remarkably different in shape from those founded on the indoor data. However, the most important ecological features such as the date of the first appearance of the parasitic wasps, the time of termination of the emergence of both the host and the parasites, and the trend that the period of emergence of the group of parasites is far earlier than that of their host, are roughly coincident with each other. The most striking contrast is a remarkable fall of the curve corresponding to the early part of emergence period for the parasites as well as the conspicuous retard of the first appearance of the chestnut gall wasp amounting to 12 days. These phenomena are attributed to the fact that the emergents at that time were attacked entirely by two species of predators. They consist of the black ant *Formica fusca japonica*

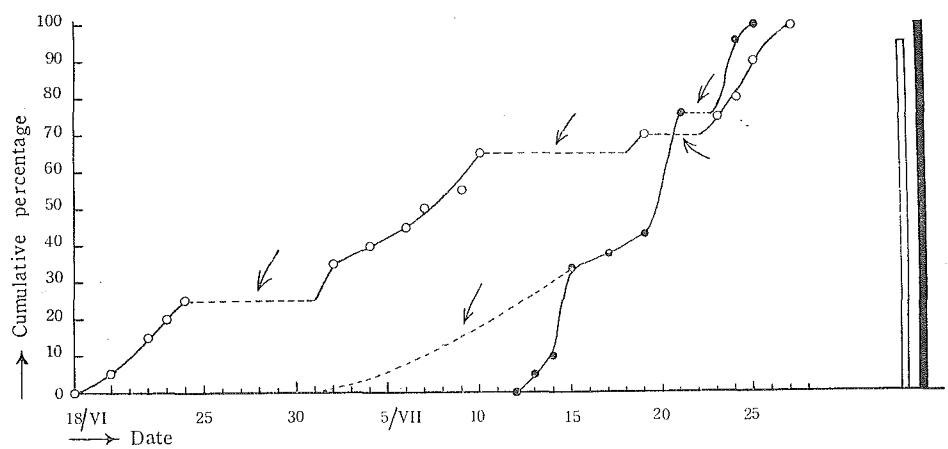


Fig. XII. The cumulative percentage emergence curves constructed with the result obtained from the outdoor emergence trap.

Note: An arrow indicates the fall due to entirely no capture of the emergents caused by the attack by their predators.

○ : The composite parasitic wasps

● : The chestnut gall wasp

(dark bar / white bar) : The percentage ratio of the total number of the emergent composite parasitic wasps (white bar) to that of the chestnut gall wasp. (dark bar).

Mots. and the micro-spider whose scientific name is unknown. The population of these predators increased in size as the number of the emergents inside the trap increased. In fact, a pretty many individuals emerging in the beginning of July were found having been captured in a web, and many ants were observed to have been active on the twigs inside the trap. This relation is also clearly refected in the fact that the percentage ratio (95.2%) of the total number of the emergent composite parasitic wasps to that of the chestnut gall wasp (100%) is abnormally large in value. The wasps in the glass tube attached to the trap, however, increased in number as the season advanced and their population became larger in size. Probably, this mutual relation between these predators and the wasps may be natural in the field, but it cannot be overlooked that the relation must have been more accentuatd than that under the natural open system because of its being established inside the closed system of an outdoor emergence trap.

From this, it may be concluded that the information of the data obtainable from this outdoor emergence trap is invalid as compared with that of indoor data in the analyses of this kind of phenomenon. At the same time, it may safely be said that the conclusions drawn from the foregoing analyses founded upon the indoor data are sufficiently reliable.

iv) *The method of prediction to be recommended*

Summing up the foregoing analyses, we shall be able to prescribe the following procedures necessary for the method of prediction to be recommended.

(i) To take count once a day or at least once each alternate day of the emergents of the group of parasitic wasps issuing from the chestnut galls in the period of the latter half of June, and sum up successively the numbers of the emergents observed on each date in the early stage of emergence to make the necessary cumulative numbers.

(ii) To estimate the maximum value of the cumulative number of the emergents in order to calculate the cumulative percentage emergence for prediction use.

(iii) To transform the values of the estimated cumulative percentage emergence into logits for prediction use, viz., the predictive logits.

(iv) To calculate the predictive logit line of least squares, L_3, and the measure, S_{E3}, of the deviation of the predictive logits from the line L_3.

(v) To fix the "period fittest for the biological control of the chestnut gall wasp" for the period put between the date corresponding to the upper limit, L_3+S_{E3}, and that corresponding to the lower limit, L_3-S_{E3}.

Attention should be paid in this case to the season of gall collecting as well as to the number, n, of days for prediction use. As previously pointed out, the best season fit for gall collecting seems to be about the middle of June. The collection made in too early a season is liable to bring an unreliable result against our purpose. The amount of galls to be gathered should be more than 500 grams at least. The number, n, of days for prediction use needs to be 5 or more at least, otherwise the prediction is apt to become inaccurate in some measure.

(8) *Advantage of the Logit Transformation*

a) Applicability to the analyses of the cumulative percentage curves in general

With the object of predicting the emergence of the rice-stem borer moth, *Chilo suppressalis* Walker, comparatively wide use of the probit method has been made in our country these few years. For instance, it is the case in point to predict the period of 50% emergence of the pest through the transformation of its cumulative percentage pupation into probits. From the the theoretical angle, apart from the practical utility, however, some defects have hitherto been pointed out by me (1959). One is that the original, not cumulative, curve, say, the percentage pupation curve, does not always conform to the normal curve, but often tends to be characterized by some other more flat-topped symmetrical curves, or sometimes by some asymmetrical curves. The other is that, when the data can not be expressed as percentage until the phenomena are over, the way to its utility will entirely be shut up. As previously demonstrated, evidences are clear that these drawbacks other than that in the case of asymmetrical curves can perfectly be overcome by the method of logit transformation.

In short, it may safely be stated that the way to the rational method of emergence prediction has broadly been opened by the introduction of the logit analysis into the domain of applied entomological science.

b) Generalized logit method

When the percentage curve, say, the percentage pupation curve, is asymmetrical in shape, its cumulative percentage curve can, of course, no longer be fitted by the integrated normal curve. If such a cumulative percentage curve be fitted well by the monotonely non-decreasing skew logistic curve

$$p=\{1-\exp[-(a+bx+cx^2+dx^3)]\}^{-1}, \quad \ldots\ldots\ldots\ldots\ldots(2)$$

the values of p can be transformed into logits just as in the case of the symmetrical logistic curve. Namely, put

$$l=\frac{1}{2}(a+bx+cx^2+dx^3). \quad \ldots\ldots\ldots\ldots\ldots\ldots\ldots(2)$$

And then the equation

$$l=\frac{1}{2}ln p/(1-p)=\frac{1}{2}(a+bx+cx^2+dx^3) \quad \ldots\ldots\ldots\ldots\ldots(3)$$

is easily derived from (1) and (2). Apparently, the relation between l and x is curvilinear. The polynomial (2) includes the straight line (when c=d=0) and the parabola (when d=0) as special cases. In this sense, the logit transformation difined by the relation (3) may be called the logit transformation in the wide sense, and consequently, the logit method based on this transformation the generalized logit method. This is one of the advantages of the logit transformation over the probit one. Needless to say, the logit transformation involved is quite equal in its numerical procedure to that in the case of usual logit transformation; the only difference between the two lying in the process of fitting a curvilinear equation (2). The process of fitting a cubic curve (2) to the data is given by the method perfected by R. A. Fisher (1950, § 27) in the special case where the successive values of x are arranged at equal intervals. The method of calculation involved is, indeed, rather complicated. But instead of such a precise curve a free-hand curve drawn through the logit dots will suffice our purpose only from the standpoint of practical use. It will be by far the better, therefore, to use a free-hand curve than to fit a straight line forcibly, in the case where the dots for the usual logit transformation do not agree with a straight line.

In this case, the value of logit, l, is zero, when p is equal to 0.5; therefore, the abscissa corresponding to the intersection of the cubic curve and the horizontal line falling on the ordinate 0 denotes the logit 50% date. It goes without saying that such a logit 50% date does not indicate the date of the maximum occurrence of the event, say, the maximum pupation, since it does not correspond to the peak of the original percentage curve of the occurrence of the event. What is to be regretted is that against such a skew logistic curve the method of estimating its maximum value is not yet found unlike the case of the simple logistic curve. This is the only bottleneck against its broad application and remains to be solved. For the present, therefore, its application must be confined to the case where the occurrence of an event can be expressed directly in terms of percentage in such a case as the rate of pupation and the rate of emergence of reared insects.

In this connection, the first-order derivative of p given by (1) is given by the equation

$$dp/dx=(\mathrm{b}+2\mathrm{c}x+3\mathrm{d}x^2)\exp(-2l)\{1+\exp(-2l)\}^{-2}, \quad \ldots\ldots\ldots(4)$$

where l is $(\mathrm{a}+\mathrm{b}x+\mathrm{c}x^2+\mathrm{d}x^3)/2$. This expresses the percentage curve in point, say, the asymmetrical percentage pupation curve.

c) Short-cut method of the logit transformation

The numerical value of $l=lnp/(1-p)/2$ can be obtained easily by the use of the table of natural, or common, logarithm. Instead of this usual method, the following short-cut method, too, may be employed as

$$l=\frac{1}{2}lnp/(1-p)=\frac{1}{2}ln(1-r)/(1-r)=Z,$$

where Z is Fisher's Z-transformation of r, correlation coefficient. In this case, it is necessary to put

$$r=2p-1.$$

The value of Z corresponding to $r=2p-1$ can easily be obtained from the table of Z-transformation. This short-cut method is very convenient when the trend of the variation of logits l is wanted to be known provisionally or cursorily. In regard to graphical transformation of the value of p into logit l, see my paper published in 1959.

(II) THE EXPERIMENTATION WITH THE LIBERATION OF A KEY PARASITIC WASP

As a part of the present study, an attempt was made to introduce a key parasitic wasp, *Torymus beneficus* Yasumatsu, into some chestnut gall wasp-infested areas in Shimoina District for three years from 1955 till 1957. This experimentation was conducted as a link in the project of the research programme of the Ministry of Agriculture and Forestry under the direction of Dr. K. Yasumatsu. It should be mentioned with gratitude in this connection that the officials concerned at the Shimoina Local Office had spared no efforts in cooperating with me in the liberation experimentation.

(1) Materials and the Environmental Conditions of the Area where Liberations were conducted

The key parasitic wasp with which the liberation experimentation was made was *T. beneficus*. Together with this species, some other species of parasites were tentatively released in some localities. In every case, the parasites released were the ones that emerged from the chestnut galls at Dr. Yasumatsu's laboratory, Kyushu University. As soon as they were received from the Entomological Laboratory at Kyushu University, they were transferred to a glass test tube and were allowed to feed on honey for two or three days before they were released. For some unavoidable reasons, it was only at Chisato area in Shimoina that the liberation could be carried out uninterruptedly for two years. At this area, the key parasitic wasp, *T. beneficus* was released with tolerably satisfactory results. The details of liberation are given in table XIII. Chisato area is situated at the scrubby valley along the left bank of the upper strean of the River Achi. The place where the liberation was carried out formed a more or less pure forest of wild chestnut trees each about 10-20 cm. thick, covering about 2 hectares in extent by eye measure. Adjacent to this wild forest, there is a grove of cultivated chestnut trees each about 30 cm. thick, extending about 1 hectare or less when measured with the eye. The other portions of the valley were dotted sporadically with pretty many wild chestnut trees. What is to be most regreted is that the great majority of the pure forest of wild chesnut trees were felled down

TABLE XIII. The dates on, and the localities at, which an effective natural enemy, *Torymus beneficus* Yasumatsu, parasitic on the chestnut gall wasp was liberated.

Year and date	Locality	No. of individuals liberated		
		Female	Male	Sum
April 20, 1955	Chisato (A)	153	17	170
May 4, 1955	Chisato (A)	155	11	166
May 6, 1956	Chisato (A)	143	34	177
April 20, 1956	Shimojo	91	78	169
April 18, 1957	Kamisato (A)	173	75	248

in the early winter of 1956 simply because of its being a private forest, and for this reason the liberation was compelled to be suspended halfway with a scant three-time liberations carried out for two years.

(2) Evaluation of Release Effectiveness

As originally discovered by Dr. K. Yasumatsu, the chestnut gall still remains greenish in colour in the latter part of autumn, when the gall wasps within it are parasitized by some other parasites, while the one free from such parasitic natural enemies is browned to be blasted early in autumn; acordingly whether or not such greenish galls can be found in abundance about the end of autumn will serve as an indicator of whether such parasitic natural enemies are active or not. In Ina district, too, such two kinds of galls can surely be observed; the brown galls are characterized by their deformed leaves dead already in the middle of August, while the greenish galls still are marked by their green leaves, though deformed, even in the end of September. Further, the latter are distinguished by a fewer exit or emergence holes as compared with the former. Judging from the physiological relation between the gall insect and its host plant, this distinctive feature of the greenish gall would be more conspicuous, if these parasitic natural enemies belonged to such species as pass the winter within the gall. In fact, such a phenomenon is reported by Dr. K. Yasumatsu to have been experienced at the southeastern climates where an overwintering single-brooded *T. beneficus* is produced in abundance. When this key parasitic wasp is, therefore, introduced into this district and established, the greenish galls will not only be detected more distinctly but also observed more abundantly than ever in autumn. From this point of view, the effectiveness of released wasps has been evaluated by the abundance of such greenish galls. The results are shown in table XIV. The census sampling methods used are similar to those described in Chapter II.

In the first year (1955) when the liberation was performed twice in the middle of spring, the number of greenish galls per unit twig in autumn was increased to about 30.5 per cent of the total number of galls. This is about 2-3 times as many as those found at the other regions far distant from the experimental station, though the latter are the results obtained in 1956. Evidence seems to be furnished by this that the released *T. beneficus* attacked the chestnut gall wasp to a considerably extent already in the first year when it was released. In the autumn of the second year (1956), the number of galls per unit twig was reduced to about 4.3 per cent of that in the first year, counting about 1.5 per unit twig in number; the great majority, i.e., about 91.2 per cet, of it was found to be greenish. At Chisato area B where is located about 6 kilometres downstream from the released station, Chisato area A, the galls per unit twig were also reduced to about 20.6 in number, which is far less than that found at Chisato area A in the first year. And besides, more than half of 20.6, viz., about 55.3 per cent of it, was greenish in colour, the percentage of which was far higher than those secured at the non-liberated areas in 1956, which ranged from 9.7% to 14.6%. In view of these results, it must be stated that the liberation 451 adult females of *T. beneficus* in all, which was carried out separately in three times during 2 successive years, produced a satisfactory result at the experi-

TABLE XIV. Comparison of the number of greenish galls observed at the *Torymus beneficus*-liberated area in fall with that observed at non-liberated areas.

	Date of which sampling survey was made	Area	Average no. of gall		No. of greenish galls Mean		%	
Torymus beneficus-liberated areas	Sept. 10, 1955 (1st year)	Chisato (A)	35.0		10.7		30.5	
	Sept. 28, 1956 (2nd year)	Chisato (A)	1.51		1.35		91.2	
		(Chisato (B))	(20.6)		(11.4)		(55.3)	
	Oct. 1, 1957 (3rd year)	Chisato (A) (Wild chestnut)	22.3	21.3	11.7	9.1	52.4	42.7
		Cultivated 〃	20.2		6.4		31.2	
		(Chisato (B))	(35.9)		(4.7)		(12.9)	
	Sept. 11, 1957 (1st year)	Kamisato (B)	23.8		13.9		58.4	
Non-liberated areas	Sept. 8, 1956	Kamisato (B)	21.3		3.1		14.6	
	Sept. 8, 1956	Uriki	9.5		1.3		13.7	
	Sept. 8, 1956	Namiai	24.0		2.5		10.4	
	Sept. 8, 1957	Goka	15.4		1.5		9.7	
	Sept. 11, 1957	Iida (A)	32.7		2.5		7.7	

Note: A twig about 1 cm. thick was sampled as a unit sample per tree. Figures underlined indicate the mean value taken concerning whild chestnut data and cultivated ones.

mental area, and was considerably effective even at the area where is located about 6 kilometres downstream.

Unexpectedly, the chestnut trees with which the release experimentation had been continued were felled down extensively in the winter of 1956; the release experiment, therefore, was obliged to be suspended thenceforth. The observation of greenish galls was, however, continued at both the environs of the experimental station and the neighboring grove of cultivated chestnut trees to see if ever *T. beneficus* was capable of being established permanently in this region. As can be seen in table XIV, the average number of galls of the two areas A and B in 1957 was only 21.3 per unit twig, counting about 65% of that secured at Iida area A in the same year; the average percentage of the greenish galls remained to be about 42.7 per cent, amounting to about 5.5 times as high as that at Iida area A. At Chisato area B, the number of galls and the percentage of the greenish galls per unit twig were both found to be practically equal to those secured at the non-liberated areas. From this, evidence is clear that the released *T. beneficus* still remained active to a considerable extent at the environs of the liberated station for at least one year after the liberation had been suspended. It should be noticed, however, that the percentage of the greenish galls that were found on wild chestnut trees has decreased to about 57.5 per cent of that secured in 1956. Probably, this fact may be ascribed for the most part, if not all, to an adverse effect of discontinuance of release upon the propagation of the parasitic wasp newly introduced. Beside this, a large scale felling done in the winter of 1956 must have exerted a vicious influence on its propagation. Another important fact to be noticed is that severe late frost damage was experienced in the southern part of Shimoina District early in May, 1956. Not only various kinds of vegetables and fruit-trees but also wild chestnut trees were damaged heavily by the killing frost. At the request of the Shimoina Local Office, I examined a lot of chestnut galls nipped by the heavy frost, and found that the mortality rate of the larvae within the gall varied remarkably according to localities, scoring 96.5, 63.6, 41.0, and 34.8 per cent at 4 different localities, respectively. The galls for which 96.5% mortality of the larvae within them was recorded were blackened to death, most of their larvae being found to be dried up. It was just at the time when the introduced *T. beneficus* was in its activity. Probably, the great majority of *T. beneficus,* which wintered at that area, must have been killed together with the eggs laid by them as well as with those introduced on 6th May in 1956. Such an effect of weather conditions upon the propagation of newly introduced *T. beneficus* can not by any means be underestimated so far as this district is concerned, since such heavy frost late in season is not unusual in this district. These findings seem to furnish almost incontrovertible evidence that the severe frost late in season is apt to do fatal damage to the larvae of both the chestnut gall wasp and its parasites within the gall, though I can not assert positively whether the fatal injury is caused directly by coldness itself or indirectly by the death of the gall occasioned by the frost damage.

At Kamisato area, too, 173 females of *T. beneficus* were released in the spring of 1957. By the census survey conducted in the autumn of that year, the result was found to be obviously satisfactory just as in the case of Chisato area A in the first year; the percentage of greenish galls amounted to about 58.4 in value, viz., about 7.6 times as high as that secured at Iida area A, a non-liberated area, in the same year.

In view of the results so far obtained, it may be interpreted that the introduced *T. beneficus* tends to propagate itself overwhelmingly for the 1st and the 2nd year of liberation, but, in a peculiar climate where severe late frost damage is experienced just in its active period, it tends to show a comparatively rapid diminution in its propagation in the case where its subsequent liberation is interrupted.

(3) Discussion

What is to be discussed may be the results secured in the autumn of 1957, one year after the release was suspended. If these results are interpreted consistently, some inference may be drawn as to the question of whether the establishment of the introduced key parasitic wasp is permanent or not.

As aptly pointed out by Thompson (1924), the time required for the attainment of host control by an introduced parasite may depend primarily upon two factors, that is, the ratio between initial number of parasites and that of hosts, and the ratio between the rate of reproduction of the parasite and that of host. Naturally, such time will be lengthened by parthenogenesis in the host; it will also be affected by such phenomena as superparasitism and co-parasitism. In our case, the host gall wasp is surely believed to be a parthenogenetic insect. But from this point alone, the results secured in the autumn of 1957 will not be explained consistently. Viewed from the body-size relation between host and parasite, superparasitism and co-parasitism, too, seem to be almost impossible to occur, and to exert so great influence upon such time, if any, in this case.

Another note worthy relation may be the ratio between the initial number of the introduced parasite and the size of the host population in question. To be sure, the individual number of the released *T. beneficus* is so small that it is incapable of bearing comparison with that of the host to be attacked. From this respect alone, however, we shall have to find difficulty in consistent interpretation of the comparatively rapid increase in the percentage of greenish galls observed in both the first year of liberation and the next year. If taken into consideration the status in the southwestern climates where *T. beneficus* is native and the chestnut gall wasp overwhelmingly large in population is controlled successfully by it, it seems to be hardly possible to attach so much importance to such host-parasite interrelationship concerning the initial number as pointed out by Thompson, if any. What should be considered more carefully is that the scarcity of the host gall wasp resulting from the heavy attack by the introduced *T. beneficus* may greatly be responsible for such a diminution in greenish gall percentage as observed in 1957. According to Dr. Yasumatsu's work (1956), a very interesting theory has been advanced by Sellers (1953) on the time factor governing the relation between the population density of host and that of its natural enemies. Sellers refers to this theory as follows: When environmental conditions are kept favorable and the connection between a pest and its natural enemy is satisfactorily, the pest population is checked greatly by its natural enemy, provided that the initial density of the pest population is so high as to make its natural enemy propagate actively. It is at this time that the effect of the natural enemy is displayed at the maximum. But the propagation as well as the effectiveness of the natural enemy will be reduced in its next generation, as a natural result of a marked decrease in its host population. Even under such conditions, however, the pest population is still checked at such a level as not to exert economic influence upon us, though the pest population is higher in density than that at the time when it was reduced markedly; thus the relation between a host and its natural enemy is balanced in this manner. Judging from the results secured at Chisato area B in 1957, however, I am still to find not a little difficulty in explaining the phenomena from this relation alone, though I never make light of the influence of such a relation between hosts and parasites.

Of Sellers' theories, what seems to be most fitted for the explanation of our case may be the theory on the relation between the time necessary for bringing about effective control of a pest and the dispersal rate of its natural enemies. He is of opinion that the slower is the introduced natural enemy in its dispersion, the quicker is its effect, since the natural enemy that is slow in its dispersion is quick in attaining to the equilibrium state of its distribution. In view of the remarkable increase of greenish galls observed at Chisato area A in the autumn of 1956, one year after *T. beneficus* was introduced thereinto, this parasitic wasp seems to belong to what Sellers calls the species that is slow in its dispersion. If that is the case, the conditions adverse to its propagation such as the large-scale felling of the chestnut trees with which its release experimentation was continued and the severe damage done by the heavy frost that took place just at the time when it was in full activity in host seeking and oviposition, that is to say, during the period spent away from a host, must have produced a fatal effect in a large way on the population of *T. beneficus,* because the population must have attained to the equilibrated dense distribution at least at the experimental station and its environs already in the spring of 1956 because of its slow dispersion.

In short, the fact that the population of *T. beneficus* was in the state most susceptible of being affected concentratively by any natural or artificial damage seems to have reduced its effectiveness far below its maximum potential in concert with adverse weather as well as environmental conditions just occasioned.

According to the book written by Wardle (1924), Howard (1924) emphasizes that, in any scheme of parasite transference, importance should be attached to the fact that there is no native parasite so closely related to the introduced form as to be capable of hybridising with it. Howard fears that the effectiveness of the introduced parasite as one on the host to be controlled is decreased due to such an interbreeding with the native parasite. It may surely not be unlikely to happen so in our case, because *T. elegantulus,* a species belonging to the same genus as *T. beneficus,* inhabits here comparatively abundantly. According to Dr. Yasumatsu's unpublished data, however, *T. beneficus* emerges early in spring, except in a very few extraordinary cases, and the period of emergence ranges from March to about 20th April in the southwestern warm climates. On the contrary, the period of emergence of *T. elegantulus* ranges from about 20th June to about 10th July in this district. Since there is a discrepancy more than one month between their emergence periods, even if the longevity of the former is taken into account, it may be said that the active period for the former cannot almost be synchronized with that for the latter. Thus, it will boil down to this conclusion that it is almost out of the bounds of possibility that such an interbreeding as emphasized by Howard should occur, so far as *T. beneficus* introduced into this district is concerned.

In regard to the factor governing the rapid increase of the introduced parasites, Howard (1924) emphasizes further that importance should be attached to the necessity for climatic similarity between the two areas concerned, or for climatic suitability of the area into which the introduction is to be made. Affirming the discussion made by Shelford (1926) on the relation between the influence of meteorological factors upon parasite abundance and the ecological conditions on the part of both host and parasite, Wardle (1929) has also pointed out that "activities of an adult female parasite as regards host seeking and oviposition may be, in particular, curtailed by weather conditions." DeBach (1958) also attaches importance to the role of weather in the natural control of insect populations. He stresses that "weather regulates insect population only by being of sufficient severity to restrict the size, quality (including food and favorability), and/or numbers of inhabitable spots in a given area, that is, its effect is obtained through interaction with microenvironment." Weather regulation is surely a factor to be paid special attention to in our case, too. As previously pointed out, *T. beneficus* emerges early in spring, and naturally it is liable to be injured by the heavy frost late in season, as is indeed the case. In reality, a great majority of the larvae of the chestnut gall wasp and probably its parasites within the galls were unable to survive the severe freeze caused by the heavy frost early in May, 1956. I have not yet accumulated abundant evidence so sufficient as to assert positively such weather regulations as the prime factor governing the ultimate establishment of the introduced

T. beneficus in this district, but from the consideration of the results so far achieved as well as of the ecological features peculiar both to this parasitic wasp and to this district, I am inclined to think that one of the critical factors adversely affecting the propagation of *T. beneficus* was weather.

Although the problem of what the ultimate establishment of this species will be in this district remains to be seen, so far, suffice it to say that, for mapping out the definite scheme of controlling the chestnut gall wasp by the aid of introduced *T. beneficus,* it is very important to clarify if there is any essential barrier to its propagation as well as its permanent establishment. In this connection, it will be very instructive to quote Howard's opinion to the effect that it is necessary to allow a sufficient period of time to elaspe before forming an opinion of the success or otherwise of the parasite introduction experiment.

Apart from the problem of permanent establishment, *T. beneficus* seems to be sufficient to become what Clausen (1950) calls an effective natural enemy on the chestnut gall wasp in this district, too, since the striking success at Chisato area A in 1956 was achieved just two years after *T. beneficus* had been released, that is, just in the time covered by only two host generations. The reason is that, according to the statement made by Clausen, "an effective natural enemy might be expected to show evidence of control at the point of release within a period of three host generations or three years". Really, even in the proven cases of excellent natural control, such a striking success as this instance has rarely been reported. Thus, it is certain that the field liberation of this parasitic wasp offers the most likely promise in the biological control of the pest wasp in question, if only an adequate scheme of liberation is mapped out. From the purely practical standpoint alone, therefore, the following line of liberation may be recommended: Namely, a considerable number of *T. beneficus,* which is divided into small groups, should be released scatteredly at several localities throughout the infested area at least once every other year until the host wasp population becomes checked down to below the level of economic density limits.

(III) HOST PREFERENCE BY THE P^{32}-LABELED PARASITIC WASPS

(1) The object of Research

As a rsult of the detailed investigations previously mentioned, it has been found that the parasitic wasps and flies emerge about 12 days or so earlier than their host gall wasp does, completing their emergence as many days earlier than the latter. From the standpoint of preservation of these native parasitic natural enemies, it will be of great importance to trace what kinds of gall insects will be chosen by these parasites as their next host after they have emerged from the chestnut gall. For this purpose, I have undertaken to investigate such host preference by using P^{32}-tagged parasitic wasps as a tracer. The experiment was carried out once each in 1958 and 1959.

(2) Method of Research

a) Parasitic wasps employed

The parasitic wasps employed were the ones which emerged from the chestnut galls kept in our laboratory in the period from 25th June to 9th July (1958) and that from 22nd June to 20th July (1959).

The galls used were collected at both Tagiri and Suwagata (1958) and at Mikoshiba (1959) each in Kamiina District on 24th June (1958), and on 20th June (1959), respectively. The kind of species and the number of their individuals were shown in the following table.

Specific name	No. of individuals			
	1958		1959	
	Female	Male	Female	Male
Megastigmus spp.	98	53	13	3
Ormyrus nigritibialis	33	24	20	6
Eurytoma rosae	14	9	4	2
Torymus elegantulus	8	11	—	—
Amblymerus amoenus japonicus	14	8	13	1
Peleumus ferrierei	9	16	1	—
Total	176	121	51	12

This composition in species and number can be regarded to be roughly proportionate to the pattern of the composite population of the wasps parasitic on the chestnut gall under natural conditions in this district in each year. They were sorted and kept in the glass tube (2.5 cm. in diameter and 13 cm. in length) on the day when they emerged, and were allowed to feed on honey which was soaked into match sticks. The specific names and sexes were again identified accurately after the experiment was over by examining their dead specimens.

b) Method of tagging

i) *Preparation of a tracer*

The honey used as bait was composed of the following materials:

Material \ Year	1958	1959
Dist. water	12 cc.	20 cc.
Honey (on the market)	30 cc.	15 cc.
P^{32}	3 mc.	3 mc.

The radioactive phosphorus P^{32} was in the form of orthophosphate in dilute hydrochloric acid and had an activity of 16.6 mc/ml. at 1 a.m. on 8th July, 1958, and 34.8 mc/ml. at 1 a.m. on 9th June, 1959, respectively. These materials were fully mixed with one another in a closed petri dish. Into this solution, 35 pieces of match sticks (decapitated and washed well) were immersed and kept for 1 day (in 1959) or 2 days (in 1958). One stick gave a reading of 1666.4 ± 297.0 counts per munite (in 1958) and that of 75.8 ± 8.4 cpm. (in 1959) at the time when feeding commenced. Counts of radial rays were taken with a shielded end-window Geiger-Müller tube. 15 (in

1958) or 5 pieces (in 1959) of the same glass tubes as mentioned above were prepared. Into each of them, the match stick prepared was put at the rate of 3 pieces a tube (see phot. III.). About 20 or fewer individuals of the same species of parasitic wasps including both sexes were put into each tube and were allowed to feed on P^{32}-labeled honey for about 2 days and a half (in 1958) or for 28 hours (in 1959), respectively. Before being let in the tube, they were starved for about 16 hours (in 1958) or about 19 hours (in 1959) in a dark room. To avoid getting to much fatigued during the feeding period, they were confined in the dark room for the most part of daytime, except for three hours a day, viz., for the time allotted to copulation and exercise.

ii) *Release of P^{32}-tagged wasps*

Before released into the cage for release use, the P^{32}-tagged wasps were transferred from the glass tube for tagging use into a clear RI-free glass tube, inside which a belt of blotting paper 2 cm. wide and 13 cm. long was laid in order to absorb the honey sticking outside the body of the tagged wasps; the wasps therein were exposed to room light to be called into activity for about 2 hours. The cage for release use is illustrated in phot. IV-a. It is a cage $40 \times 40 \times 75$ cm.^{3} in dimension, the four sides and the ceiling of which are netted with the brass net of very minute mesh. 9 (in 1958) or 10 flasks (in 1959) 300 cc. in capacity were placed in each cage; one twig, or stem, with a definite kind of insect galls was put into each flask at the rate of a twig per flask. The P^{32}-labeled parasitic wasps were released into each cage at the combination shown in table XV in 1958. In 1959 the arrangement of the plants caged with them was more simplified and systematized. Four cages A-D were employed; in each of which a set of 13 kinds of plant specimens (which are listed in table XXI) were placed in such a manner as one specimen occupies one flask, although the two specimens (3) and (4), (6) and (7), and (9) and (10) (see the same table) were put together into one flask, respectively. The parasitic wasps released into each cage are as follows:

Cage	Species	No. of individuals	
		Female	Male
A	*O. nigritibialis*	20	6
B	*Megastigmus* spp.	13	3
C	*A. amoenus japonicus*	13	1
D	*E. rosae*	4	2
	P. ferrierei	1	—
	(1 male died after copulation)		

Four cages were all placed in the large outdoor cage $90 \times 90 \times 175$ cm^3 in dimension as illustrated in phot. IV-b. The large outdoor cage is also netted with the same net as that of the cage for release use. Various kinds of insect galls and blotches as well as deformed leaves due to piercing-sucking insects, as shown in tables XV and XI, were selected as samples for presumptive second-generation hosts to be chosen by the parasitic wasps under inquiry. They were all collected at the scrub forest near our college grounds (situated at Mikoshiba in Kamiina district) on 11th to 12th July in 1958 and on 23rd July in 1959. Up to this time, the chestnut gall wasp thereat

TABLE XV. Released parasitic wasps and the plants caged with them as supposed hosts on which the released wasps are to lay their eggs. (The data in 1958).

Cage	Plants caged with the released parasitic wasps: Designation	No. of twigs or stems	Parasitsc wasps released into each cage: Specific name	No. of individuals: Female	Male	Sum
No. 1	Oak knot galls	8	*M.* spp.	23	8	31
	Oak scale galls	1	*O. n.*	5	3	8
	Chestnut young terminal shoots with chestnut galls	4	*E. r.*	5	2	7
			A. a. j.	3	2	5
			P. f.	2	3	5
No. 2	Chestnut twig with chestnut galls	1	*M.* spp.	25	12	32
	Oak twigs with leaf rolls	2	*O. n.*	9	4	13
	Oak twig with ball-galls on leaf veins	1	*E. r.*	2	4	6
	Oak twig with leaf-miner blotches	1	*T. e.*	4	2	6
			A. a. j.	5	0	5
			P. f.	3	3	6
No. 3	Oak twigs with large oak apple gall	2				
	Chestnut young terminal shoots with chestnut galls	2	*M.* spp.	22	13	35
			O. n.	8	4	12
	Mugwort stems with leaf warty galls	2	*E. r.*	5	1	6
	Mugwort stems with bowl-like galls	1	*T. e.*	1	6	7
	Cherry twigs with leaf aphid burns	2	*A. a. j.*	4	2	6
	Oak twig with leaf rolls	1	*P. f.*	3	4	7
No. 4	Chestnut young terminal shoots with chestnut galls	4	*M.* spp.	18	15	33
	Chestnut young terminal shoots without chestnut galls	3	*O. n.*	6	5	11
			E. r.	4	2	6
	Chestnut twig with chestnut galls	1	*T. e.*	3	3	6
	Quercus Acutissima young terminal shoot (with a leaf having one red ball gall)	1	*A. a. j.*	2	4	4
			P. f.	1	6	7

Note: (1) In each cage, nine flasks were put, each with a unit sample. 2 mugwort stems with leaf warty galls in the cage no. 3 were put together in one flask.

(2) *M.* spp.: *Magastigmus* spp.
T. e.: *Torymus elegantulus* Yasumatsu.
E. r.: *Eurytoma rosae* Nees.
O. n.: *Ormyrus nigritibialis* Yasumatsu.
A. a. j.: *Amblymerus amoenus japonicus* Yasumatsu.
P. f.: *Peleumus ferrierei* Yasumatsu.

(3) The relative rate of the number of the released parasitic wasps is roughly proportinate to that of the parasitic wasps emerging from the chestnut galls. See also the table given in Chap. III, Sect. (2), Par. a).

must have already emerged to the extent of about 59-64 per cent (in 1958) and that of about 85 per cent (in 1959) of its total emergence at least, as can be inferred from the cumulative percentage emergence curve shown in Figs. VI and VII. As a natural result, the buds of the young shoot of chestnut trees thereat are to have been exposed to the attack by the chestnut gall wasps that have already emerged by that time. The release experimentation was conducted for 116 hours from 12th to 17th July in 1958, while in 1959 for 48 hours from 23rd to 25th July. During this period, water was added into each flask once a day by way of precaution against evaporation.

c) Preparation of specimens of dried leaves and galls

According to the usual method, the specimens of dried leaves and galls under examination were prepared on 17th July in 1958 and on 25th July in 1959. Utmost precautions were, of course, exercised against contamination due to contact with each other. The pressing of specimens was continued until 5th August in 1958 and till 8th August in 1959 to dry them up to the full. For autoradiographical use some representative specimens were selected from each sample. These were sorted into several groups and each of them was fixed on a thick setting board $34 \times 14\,cm^2$ in dimension with Scotch tape, respectively, as illustrated in phots V-X (1958) and in phots XI-XIV (1959).

d) Preparation of macro-autoradiographs

The usual contact method was adopted. Each setting board with several kinds of specimens was covered with a sheet of damp-proof cellophane paper by way of precautions against chemical fog. Fuji non-screen type X-ray film no. 200, which is very sensitive to X-ray, was used. The X-ray film plate was brought into contact with a setting board on which several kinds of specimens were fixed, and then the two were bound into a bundle with a rubber band. When binding them, too much pressing was, of course, avoided for precautions against pressure fog. Each bundle was wrapped up in a sheet of large black light-tight paper and further was folded in a sheet of large vinyl wrapper to be kept under moisture-tight conditions. All the bundles were wrapped up in a sheet of large vinyl wrapper and were kept cold within an electric refrigerator whose internal temperature was kept constantly at 5°C. or so. By way of precautions against under exposure, these bundles were left at that condition for 4 months and 23 days in 1958 and for 30 days in 1959, respectively, though any longer time of exposure than 4-fold the half-life time, 14.3 days, of radioactive P^{32}, is quite meaningless. Fuji-rendol and Fuji-fix were employed as a developer and/or a fixative, respectively. Before the developing operation started, the bundles which were taken out of the refrigerator were left as they were for about one day at room temperatures for precautions against their being moistened by the sudden exposure to warm air in the room. Conditions under which a release experiment was carried out are listed in the following table.

Treatment \ Year	1958	1959
Plant collection	11–12th July	23rd July
Release	12–17th July (for 116 hrs.)	23rd–25th July (for 48 hrs.)
Pressing of plant specimens	17th July–5th Aug. (for 19 days)	25th July–20th Aug. (for 26 days)
Contact with X-ray film	5th Aug.–28th Dec. (for 145 days)	20th Aug.–19th Sept. (for 30 days)
(kept constantly at 5°C. or so)		

(3) Results

a) Radioactivity of P^{32}-tagged parasitic wasps

i) *Results obtained in 1958*

At the time of release, viz., on 12th July, some individuals belonging to two species of parasitic wasps, *Megastigmus* spp. and *O. nigritibialis,* which are most abundant in number and the largest in body size, were selected and their radioactivity were measured with a shielded end-window of a Geiger-Müller tube. These were selected from the 8 glass tubes that were chosen randomly from the whole 15 glass tubes for tagging use. All the wasps selected were killed with ethyl acetate, and the counts were taken of the radial rays of an intact wasp singly for 1 minute repeatedly and then of that of such dissected portions as the head, the thorax with legs and wings, and the abdomen, respectively. The average number of their readings of counts per minute is presented in table XVI. The readings of cpm. for each in-

TABLE XVI. Radioactivity (averages of cpm. taken at 5-cm. distance) of adult parasitic wasps (on 12th July) after feeding for about 60 hours on a honey solution (well soaked up in matchsticks) containing 3 millicuries of radioactive P^{32} in 42 cc.

Species examined	Sex	Part examined									No. of individuals examined
		Before dissection	After dissection								
		Intact body	whole body		Head		Thorax		Abdomen		
		cpm	cpm	%	cpm	%	cpm	%	cpm	%	
M. spp.	Female	41.0±5.9	37.4	100	3.7	9.9	12.8	34.2	20.9	55.9	10
M. spp.	Male	40.7±6.4	38.5	100	3.9	10.1	13.6	35.3	21.0	54.6	5
O. n.	Female	27.1±4.7	23.4	100	2.2	9.4	7.7	32.9	13.5	57.7	5
O. n.	Male	25.5±4.3	26.2	100	2.0	7.6	8.6	32.8	15.6	59.6	8

Note: (1) *M.* spp.: *Megastigmus* spp.; *O. n.*: *Ormyrus nigritibialis.*
(2) The cpm. of the dissected portions was taken with 3 or 5 portions in a lot for 1 minute repeatedly.

dividual varied to a considerable extent, indicating that there was a remarkable lack of uniformity in the amount of P^{32} acquired by each individual, though no individuals untagged. Generally speaking, however, *Megastigmus* spp. is observed to tend to be more strongly tagged than *O. nigritibialis* did, probably because of its greatness in body size. From this, evidence is clear that the adult-feeding method succeeded in tagging the parasitic wasps used, leading to the production of the wasps having a pretty strong radioactivity sufficient to permit their distinction from non-labeled wasps. When dissected, the abdomen showed the strongest radioactivity among the three portions, the thorax with legs and wings showing comparatively strong radioactivity next to the abdomen, and the head the least. In the distribution of P^{32} in the body, this result bears some resemblance to those described by Hoffman, Lindquist and Butts (1951) for adult blow flies and/or to those by Yates, Gjullin and Lindquist

(1951) for Aedes mosquitoes. From this result alone, however, it is impossible to clarify whether the reading of cpm. means really the internal tagging of the body or merely its external contamination with radioactive honey. Although no further examination was conducted in this respect, it will not be impossible to get the rough idea of it acording to the results obtained by many investigators of a certain species of insects and mites under diversified conditions.

With regard to the P^{32}-labeled Douglas-fir beetle, Fang and Allen (1955) have clarified that only 2 per cent of the total radioactivity was removed by washing, indicating that nearly all the P^{32} was in the insect, and that about 16.6, 14 and 53 per cent of the remainder in the head, thorax and abdomen, 6.8 and 9.3 per cent in the legs and wings, respectively. According to the result secured by Foott (1955), the radioactivity of the P^{32}-labeled adult cabbage maggot, *Hylemyia brassicae* (Bch.), was not removed by washing, and females developed significantly more radioactivity than males, some laying radioactive eggs. In his study on radiophosphorus in metabolism in the two-spotted spider mite, Rodriguez (1954) has pointed out that utilization of phosphorus in egg production proceeded at a rate similar to uptake, and phosphorus was three times as concentrated in the egg as in the mite body. Concerning the distribution of radiophosphorus in wax moth, meal worm, cochroach and firebrat, Lindsay and Craig (1942) have also shown that in all the insects used the P^{32} was least concentrated in the fat body and most concentrated in the epithelium of the mid-intestine, in the reproductive ducts and in the gonads.

The species described above are, to be sure, wide apart taxonomically from the parasitic wasps employed in the present investigation. But, considering all the above results collectively, it may safely be stated that the difference in the readings of cpm. for three major portions of the body of the wasps examined should not be ascribed solely to the external contamination of such portions, but principally to the very difference in the amount of P^{32} present in such three portions of the body. This interpretation may also be supported by the fact that the parasitic wasps were observed to flock on the matchsticks tagged with P^{32} containing honey to lap up honey as soon as they were put into the feeding glass tube after long-hour starvation. (cf. phot. III.)

After the release experiment was over, the counts were taken again of the radial rays of the three major dissected portions of the body of the released parasitic wasps. The readings of cpm. in this counting are indicated in table XVII. As can be seen by comparison with the readings shown in table XVI, the relative decrease in percentage radioactivity is most great in the female abdomen of the two species examined. Besides, the counts for female abdomens were found to be smaller by about 11.5-13% than those for male ones, while they were substantially equal to each other at the time of release. This must be a noteworthy fact. According to the result described by Roan (1952) concerning the P^{32}-labeled adult Oriental fruit flies, the rate of loss of P^{32} by females is always more rapid than that by males up to 40 days after emergence, and the difference between the two was regarded by him as considerably attributable to the oviposition by the parent female, since an 80-mg. sample of eggs collected from the flies 15 days after their emergence gave a reading of 1000 cpm. His conclusion

TABLE XVII. Radioactivity (averages of cpm. taken at 5-cm. distance) of adult parasitic wasps on 18th July (after 6 day- release).

Species examined	Sex	Part examined: Before dissection, Intact body	After dissection: whole body		Head		Thorax		Abdomen		No. of individuals examined
		cpm	cpm	%	cpm	%	cpm	%	cpm	%	
M. spp.	Female	32.7±6.5	30.8	100	4.2	13.7	15.3	49.6	11.3	36.7	15
M. spp.	Male	33.8±5.1	32.4	100	3.3	10.2	13.0	40.1	16.1	49.7	10
O. n.	Female	20.9±3.7	17.3	100	2.5	14.5	8.0	46.2	6.8	39.3	8
O. n.	Male	19.8±3.5	18.5	100	1.9	10.3	7.2	38.9	9.4	50.8	5

has, of course, been drawn after due consideration of the loss of P^{32} due to excretion. In their study on the translocation of radioactive phosphorus injected by the green peach aphid into tobacco plants, Lawson, Lucas and Hall (1954) have pointed out that the tracer was found in the honeydew and excreta by the aphids, in their cast skins and in their young born of radioactive individuals, though some of them may have been partly ascribed to contamination with honeydew. It is suggested in their study, therefore, that the oviposition by these insects plays an important role in the loss of P^{32} acquired by them in concert with their excretion. On the contrary, more stress is laid on the role of excretion in the loss of radioactivity by the P^{32}-labeled insect by Fuller, Riegert and Spinks (1954) in their study on the persistence of P^{32}-tagged grasshoppers. On the other hand, Tones and Wallace's data (1955) concerning the fruit fly show that the females having received heavy dosages of P^{32} failed to oviposit in cherries probably due to an adverse effect of P^{32} on the ovary, while their excreta and the juice and pulp of the cherries caged with them became radioactive. Their result also suggests the possibility that P^{32} ingested by the fruit fly is apt to be concentrated in the ovary as well as the other internal organs of the fly to a considerable extent. According to the data described by Babers, Mitlin and Shortino (1956) concerning house flies, the ovary of the house fly fed on the milk containing radioactive P^{32} for several days has undergone a remarkable change in its morphological characteristic, the eggs laid by them having considerably strong radioactivity.

Putting these results together and comparing the result given in table XVI with that given in table XVII, it would be regarded as appropriete to consider that in the present experiment, too, the parasitic wasps having fed on the honey containing P^{32} must have undergone a considerable change in their ovary and as a result their eggs must have labeled with P^{32} to a considerable extent.

b) Radioactivity of the plants caged with the P^{32}-labeled parasitic wasps

Before the dried plant specimens were exposed to the X-ray film, their radioactivity was measured with the Geiger-Müller counter above described. Counts of radial rays were taken at nearly zero distance for 1 minute repeatedly. The readings of 8 to 36.8 cpm. in average, which are presented in table XVIII, vary according as

TABLE XVIII. Radioactivity (averages of cpm. taken ten times for each specimen at nearly zero distance) of the dried plant specimens caged with the P^{32}-labeled parasitic wasps. Cpm. was taken on 5th August.

Cage	Dried specimens of the plants examined	cpm	Mark in phots. V~X
No. 1	Oak knot galls	32.0	E
No. 1	Oak scale galls	0	
No. 2	Chestnut young terminal shoots with chestnut galls		
	Chestnut galls	36.8	D
	Young terminal shoots	0	D
No. 2	Ball galls on oak leaf veins+oak leaf miner blotches +chestnut serpentine leaf miner winding trials	12.0	J
No. 2	Chestnut twigs with chestnut galls	24.0	I
No. 3	Chestnut young terminal shoots with chestuut galls		
	Chestnut galls	8.0	B
	Young terminal shoots	0	B
No. 3	Cherry leaf aphid burns	16.8	C
No. 3	Oak leaf rolls+oak leaf miner blotches	30.0	L
No. 3	Bowl-like galls on mugwort stem	10.4	G
No. 3	Mugwort leaf warty galls	9.6	F
No. 3	Oak apple galls	0	
No. 4	Chestnut twigs with chestnut galls	24.0	K
No. 4	Chestnut young terminal shoots with chestnut galls		
	Chestnut galls	9.6	H
	Young terminal shoots	13.6	H
No. 4	Chestnut young terminal shoots without chestnut galls+chestnut leaf minute, pitted spottings	0	M
No. 4	Young terminal shoots of *Quercus acutissima*+*Quercus acutissima* leaf minute, pitted spotting	10.8	N
No. 4	Oak leaf rolls+chestnut leaf red ball gall	0	A

the plant specimen changes. These values do not, of course, indicate the amount of P^{32} localized in some portions of each specimen, but merely its total amount present in each specimen. It is apparent, however, that there is a remarkable difference in radioactivity among the plants examined. Thus, according to the order of the magnitude of radioactivity, all the specimens may be arranged in the following grades:

> Chestnut galls>Oak knot galls>Oak leaf-rolls+oak leaf miner blotches>Cherry leaf aphid burns>Young terminal shoots of the chestnut trees with chestnut galls>Ball galls on oak leaf veins>Young terminal shoots of *Quercus acutissima* (a kind of oak)>Bowl-like galls on a mugwort stem>Warty galls on mugwort leaves.

If what Flanders (1953) calls the three types of incomplete parasitism in natural enemies are taken into consideration, some questions may be raised against this graduation, that is to say, a question of whether or not these grades really stand for the degree of the preference by the P^{32}-labeled parasitic wasps to these plants tagged. Furthermore, it must be taken account of that the contamination due to excreta as well as the honey sticked to the insect body may have had some effect upon the above result of labeling. Attention should be given at the same time to the possibility that considerable individual variations in the radioactivity of the tagged wasps may have affected the present result in a measure. About these questions detailed discussion will be given in the next section.

c) Analysis of macro-autoradiographical results

i) *Descriptive features*

The macro-autoradiographs obtained are shown in phots. V-X. The position of the left-hand side plant specimens just corresponds to that of their right-hand side macro-autoradiographs. It will easily be found out by inspection that there are 2 kinds of sensitized portions at least in the graphs; one is a tiny spot sharply sensitized and/or a pretty large spot comparatively strongly sensitized, the other being the portions more weakly sensitized, which lie around or between the former spots. Here let the former spots be called the sensitized spots of the first kind, and the latter spots the sensitized spots of the second kind.

The sensitized spots of the first kind can be found in such portions as correspond to the following specimens or portions:

Chestnut galls (phots. B, D, H, I, and K), oak knot galls (phot. E), cherry leaf aphid burns (phot. C), chestnut leaf miner blotches (phot. A), chestnut serpentine leaf miner winding trails (phot. J), buds of the young terminal shoot of the chestnut twigs with chestnut galls (phots. B, D, and H), warty galls on mugwort leaves (phot. F), bowl-like galls on mugwort stems (phot. G), ball galls on oak leaf veins (phot. J), minute, pitted spottings on a chestnut leaf (phot. K), oak leaf rolls (phots. J and L), oak leaf miner blotches (phots. A and L), minute, pitted spottings on a *Quercus acutissima* leaf (phot. N), buds of the young terminal shoot of *Quercus acutissima* (phot. N).

Of these specimes, the small-sized galls and the buds of the young terminal shoot correspond to tiny, sharply sensitized spots, while such large-sized galls as chestnut galls, oak knot galls, and bowl-like galls on the mugwort stem correspond to large, but rather unsharply outlined, sensitized portions. No tiny, sharply sensitized spots can be found at any portions other than those corresponding either to various kinds of galls, the buds of the young terminal shoot, leaf blotches and leaf pitted spottings, or to other injured portions caused by various kinds of piercing-sucking insect pests. If the above large, but rather unsharply outlined, sensitized portions are scrutinized, one or a few tiny, sharply sensitized portions can be detected in the interior of each one of them in most cases. Hardly any sensitized portions of the first kind, which are randomly scattered and indicative of contamination due to excreta by released wasps, can be seen at any portions of all the specimens. The sensitized portions of the second kind can be seen at such portions as correspond to twig specimens, leaf veins, some buds of the young terminal shoots and some leaf margins.

When compared with the magnitude of the readings of cpm. as given in table XVIII, these two kinds of sharply sensitized portions can be found to correspond exactly to the specimens whose radioactivity was strong, excepting some young terminal shoots of the chestnut twigs (phots. B, D, and M) whose radioactivity could not be detected with a G.-M. tube. Consequently, there is almost no room for doubt that these sensitized portions are certainly not an outcome caused by various kinds of background fogs. Needless to say, no such sensitized portions were found at all on the autoradiographs of the P^{32}-free control specimens.

In short, of the 15 sorts of plant specimens examined, the following 13 may be recognized as having been tagged with P^{32} by the P^{32}-tagged parasitic wasps which were caged with them:
Namely,

6 sorts of galls;
5 sorts of injured leaves;
2 sorts of young terminal shoots.

What attracts our special attention is that some of the tiny, sharply outlined, sensitized portions are found on the portions corresponding to some of the buds of the young terminal shoots of the chestnut twigs with chestnut galls and/or without them (phots. B, D, and M), whereas these specimens had no radioactivity when the counts of their radial rays were taken with the end-window of a G.-M. tube.

ii) *Biological interpretations*

It goes without saying that the sensitivity of any portion on an autoradiograph depends entirely upon the local amount of P^{32} present in the specimen corresponding to that portion. The sensitized portion of the first kind must therefore be an indication of localization of strongly concentrated P^{32} in the corresponding specimen. What is the origin of such local concentration of P^{32} ? What is conceivable first of all may be either oviposition of radioactive eggs or excretion of radioactive substances by the released wasps. If not so, it must be ascribed to the other mechanism, viz., to the accumulation of P^{32} translocated from the other portion that was directly tagged with P^{32}. In view of the whole course of the present experimentation, the latter case seems to be almost impossible. What needs to be clarified is the first case. Judging from their locality and the degree of their sensitivity, the sensitized portions of the second kind may be considered as an indication of indirect marking due to translocated P^{32}.

In order to clarify the first case, attention should be paid to the whole course of the present experimentation as well as the host seeking habit of the released wasps. What should be noticed first of all is that no chance of oviposition had been given to the female wasps before they were released into the experimental cage, and accordingly they were all capable of ovipositing, if only they could find any suitable host. The next point to be considered is the positions of the sensitized portions of the first kind, which correspond exactly to the parts suited for their oviposition under natural conditions, viz., to the positions just infested by their presumptive host insects, i.e., the insects parasitic on the plants examined. Putting these facts together with the results secured by many investigators regarding the radioactivity of the eggs laid by the P^{32}-tagged insects and mites such as described in section (3), i), a), of this chapter, it may safely be stated that the sensitized portions of the first kind must be interpreted as the result from the radioactive eggs laid by the released parasitic female wasps, one single tiny, sharply outlined, sensitized spot corresponding to one egg or a very few eggs deposited, and a large, rather unsharply outlined, sensitized spot corresponding to a lot of deposited eggs. The contamination due to the excreta expelled from the body of the released wasps should not, of course, be missed. Were some of the sensitized portions, however, to be produced as a result of contamination due to such excreta as faeces and vomits of the released wasps, such portions would be scattered more randomly over the plants submitted to examination, that is, over the leaves, twigs and young terminal shoots, because there is no knowing that parasitic wasps prefer only to such special portions, when they excrete. Thus, according to the autoradiographical sensitivity, viz., the sensitivity and the number of the sensitized portions, the following order of labeling intensity may be graded:

> Chestnut galls (1).>Oak knot galls (2).>Warty galls on mugwort leaves (9).≒ Oak leaf miner blotches (3).>Cherry leaf aphid burns (4).≒Buds of the young terminal shoots of *Quercus acutissima* (7).>Oak leaf rolls.≒Bowl-like galls on mugwort stems (8).≒Ball galls on oak leaf veins (6).>Chestnut serpentine leaf miner winding trails.>Buds of the young terminal shoot of the chestnut twigs with chestnut galls (5).>Buds of the young terminal shoots of the chestnut trees resistant to chestnut gall wasps.>Chestnut leaf pitted spottings.≒Minute, pitted spottings on a *Quercus acutissima* leaf.

This order differs considerably from that based on the readings of cpm., which is indicated with the figures in parentheses. But nothing is to be wondered at this order, since an autoradiograph is indicative of the radioactivity localized in some portions of each specimen, whereas the readings of cpm. merely indicate the strength of total radioactivity present in each specimen as a whole. Notwithstanding this relation, such specimens as chestnut galls, oak knot galls, oak leaf miner blotches and cherry leaf aphid burns still rank high in both orders. Particular stress should be laid on this fact. In this case, there is hardly any fear of making the P^{32}-tagged wasps lay their eggs compulsively on some unfavorable places to which they do not prefer, since various kinds of gall plants all available were caged with them, and accordingly they could have oviposited quite freely on anywhere they prefer, so far as they were endowed with the ability of discerning their host. If they should not have such ability, they would have deposited their eggs into any plants quite at random, and as a result all the plants caged with them would have been oviposited nearly evenly by them, since relatively a lot of parasitic wasps were released. Judging from the present results, however, such a case seems to be utterly out of the bounds of possibility. If this is tenable, the order of radioactivity discribed above may be regarded as suggesting the grade of the host preference by the releasd parasitic wasps. Thus, the following order in terms of the selected host insect may be accepted:

> *Dryocosmus kuriphilus* (×, pupae).>*Pelataea bicolor* Walsingham (♯; larvae). >*Rhopalomyia foliorum* Kieffer (+; larvae).≒*Tischeria complanella* Hübner (*; larvae).>*Phomyrus sasaki* Mats. (◎; larvae).≒Eggs of the unknown species of parasitic insect† laid on the buds of the young terminal shoot of *Quercus acutissima* (×).>*Phyllaphia korarae* Shin. (◎; larvae).≒*Rhopalomyia tubifex* Kieffer (+; lavae).≒A kind of oak gall wasp (×, specific name unknown, larvae).>A kind of serpentine leaf miner maggot (○, larvae, specific name unknown).>Eggs of *Dryocosmus kuriphilus* laid on the buds of the young terminal shoot of the chestnut

† This species has been submitted to Dr. K. Yasumatsu's identification.

twigs with chestnut galls (×).>Eggs of *Dryocosmus kuriphilus* laid on the buds of the young terminal shoot of the chestnut twigs without chestnut galls (×).> *Trizoa querci* Shin. (△; larvae).

These are classified into the following 10 species:

i) 2 species of gall wasps (marked with ×): Eggs, larvae, pupae; Fam. Cynipidae, Suborder Apocrita, Order Hymenoptera.

ii) 2 species of gall flies (marked with +): Larvae; Fam. Cecidomyiidae, Suborder Orthorrhapha, Order Diptera.

iii) 2 species of aphids (marked with ◎): Youngs; Fam. Aphididae, Suborder Homoptera, Order Hemiptera.

iv) 1 species of stem gall moth (marked with ♯): Larvae, Fam. Olethreutidae, Suborder Heteroneura, Order Lepidoptera.

v) 1 species of leaf miner moth (marked with *): Fam. Tischeriidae, Suborder Heteroneura, Order Lepidoptera.

vi) 1 species of a leaf miner fly (marked with ○): Larvae; Order Diptera.

vii) 1 species of tree louse (marked with △): Youngs; Fam. Psyllidae, Suborder Heteroptera, Order Hemiptera.

In this experiment, however, it could not be traced exactly which of the released parasitic wasps is ever responsible for an attack on any of these host insects. At any rate, the following conclusion may be drawn from the results so far achieved that in Kamiina District the 6 species, if not all, of parasitic wasps, which emerged from the chestnut galls by about the 10th of July, tend to deposit their eggs into the galls, or the injured leaves, caused by any of the above 10 species of parasitic insects, or into the buds of the chestnut young terminal shoots, into which the chestnut gall wasps are sure to have already laid their eggs.

ii) *Results obtained in 1959.*

In view of the results secured in 1958, the investigation was carried out principally with an eye to the elucidation of the following two points: i) To trace the host-preference by each species of parasitic wasps. ii) The relative relation between the duration of release and oviposition, especially the oviposition after a release of short duration.

a) Radioactivity of P^{32}-tagged parasitic wasps

The radioactivity was measured of the three species such as *Megastigmus* spp., *O. nigritibialis* and *A. amoenus japonicus* which are numerically abundant. The method and techniques are alike with those in the case of 1958. The readings of cpm. taken at the time of release and after the end of release are shown in tables XIX and XX. The values of cpm. are comparatively larger than those secured in 1958, since the counts of radial rays were taken at 0.5-cm. distance. In this case, too, it is clear that a female abdomen is relatively larger in cpm. as compared with a male one. Furthermore, evidence is clear that the loss of radioactivity in a female abdomen is far larger than that in a male one, even in the case of relatively short duration of release of 2 days. This tendency bears a striking resemblance to that observed in 1958. From this, the following conclusion may safely be drawn. a) The internal organs of the parasitic

TABLE XIX. Radioactivity (averages of cpm. taken at 0.5-cm. distance) of adult parasitic wasps after feeding for about 28 hours on a honey solution (well soaked up in matchsticks) containing 3 millicuries of radioactive P^{32} in 35 cc.

Species examined	Sex	Part examined: Before dissection, Intact body (cpm)	After dissection: whole body (cpm)	whole body (%)	Head (cpm)	Head (%)	Thorax (cpm)	Thorax (%)	Abdomen (cpm)	Abdomen (%)	No. of individuals examined
M. spp.	Female	143.6±3.2	149.3	100.1	18.5	12.4	60.4	40.5	70.4	47.2	3
	Male	170.0±8.4	167.6	100.0	19.4	11.6	88.8	53.0	59.4	35.4	2
O. n.	Female	76.9±7.5	78.5	100.0	3.4	4.3	26.3	33.5	48.8	62.2	4
	Male	61.2±8.6	63.4	100.0	6.4	10.1	31.6	49.8	25.4	40.1	2
A. a. j.	Female	81.2±3.6	79.4	100.1	21.4	27.0	26.8	33.8	31.2	39.3	2
	Male	116.4±5.2	125.6	100.0	26.4	21.0	69.2	55.1	30.0	23.9	1

TABLE XX. Radioactivity (averages of cpm. taken at 0.5-cm. distance) of adult parasitic wasps measured after 2 days release.

Species examined	Sex	Part examined: Before dissection, Intact body (cpm)	After dissection: whole body (cpm)	whole body (%)	Head (cpm)	Head (%)	Thorax (cpm)	Thorax (%)	Abdomen (cpm)	Abdomen (%)	No. of individuals examined
M. spp.	Female	157.2±2.8	163.4	100.0	28.2	17.3	72.6	44.4	62.6	38.3	9
	Male	158.1±4.0	169.7	100.0	41.7	24.6	77.0	45.4	51.1	30.0	3
O. n.	Female	89.9±5.6	93.4	99.9	10.0	10.7	34.6	37.0	48.8	52.2	5
	Male	85.5±6.9	83.2	99.9	10.6	12.7	39.2	47.1	33.4	40.1	3
A. a. j.	Female	133.0±3.8	142.9	100.1	25.1	17.6	59.4	41.6	58.4	40.9	5
	Male	97.8±7.1	96.6	100.0	16.6	17.2	60.2	62.3	19.8	20.5	1

wasps fed on P^{32}-tagged honey for 28 hours were fully labeled with P^{32}, and especially their ovary was strongly marked. b) The relatively great loss of radioactivity in a female abdomen after two-day release may be ascribed to the deposition of P^{32}-tagged eggs.

These results also show that the parasitic wasps released in the latter part of July are capable of ovipositing, even in the case of only two-day release, in so far as some adequate hosts are offered. It is quite natural that they could lay their eggs even at the lapse of nearly one month after they had emerged, since they are considered to belong to what Flanders (1950) calls a synovigenic species. It is to be regretted, however, that the ovipositing habit of these parasitic wasps fresh from emergence could not be traced entirely owing to the unavoidable circumstances pertaining to the present experiment.

At the time when the plant specimens were brought into contact with a X-ray film, the radioactivity was measured at 0.5-cm. distance of the parasitic wasps which were used as the control at the time of release. The results are given in the following table.

Species	cpm.		No. of individuals examined
	Female	Male	
M: spp.	79.2	84.6	5♀♀, 3♂♂
O. n.	43.2	44.9	5♀♀, 3♂♂
A. a. j.	68.0	49.0	5♀♀, 1♂

(As regards abbreviated specific names see table XV.)

The females are of course the not-yet-egg-laid ones. From this, it may safely be inferred that the P^{32}-labeled eggs laid by the released parasitic wasps must have had comparatively strong radioactivity at the time when autoradiographical preparations were made. This inference warrants the reliability of the results obtainable from autoradiographical preparations.

b) Radioactivity of the plants caged with the P^{32}-labeled parasitic wasps

The readings of cpm. of the dried plant specimens are shown in table XXI. The counts of radial rays were taken at the very time when autoradiographical preparations were made. Although their values are so small as indistinguishable from those of the natural background, the general trend in the radioactive strength can roughly be grasped. When viewed from readings of cpm., the general trend in the order of radioactive strength shown in the plant specimens examined seems to be roughly equal to that observed in the experiment in 1958.

The radioactivity of both oak knot galls and chestnut galls tends to become comparatively stronger than that of the other plant specimens in most cases. This is coincident with the tendency observed in 1958. It is to be noted, however, that, unlike the results secured in 1958, the value of cpm. of the oak scale gall specimen was found to be relatively strong as shown in the data obtained from cage C where a pretty many *A. amoenus japonicus* were released. Moreover, it seems undeniable that, newly adopted plant specimens such as bowl-like galls on willow leaf and burry galls on oak twig were also marked with P^{32}. Together with oak scale galls, these two kinds of plant galls should be added to the 13 sorts of host plants ascertained in the experiment carried out in 1958.

c) Macro-autoradiographical results

The results are shown in phots. XI-XIV. Generally speaking, the sensitized portions are not so sharp as compared with those shown in the data of 1958. This may be attributed to the weakness in radioactivity of the parasitic wasps tagged with P^{32}. It is quite similar to the results secured in 1958 that the two kinds of sensitized portions, viz., the sensitized portion of the first kind and that of the second kind, can be distinguished, though rather faintly. The difference in such local sensitivity shown in the autoradiographs permitted the detailed rating of the intensity of radioactivity in each plant specimen examined. The result was inserted in table XXI with marks * and +. As can be seen, the trend does not always coincide with that of the readings of cpm., as is usual with the experiment with radioactive isotopes. But

TABLE XXI. Radioactivity (averages of cpm. taken ten times for each specimen at nearly zero distance) of the dried plant specimens caged with the P^{32}-labeled wasps parasitic on the chestnut gall.

Plant specimens	Cages and released parasitic wasps			
	A (*O. n.*)	B (*M.* spp.)	C (*A. a. j.*)	D (*E. r.* & *P. f.*)
1) Oak knot galls	5.4*	0.4*	0.6*	5.8*
2) Oak scale galls	1.8*	—+	3.4*	—*
3) Chestnut galls	0.8*	—+	3.2*	3.2*
4) Chestnut young terminal shoots with chestuut galls	—+	—	—	—
5) Chestnut young terminal shoots withont chestnut galls	3.2*	—	—*	—*
6) *Quercus acutissima* galls	—*	0.4+	1.0*	4.2*
7) *Q. a.* young terminal shoots with *Q. a.* galls	—	—	0.4+	0.6+
8) *Q. a.* young terminal shoots without *Q. a.* galls	0.6*	0.2*	1.3*	1.6*
9) Mugwort stems with bowl-like galls	—+	—+	—+	1.2+
10) Mugwort leaves with warty galls	—	0.4*	—	—+
11) Mugwort leaves with aphid burns	—+	0.2+	—	—
12) Willow twigs with bowl-like leaf galls	—	—	—	2.6*
13) Burry galls on oak twig	—+	0.6+	—*	—+

Note: 1) All the plant specimens were covered with a sheet of damp-proof cellophane paper.
2) The line—denotes the case when cpm. was foudd to be nearly equal to that of the natural background.
3) The asterisk * denotes the case when some intensely sensitized portions were found on the autoradiograph concerned, the mark + denoting the case when the similar sensitized portions were found to be a little weaker than those in the former case.
4) As regards the abbreviated scientific names in parentheses see table XV.

both resembles with each other in their general trend, and by this fact the positions where eggs were laid by the released parasitic wasps can easily be traced systematically. It should be noted in this connection that some sensitized portions arising from contamination probably due to excreta in part as well as to sweat on finger tips in course of the X-ray film operation was discerned on some plant specimens.

Based on these results, we shall be able to trace the ovipositing habit of, or host preference by each parasitic wasp examined, since the release experiment was planned so as to be able to detect it, as previously described. What arrests our attention first of all is that the young terminal shoots of a chestnut tree, irrespective of a variety resistant or susceptible to the chestnut gall wasp, tend to be egg-laid rather slightly by the released parasitic wasps as compared with those of *Q. acutissima,* whereas evidence is clear that the former are more frequently egg-laid than the latter. This is the only point quite different from the results in 1958. This may be interpreted as follows. In this district, the period of emergence of the *Q. acutissima* gall wasp is earlier by about half a month than that of the chestnut gall wasp. The emergence of the former ends at the first part of July, its maximum period being at the end of June. At the time of a release experiment, viz., on and around 23rd July, therefore, the eggs laid by chestnut gall wasps on the chestnut buds must have been lagging pretty behind those laid by the *Q. acutissima* gall wasp on the buds of *Q. acutissima* in their development. Presumably, the above result may be ascribed to such difference in the developmental state of their eggs.

What should be paid attention to in the next place is that there seems to be relatively distinct differentiation in the ovipositing habit among the 4 species of P^{32}-tagged parasitic wasps. They tend to oviposit on the whole more frequently on woody galls than on herbaceous galls and/or leaf galls, especially so does *A. amoenus japonicus*. This fact seems to imply that *A. amoenus japonicus* tends to choose perenial woody plants in preference to annual herbaceous plants as its host plant, when it is compared with the other three species of parasitic wasps. If that is the case, this has an ecological interest a great deal, since *A. amoenus japonicus* is believed to emerge once a year, whereas the other three species to emerge at least twice a year. Furthermore, this seems to serve as a positive proof of the statement made by Dr. Yasumatsu (1955) that, from the angle of the utilization of the wasps parasitic on the chestnut gall wasp, the single-brooded ones are utilized against the scrubless pure forest of chestnut as well, while the double-brooded ones are more fitted to the liberation against the scrub forest. This finding seems to be very suggestive at the same time for the utilization as well as the conservation of these indigenous parasitic wasps, since they are produced relatively abundantly in this district, and are considered powerful natural enemies against chestnut gall wasps.

Species to be added to the 10 species of insects ascertained as a host in 1958 are as follows:

a) The species belonging to Fam. Cynipidae.

i) *Neuroterus* spp. which produces a scale gall on the male flower of a kind of oak tree, *Quercus grandulifera* Sieb.

ii) *Diplolepis japonica* Ashmead, which produces a burry gall on the twig of *Q. grandulifera.*

b) The species belonging to Fam. Tenthredinidae.

i) *Pontania viminalis,* which produces a bowl-like gall on the willow leaf.

To sum up the results secured in 1958 and 1959, the statement may be made that the plant galls, injured leaves, or the egg-laid buds of the young terminal shoot, which are attacked by the parasitic wasps on the chestnut gall, amount to 16 kinds in number, and the insects to be utilized as the host of these parasitic wasps to 13 species belonging to 7 Families and one Dipterous Order in Ina district.

(4) Discussion

a) From the viewpoint of parasite biology

What arrests our attention first of all is that there is almost undeniable evidence that the released parasitic wasps have concentratively laid their eggs on the chestnut gall. This finding suggests the possibility of either their progeny may emerge from the chestnut galls again in the autumn of the same year, or winter inside the gall as larvae. In fact, Dr. K. Yasumatsu (1955) pointed out affirmatively such a life cycle, in his studies on the wasps parasitic on the chestnut gall wasp. According to him, such a species as *Megastigmus* spp. could easily pass winter as adults, when it emerged in autumn. Furthermore, according to his unpublished data,* a species such as *Megastigmus* spp., *M. maculipennis, Peleumus ferrierei, Ormyrus nigritibialis* and *Eurytoma rosae* continue to emerge from the chestnut galls from the beginning of September up to the end of October, and of them 2 species belonging to genus *Megastigmus* have the longest period of emergence, ranging from the end of May to the end of October in the southwestern warm climates in Japan. Judging from the short period of their larval stage in the warmer season, it may be interpreted that the late emergence of the said parasitic wasps in the middle part of autumn in the warmer climates may be ascribed to the progeny issuing from the eggs laid by their parent females in the latter half of June; in other words, to the progeny in their second generation. This version seems to be warranted also by the fact that there is a break from about 10 to 30 days during August in the period of their emergence for such species as *Peleumus ferrierei, M. maculipennis,* and *Ormyrus nigritibialis,* as is shown in the unpublished chart of their seasonal occurrence constructed by Dr. Yasumatsu (1958). In my release experiment carried out in 1958 and 1959, too, the same 4 species as above-mentioned, excepting *M. maculipennis,* were employed, and *M.* spp., *O. nigritibialis* were comparatively large in individual number among them. If that is the case, it may be said that the above finding concerning the intensely P^{32}-tagged chestnut gall affords us a positive evidence for the statement made by Dr. Yasumatsu (1955) and the above version of his unpublished data (1958). In the experiment conducted in 1959, evidence has been produced that *A. amoenus japonicus* also lays eggs relatively concentratively on the chestnut gall. According to Dr. Yasumatsu's unpublished data (1958), no record was reported that this species emerges in autumn. It is sure, therefore, that this species winters even inside the chestnut gall. Then, it may safely be concluded that the 4 species at least such as *Megastigmus* spp., *O. nigritibialis, Eurytoma rosae,* and *A. amoenus japonicus* are responsible for the greenish galls amounting to about 10 per cent in this district.

In short, it may be said that the pretty hardened chestnut galls, within which the gall wasps are probably either in a state of matured larvae or in the pupal stage, tend to be utilized again during summer for the second-generation host of these parasitic wasps issuing from them early in summer. From the viewpoint of the preservation of native parasitic wasps, therefore, utmost care should be taken to the prevailing control measure against the chestnut gall wasp, since it rests mainly on a stopgap measure of plucking galls and burning them in summer season. This interpretation will furnish us with a new basis to the biological control aided by the resident parasitic wasps, and at the same time will tell us the need to improve the prevailing control measure, since, as previously mentioned, these species are most abundant in individual number in this district. From the angle of parasite biology, it should be noted that it is suggested by the above finding that these parasitic wasps attack not only the larvae but also the pupae of the chestnut gall wasp. It must be added further

* The report submitted to the Ministry of Agriculture and Forestry in 1958.

that there is almost no fear of what Flanders (1953) calls three kinds of incomplete parasitism being involved in this case. The reason is as follows. Since all of these parasitic wasps issued evidently from chestnut galls, they are naturally able to attack the chestnut galls voluntarily under natural field conditions.

The second point to be paid special attention to is that the buds of the chestnut young terminal shoot tend to be undoubtedly attacked by the released parasitic wasps indiscriminately, irrespective of a variety resistant or susceptible to the chestnut gall wasp. Judging from the ovipositing habit of the released parasitic wasps, this must be an indication that some of the chestnut buds examined, resistant or susceptible to the chestnut gall wasp, had already been egg-laid by the chestnut gall wasp before they were subjected to this release experiment. This is quite probable. Why so? Because of two reasons at least. One is that the chestnut gall wasps must have been active in seeking their host, since the larger part of their larvae must have already been in a matured state on the day when the sample specimens were collected, as can be seen in Figs. VI and VII. The other is that, as has been recognized by Yokoyama and Kinoshita (1951) and by Nitto and Shimizu (1954), respectively, the chestnut gall wasp lays its eggs in most cases on both resistant and susceptible varieties indiscriminately. Thus, it neccessarily follows that the chestnut gall wasp tends to be attacked by its parasitic wasps not only in the larval stage but also equally in both the egg and the pupal stage, since, at that time of the season, most of the eggs laid by the chestnut gall wasp can not yet have been hatched out, but must have remained in the egg stage. This means so much increase in the range of economic utilization of these parasitic wasps for the control of the chestnut gall wasp.

It may be interpreted in like manner that the buds of the young terminal shoot of *Quercus acutissima* had also pretty strong radioactivity. In this case, however, the scientific name of its gall wasp is not yet known, but it is certain that the wasp is quite different from the chestnut gall wasp. There is hardly any doubt that these buds had already been attacked by this gall wasp, and that on such eggs the released parasitic wasps must have laid their eggs. Two species of parasitic wasps* were found to emerge from the galls on *Q. acutissica* twigs, one bearing striking resemblance to *E. rosae,* and the other to *O. nigritibialis.* This has an ecological interest, because the latter two species tend to lay eggs relatively concentratively on the buds of *Q. acutissima* young terminal shoots. (Table XXI.)

In the third place, allusion should be made particularly to the fact that the parasitic wasps employed, if not all, are capable of selecting comparatively various kinds of hosts. As already shown, their hosts amounted to about 13 species of parasitic insects, and the plants infested by them can easily be found everywhere in the scrubs and the forests. This is quite nothing to be wondered at, if it is taken into consideration that they had inhabited here as indigenous wasps parasitic on various kinds of gall, or parasitic, insects, before the chestnut gall wasp invaded here. From the angle of the conservation of resident parasitic wasps, however, it is of great importance to have clarified positively that they utilize pretty many species of ordinary parasitic insects for the host in their second generation, to say the least of it,

* These are submitted to Dr. Yasumatsu's identification together with their host wasp.

immediately after they emerge from the chestnut galls. This finding is nothing but a positive proof of the statement made by Dr. Yasumatsu (1955) that scrub forests are generally fitted better than the pure forests for the liberation of a double-or more-brooded natural enemy.

Finally, but not least, the question which needs to be considered may be whether the parasitic wasps employed in this investigation belong to what Flanders (1950) calls the pro-ovigenic species or to the synovigenic ones. According to my small experience both in 1958 and 1959, the species that lived longest of all the six species employed were *M.* spp. and *O. nigritibialis* which emerged late in summer. The record of the longevity of these species amounted to more than 2 months, while the other species lived for less than one month, the minimum being 11 days, when they were allowed to feed on honey. Their life is surely by far the longer than that of the chestnut gall wasp which, acording to Nohara (1956), belongs to the pro-ovigenic species; the latter lived no more than about 14 days under the same feeding conditions. From this, I should like to think that these parasitic wasps belong to the synovigenic species, just like some other species demonstrated by Dr. Yasumatsu (unpublished data). The release experiment with P^{32}-taggd parasitic wasps in 1959 is also another positive proof for this interpretation. If this is tenable, it may be said that the utilization of these native parasitic wasps gives great promise for the success in biological control of the chestnut gall wasp.

b) Chemical control combined with biological control

1) The present situation

In taking a certain chemical control measure, if any, the question of how to combine it with the utilization of natural enemies is of great importance in the field of biological control. A lot of difficulties are involved in this question, and are left not yet overcome.

As is generally acepted, it is a weakness common to most natural enemies that their propagation commences later in general than that of their host insect pest. Naturally, they are very few in number as compared with their host, being unable to exert much influence upon the host at the period from the initial to the most active outbreak of the host. Such a time-gap is, so to speak, a fatal one between the periods of propagation of the two. In order to bridge this time-gap, some proper measures, such as chemical control or a removal of infested trees, must be adopted. As emphasized by Dr. Yasumatsu (1958), however, such a measure should of course be employed only as a so-called stopgap one, and be replaced at a proper time with the control based on the activity of the natural enemies, since the natural enemies are sure to increase in number some time later, necessarily acquiring an ample scope for their activity. The case of the resident natural enemies parasitic on the chestnut gall wasp cannot be of course an exception as well. In this case, therefore, a chemical control measure available, if any, should be adopted from this standpoint, and never be used habitually. We should always bear in mind the warning given by Dr. Yasumatsu (1958) that the habitual use of insecticides without due reflection will surely result not only in the waste of cost but also in mere killing of the natural enemies that are propagating vigorously. When only combined with biological control as a stopgap measure, the chemical control of the chestnut gall wasp will be compatible with the utilization of its natural enemies. Needless to say, "a timely application of a proper dose" is the secret of success in this case, too.

On the other hand, it is actual that some chestnut growers have been earnestly desirous of any more immediately effective measures, partly due to their being unable to stand the trouble involved in plucking galls, and partly due to the difficulty in mass introduction of effective parasites. Circumstances at Shishiko area in Dejima Village, Ibaraki Prefecture, are just the case in point. The area is one of the most famous chestnut-producing centers in Japan. Under the stress of unavoidable necessity, the chestnut growers thereat devised a certain chemical control measures, and have had recourse solely to it these three years, although they have apparently taken a wrong course when viewed from the angle of rational chemical control above-mentioned. They applied γ-BHC 3 per cent dust to their chestnut groves at the rate of 2 kg. per Tan (about one forth of an acre). After making an inspection of their groves one year after it was carried out, viz., in the summer of 1958, I could not help feeling that there is a great need to re-examine the effect more carefully. Probably, the effect of γ-BHC upon the chestnut gall wasp may be enormously great, since parasitic insects are, as a rule, seriously affected when exposed to contact insecticides, as pointed out by Graham (1952). But at the same time, so may it be upon the wasps parasitic on the chestnut gall wasp as is generally accepted when organic chlorinated insecticides are applied. This is a persistent disadvantage which we often encounter in the application of chemical control. According to the present results, however, it seems to be not neccessarily impossible to overcome such a difficulty, if only the timing of dusting is proper, because it has been traced autoradiographically that the female wasps parasitic on the chestnut gall wasp tend to oviposit in all available kinds of gall insects comparatively soon after emergence, when only conditions are favorable for oviposition, although most of them belong to a long-lived, and probably to a synovigenic species. The problem of timing of an application has clearly been solved, since it resolves itself down to how to predict what I call the "period fittest for biological control". Another problem to be solved is the systemic action of γ-BHC. By many investigators, such as Questel and Connin (1947), Hoskins (1949), Kozlova (1950), Starnes (1950), Casida and Allen (1952), Terrier and Ingalsbe (1953), Haines (1956), Foster and others (1956), Lilly and Fahey (1956), and Koshihara and Okamoto (1957), there has been reported that γ-BHC which was applied to soil is capable of being translocated comparatively easily to the body of the crop plants grown on the treated soil. As regards the rate of dosage which is effective in killing the insects living inside the body of the crop plants, however, their results are not always consistent with one another. On the same problem concerning woody plants, hardly any reports have hitherto been published, so far as I am aware. According to Dr. Ishii and coworker's achievements* recently produced (1958), however, the systemic action or the translocation of γ-BHC seems to be practically negligible,

* Presented at the annual meeting of the Japanese Society of Applied Entomology and Zoology held at Tokyo, April 5, 1959.

when it is sprayed upon the crop plants, or the crop plants are dipped into its emulsion. His conclusion rests on the results obtained from the experiment with the rice plant and the soybean plant by using radioactive γ-BHC-1-C^{14}. Although it does not directly concern itself with woody plants, his finding furnishes a reliable clue to the solution of the second problem with which we are confronted.

2) An experiment on dusting or spraying γ-BHC

The effect of γ-BHC on the chestnut gall wasp was investigated in 1959. This insecticide was applied in the following manner. The application of γ-BHC 1% dusts was made at the rate of 3 kg. per Tan (about one forth of an acre), and 70 grams of γ-BHC 5% wettable powder was diluted in 10 liters of water. In the application of these insecticides, it was assumed that the chestnut grove is planted with chestnut trees at the rate of 12 per Tan on the average, each tree having about 200 galls weighing about 280 grams on the average.

i) *Application to the chestnut galls collected in the early season.*

The galls were collected at Mikoshiba in Kamiina on 18th June, 1959. To 112 grams of them, γ-BHC 1% dusts were applied, and 100 grams of them were sprayed with the suspension of γ-BHC 5% wettable powder. After the treatment, each set of galls were kept in the paper bag separately. The number of the emergents was counted every day in principle.

Not an insect, including the chestnut gall wasp and its parasitic wasps, was observed to have emerged from each set of galls within the period from 18th June to 20th August. This result could not be altered by the observation carried out after that time till the middle part of December.

Some sample galls were selected randomly from each set of galls, and were dissected carefully in the middle part of October. Through this dissection, a very interesting fact was revealed. Namely, nearly all the chestnut gall wasps and their parasitic wasps were found dead in an adult form. Furthermore, it was found that they burrowed their exit hole up to the portion within about 0.5 mm. of the surface of the gall to die there. In such a case, the exit hole was filled with frass or woody refuse, which was produced as a result of their burrowing the way to the outside. (phot. XVI.)

ii) *Application to the chestnut galls collected at the maximum period of emergence of the chestnut gall wasp.*

Some sample galls were collected on 18th July, viz., at the predicted maximum period of emergence of the chestnut gall wasp, at Mikoshiba in Kamiina. (The period was determined to have been 16th July in reality.) Dusting was made against 90 grams of them, spraying being made against 100 grams of them, respectively, on that day. After the treatment, each set of the galls were kept in a glass jar with a cap netted with the brass net of 50 mesh. (phot. XV.) The observation was of course carried out every day.

The results are as follows:

In the case of spraying experiment, 2 or 3 chestnut gall wasps (the maximum number of them being 6) were found to have emerged from the pile of galls for an initial few days, although most of them were of course found dead. After that initial period, no individuals were found to emerge until the middle part of December. In the case of dusting, not an individual was found at all to have emerged from the start of observation to its finish.

The state of the insect inside the gall is substantially the same as that observed in the preceding experiment. Needless to say, far greater part of the gall insects were found dead in the adult state as compared with those observed in the former experiment.

iii) *Discussion and conclusions*

In the above experiments, it was revealed that almost all the chestnut gall wasps inside the gall were found dead at the portion about 0.5 mm. beneath the surface of the gall, when the galls were treated with γ-BHC. This is to be interpreted that the wasps must have died of contact with γ-BHC at that portion without completing the way to the outside. The reason is that, if the wasps should have died of the direct action of γ-BHC that had penetrated far deeply into the interior of the gall, they could not have been in the perfectly adult stage and could have been unable to burrow their exit hole as observed in the experiment made in the early season, since the galls were collected 13 days earlier than the first appearance of the wasps. It is true that some were found dead in the pupal stage. But, in such a case, their chambers are found to be situated, almost without exception, at the portion very near to the surface layer of the gall which was occupied by comparatively large number of wasps. The case is similar in the later season experiment. If this inference is tenable, it may be said that γ-BHC, irrespective of its dusts or suspension of its wettable powder, is able to penetrate barely into the tissue of the chestnut gall up to the depth of about 0.5 mm. from the surface of the gall, when it is applied to at the rate or standard of dilution above mentioned. Such as action of γ-BHC seems to be more powerful, which it is dusted, as is demonstrated in the experiment (ii) of this section.

This is very suggestive of the action of γ-BHC. On this point, further investigation will be published in the near future, since the autoradiographical experiment on this problem is now in progress with the use of γ-BHC-1-C^{14}.

Now, if the above interpretation is supported, the following inference may be drawn. Namely, even though the chestnut gall wasp inside the gall should have been parasitezed after the middle of July by some of its parasitic wasps which emerged early in July, the eggs laid by its parasitic wasps may probably be able to escape the action of γ-BHC, since the residual effect of γ-BHC is considered not to continue so long as the eggs develop into imagoes. Presumably, this inference may be applied in general to the eggs laid by the parasitic wasps in the other plant galls. If so, the way to the combined use of γ-BHC and resident wasps parasitic on the chestnut gall wasp may be opened, if only the proper time of applying γ-BHC is determined.

iv) *Recommended control measures*

From the results so far obtained, the following two may be conceivable as stopgap chemical control measures. One is the dusting γ-BHC which is applicable to the grove of cultivated chestnut trees. The other is that applicable to the prevailing control measure by burning the plucked chestnut galls. In the former case, the results obtained from the preceding experiments and Dr. Ishii's finding seem to warrant the safety of the measure. The reason is as follows. The γ-BHC sprayed or dusted does not exert any serious influence upon the eggs that the parasitic wasps deposited into various kinds of hosts in their second generation, since there is hardly any fear that such γ-BHC may deeply pass into the interior of the galls, or indirectly be translocated from any other portions of the treated plants to the interior of the galls, insofar as a proper dose of it is applied timely. In the latter case, the possibility is suggested of the solution of a new question raised in the prevailing control measure of the chestnut gall wasp. It is meant by a new·question here that, as previously discussed, the parasitic wasps in question tend to choose again the chestnut galls as their host after emergence. From the angle of protecting the native parasitic wasps, therefore, a new step must be devised to cope with this situation; in other words, it must be considered how to make their eggs escape from being killed after the galls were plucked. This may be overcome if only the collected galls are treated with γ-BHC, instead of their being committed to the fire. Namely, it consists of the following two steps; (i) the galls are gathered at, or shortly before, the "period fittest for biological control"; (ii) a proper dose of γ-BHC is dusted thoroughly upon the pile of collected galls once immediately after the collection was over, the pile being left at that condition within the airy walls until the end of autumn at least. In this case, precautions should be taken against rotting of the galls. If these steps are carried out successfully, the eggs newly laid by the parasitic wasps into chestnut galls will probably not be affected by γ-BHC, since its systemic action, if any, seems to be practically negligible. Needless to say, it is sure that the chestnut gall wasps issuing from the galls may be killed extensively. There is, however, hardly any fear of the residual effect of γ-BHC upon the eggs of the parasitic wasps inside the gall, because it takes more than two months at least till they develop into adults, and its residual effect does not last for so long a time. About the end of autumn, the second-generation parasites will emerge from such collected galls. Then, a proper step should be taken to let them go out the walls. It should be noticed in this case that there is not without some apprehension that the eggs may be affected in some measure by gaseous γ-BHC which may possibly penetrate into the interior of the gall through the exit holes burrowed by their parent groups as well as the other host gall wasps. This problem remains to be solved.

SUMMARY AND CONCLUSIONS

In recent years the chestnut gall wasp has become a serious pest of the chestnut trees, wild or cultivated, in Ina district. So far, the standard recommended measures to control this pest have been centred on burning the galls that were picked almost recklessly off the infested trees.

In the present studies, attempts have been made, from the standpoint of biological control, to devise the method of conserving the indigenous natural parasites as much as possible and to find out the biological control measure aided by such indigenous natural enemies, which is applicable to any infested areas. The gists of the studies are summarized as follows:

(1) The 10 species that are classified into 5 families belonging to the superfamily Chalcidoidea and one unidentified Dipterous parasite have been found to be the natural enemies parasitic on the chestnut gall wasp inhabiting Ina district. These belong to the eleven effective natural parasites discovered by Dr. Yasumatsu. Of these 10 species, *Megastigmus* spp. and *Ormyrus nigritibialis* have been found to be overwhelmingly predominant in relative population density in this district. If these species can be regarded as powerful natural enemies most adapted to this district, their utilization and positive protection will make a great contribution to the biological control of the chestnut gall wasp in this district.

(2) On the basis of the results derived from the indoor data which were secured for 5 years from 1955 to 1959, the biostatistical analyses were carried out against the cumulative percentage emergence curves concerning the chestnut gall wasp and its composite parasitic wasps.

The logistic curves well fitted to the cumulative percentage emergence curves for both have all been recognized as substantially uniform in shape with one another. From this, it has been concluded that, when taken as a whole, fluctuations in the trend of yearly emergence for both are substantially uniform in type, regardless of years and districts.

(3) In all the districts observed, the date of the first appearance of the emergents for the composite natural parasites has been estimated at about 20th June, and that for the chestnut gall wasp at about 1st July every year. A large majority of chestnut gall wasps end their emergence by about 5th August at the latest every year, about 10-15 days later than the composite natural parasites do. The composite natural parasites emerge about 14 days earlier than their host gall wasp does, completing their emergence as many days earlier than the latter.

(4) For the analysis of data, a logit method has been adopted anew. As compared with the usual probit method, the logit line of least squares based on the transformation of the cumulative percentage emergence into logits furnishes the most reliable step for determining what the writer calls "the period fittest for biological control" aided by the native natural enemies parasitic on the chestnut gall wasp.

(5) On the logisticity of the cumulative emergence curve for the composite natural parasites as well as for the chestnut gall wasp, a new version founded upon biological considerations has been put that any logistic curve concerned may be regarded as a resultant of various normal curves which differ with one another in the range of distribution and the size of population, each normal curve representing the percentage emergence peculiar to each locality characterized by its unique habitat conditions. A schematic diagram has been presented analytically, and evidences are adduced in support of this new version.

(6) The "period fittest for biological control" of the chestnut gall wasp in Ina district has been determined by the logit 75% date. It has been estimated as ranging from 4th July to 12th July with 8th July as centre. By this period, the composite natural parasites end about 75% of their emergence, while only about 10% of all

the chestnut gall wasps emerge. If some adequate control measure is, therefore, taken against the chestnut gall wasp on and after this period, it will perhaps be possible to destroy a large majority of chestnut gall wasps, protecting all the indigenous natural parasites having emerged by that period, which amount to about 75% of all their emergence in that season.

(7) From the theoretical inference, it has been suggested that, if some adequate control measure is adopted substantially on and after the logit 75% date, the percentage of parasitism of the composite natural parasites to the host gall wasp can be increased up to about 7.5 times as much as that under natural conditions, and up to such an extent as high as 100 per cent or more in the very year when such a control measure is taken.

(8) From the analyses of the data derived from the outdoor emergence trap, it has been concluded that the information of the data obtainable from an outdoor emergence trap is invalid as compared with that of indoor data in the analyses of the cumulative percentage emergence curve concerned, on account of the former data being seriously affected by some predators.

(9) The method of prediction of the "period fittest for biological control" of the chestnut gall wasp has been devised. It rests on the logit line of least squares based on the trend of the early stage of emergence of the composite natural parasites.

(10) Considerations have been given to the advantages of the logit method. The generalized logit method applicable to skew logistic curves has been presented and discussed.

(11) Attempts were made to introduce an effective parasitic wasp, *Torymus beneficus* Yasumatsu, into some heavily infested areas in Shimoina District for three years from 1955 to 1957. Taking into consideration the number of the whole galls formed on a unit twig sample as well as that of the galls remaining greenish in autumn, the liberation of this parasitic wasp was evaluated to have been strikingly effective in the first and the second years of liberation. In the autumn of the second year, the number of galls per unit twig sample was about 1.5, on the average, being reduced to about 4.3% of that in the first year; and about 91.2% of it was found to be greenish in colour indicating that most of the galls had been parasitized by the introduced *Torymus beneficus* Yasumatsu. This species belongs to what Clausen calls an "effective natural enemy", and further to what Sellers calls the species that is slow in its dispersion, in this district, too.

(12) *Torymus beneficus* Yasumatsu introduced into Ina district has, however, tended to show a comparatively rapid diminution in its propagation in the case where its subsequent liberation is interrupted. Full discussion has been given to this phenomenon from every angle conceivable, and great importance has been attached to the adverse effect of the heavy frost late in season on the propagation of this parasitic wasp introduced into this district. What the ultimate establishment of this introduced parasitic wasp will be in this district remains to be seen.

(13) From the practical standpoint, the following line of liberation has been recommended: a considerable number of *Torymus beneficus* Yasumatsu, which are divided into small groups, should be released scatteredly at several localities throughout

the infested area at least once every other year until the pest wasp population becomes checked down to below the level of economic density limits.

(14) With the object of finding out the method to ensure the protection of native natural enemies parasitic on the chestnut gall wasp, their egg-laying habit was pursued with the radioactive phosphorus P^{32} as a tracer. 6 species of parasitic wasps issuing from the chestnut galls early in summer were allowed to feed on honey containing 3 millicuries of P^{32}.

(15) No individual of the parasitic wasps allowed to feed on radioactive honey was found to be untagged. The reading of cpm. for some sample species varied to a considerable extent. In general, *Megastigmus* spp. tended to be more strongly tagged than the other small sized species did, presumbaly because of its comparatively large body size. When the body was dissected, radioactivity was the strongest in the abdomen, the next in the thorax with legs and wings, and the weakest in the head.

(16) 6 species of P^{32}-labelled parasitic wasps including both sexes were caged with several kinds of plant galls which were collected at the chestnut gall wasp infested scrub forests, and were allowed to lay their eggs on the caged plant specimens. As remarkable decrease in radioactivity in the abdomen was revealed at the time when the release experiment was over, it has been inferred that the caged P^{32}-labelled parasitic wasps, if not all, must have deposited their radioactive eggs on the plant specimens subjected to their selective oviposition.

(17) Evidences are adduced in support of the above conclusion by the results from macro-autoradiographs as well as by the radioactivity of the plant specimens caged with the P^{32}-tagged parasitic wasps. On the basis of these evidences, it has been concluded that, on emerging from the chestnut galls early in summer, six species of parasitic wasps examined, if not all, tend to lay their eggs on 16 sorts, at least, of galls and injured leaves caused by 13 species of phyto-parasitic insects belonging to 7 families and one Dipterous order. A definite gradient has been observed of the degree of host preference by these parasitic wasps. A single-brooded species such as *A. amoenus japonicus* tends to choose perenial woody plants in preference to annual herbaceous ones as its host plant, whereas double-brooded species such as *Megastigmus* spp., *O. nigritibialis,* and *Eurytoma rosae* take annual herbaceous plants for preference as well.

(18) It has been evidenced that the chestnut galls examined, in other words, the chestnut gall wasps in the pupal stage, must have been oviposited most abundantly by the six species of parasitic wasps caged with them. This finding indicates that there is much possibility that, under natural conditions, too, the chestnut galls are apt to be oviposited abundantly again by the native natural parasites issuing from them early in summer, and consequently that the utmost care should always be taken to be prudent in disposing of the chestnut galls collected in such a season, in order to protect the native natural parasites as much as possible. The 4 species at least such as *Megastigmus* spp., *Ormyrus nigritibiglus, Eurytoma rosae* and *A. amoenus japonicus* have been judged to be most responsible for such oviposition, and consequently for the formation of the chestnut galls remaining greenish in colour in autumn in this district.

(19) It has been revealed that the chestnut gall wasps inside the gall were found dead at the portion about 0.5 mm. beneath the surface of the gall, when the galls were applied with γ-BHC. This is interpreted that the wasps must have died of contact with γ-BHC at that portion without completing the way to the outside. From this, it has been concluded that γ-BHC penetrates barely into the tissue of the chestnut gall up to the depth of about 0.5 mm from the surface of the gall, when it is applied to at the usual rate or standard of dilution.

(20) In view of the ovipositing habit of the parasitic wasps examined, any application of any systemic insecticide should be avoided as much as possible in order to prevent the eggs laid by the resident parasitic wasps in the host galls early in summer from being affected by the action of the insecticide applied. If this precaution is taken to the full and what the writer calls the "period fittest for biological control" is utilized, it will perhaps not necessarily be impossible to expect success in combined usage of the chemical control and the biological one aided by resident natural parasites, so far as the cultivated chestnut grove is concerned. Spraying, or dusting, of γ-BHC seems to be capable of answering this purpose, since there is almost no fear of a systemic action of γ-BHC, as has been affirmed by Dr. Ishii and coworkers by the experiment with the rice plant and others by using γ-BHC-1-C^{14} and by the present experiment on the application of γ-BHC.

(20) As the stopgap chemical control which can be combined with biological control, a new control measure has been presented. It consists of the following two steps: (i) To gather the chestnut galls at, or shortly before, the "period fittest for the biological control of the chestnut gall wasp". (ii) To dust thoroughly a proper dose of γ-BHC upon the pile of collected chestnut galls once immediately after the collection was over, the pile being left at that condition within the airy walls against rotting until the end of autumn at least so that the second-generation parasites may emerge completely from the collected galls.

LITERATURE

Allee, W. C., P. Orlando, A. E. Emerson, T. Park, and K. P. Schmidt. (1950): *Principles of animal ecology*. pp. 301-315, W. B. Saunders Co.

Babers, F. H. and C. C. Roan. (1954): Distribution of radioactive phosphrous in susceptible and resistant house flies. Jour. Econ. Ent. 47(6) pp. 973-980.

Babers, F. H., N. Mitlin, and T. J. Shortino. (1956): The fate of radiophosphorus ingested by house flies. Jour. Econ. Ent. 49(6) pp. 820-823.

Berkson, J. (1951): Relative precision of minimum chi-square and maximum liklihood estimates of regression coefficients. Proc. Second. Berkelay Symposium on Mathematical Stat. and Prob. pp. 471-479. California.

———. (1944)*: Application of the logistic function to bio-assay. J. Amer. Stat. Assn. 39 pp. 357-65.

Clausen, C. P. (1951): The time factor in biological control. J. Econ. Ent., 44(1) pp. 1-9.

DeBach, J. (1958): The role of weather and entomophagous species in the natural control of insect populations. Jour. Econ. Ent. 51(4) pp. 474-484.

Dews, P. B. and J. Berkson. (1954): On the error of bio-assay with quantal response. *Statistics and Mathematics in Biology*. Chap. 27. pp. 361-370. Iowa State Coll.

Esaki, T. et al. (1951): *Iconographia Insectorum Japoniorum.* Hokuryukan.

Esaki, T., S. Isshiki, et al. (1957): *Genshoku nihon garui zukan* (Jo) p. 64. Hoikusha.

Fang, S. C. and D. Allen. (1955): Distribution and incorporation of radioactive phosphorus in the Douglase fir beetle. Jour. Econ. Ent. 48(1) pp. 79-82.

Fisher, R. A. (1950): *Statistical Methods for Research Workers.* Chaps. V-VI. pp. 114-210.

Flanders, S. E. (1952)*: The deposition of nonviable eggs by Hymenoptera. Jour. Econ. Ent. 35 pp. 283.

———. (1942)*: Oosorption and ovulation in relation to oviposition in the parasitic Hymenoptera. Ann. Ent. Soc. Amer. 35 pp. 251-266.

———. (1950): Regulation of ovulation and egg disposal in parasitic Hymenoptera. Canadian Entomologist. 82 pp. 134-140.

Foott, W. H. (1954)*: Phosphorus-32 labeling of the adults of the cabbage maggot, *Hylemyia brassicae* (Bouche) (Diptera: Anthomyiidae). 85th Rep. Ent. Soc. Ont. pp. 56-61. (R.A.E.: A, 45(9) pp. 344-345, 1957).

Foster, A. C. and Others.* (1956): Some effects of insecticide spray accumulations in soil on crop plants. Tech. Bull. U. S. Dep. Agr. no. 1149. 36 pp. (R. A. E: A. 45(10) pp. 408-409, 1957.)

Fukuda, J. and S. Okudai (1950): Kuritamabachi ni tsuite (On the chestnut gall wasp.) Appl. Ent. 6(1). p. 18. (in Japanese.)

Fuller, R. A., P. W. Rieger and J. W. T. Spinks.* (1954): Persistence of radio-activity in grasshoppers (Acrididae) tagged with phosphorus-32. Cand. Ent. 86(5) pp. 201-203. (R. A. E.: A. 44(4) pp. 109-110, 1956.).

Graham, S. A. (1952): *Forest Entomology.* Chaps. XI-XII. pp. 147-164, McGraw-Hill.

Himoto, K. (1957): Kyotofu Ka niokeru kuritamabachi no tenteki to gooru no suii. Shinrinboeki news. 6(4) pp. 89-90.

Hoffman, R. A., A. W. Lindquist and J. S. Butts. (1951): Studies on treatment of flies with radioactive phosphorus. Jour. Econ. Ent. 44(4) pp. 471-473.

Imms, A.D. (1937): *Recent advances in Entomology.* (Translated into Japanese by Ishikura, H. and M. Fukaya. (1943). Chap. XIII. pp. 334-427).

Ishii, S. (1958): *Agricultural Chemicals.* Chap. II. 17-87, Asakuka. (in Japanese.)

Kawada, K. and F. Maruyama. (1951: *Mathematical Statistics.* pp. 90. χ^2-Chart. Shokabo. (in Japanese.)

Kitagawa, T. and M. Masuyama. (1952): *Tokeisuchihyo* (Statistical tables) pp. 186-187. Kawade. (in Japanese.)

Koehler, C. S. and G. G. Gyrisco. (1957): The systemic action of lindane in alfalfa upon the meadow spittlebug. J. Econ. Ent. 50(3) pp. 346-347.

Komoto, K. (1957): Ishikawa-ken ka niokeru kuritamabachi tenteki ishoku no jokyo. Shinrinboeki news. 6(4) pp. 84-87.

Koseki, Y. and others. (1956): Experimental Technique on Autoradiography. 105-147. *Experimental Technique with Isotope.* No. 2. pp. 105-147. Nankodo. (in Japanese.)

Koshihara, T. and D. Okamoto (1957): Control of rice stem borer by the application of BHC dust in the paddy field soil. Jap. Jour. Appl. Ent. Zool. 1(1) pp. 32-35 (in Japanese with a summary in English.)

Lawson, F. R., G. B. Lucas, and N. S. Hall. (1954): Translocation of radioactive phosphorus injected by the green peach aphid into tobacco plants. Jour. Econ. Ent. 47(5) pp. 749-752.

Lilly, J. H. and J. E. Fahey. (1956): Translocation of BHC from high-dosage soil treatements applied before planting. Jour. Econ. Ent. 49(6) pp. 815-818.

Lindsay, E. and C. Craig. (1942): The distribtuion of radio-phosphorus in wax moth, meal worm, cochroach and fire brat. Ann. Ent. Soc. Amer. 35 pp. 5-6.

Jones, S. C. and L. Wallace. (1955): Cherry fruit fly dispersion studies. Jour. Econ. Ent. 48(5) pp. 616-617.

Masuyama, M. (1948): A graphical method of estimation of parameters contained in the Langevin's non-central stochastic equation. Study of Math. Stat. 12(2). pp. 28-32. (in Japanese with a summary in English.)

Masuyama, M. (1950): *The Way to Stochastics.* pp. 103-104. Kawade. (in Japanese.)

———. (1953): *How to treat small Samples.* pp. 195-203. Kawade. (in Japanese.)

Metcalf, C. L. and W. P. Flint. (1951): *Destructive and useful insects, their habits and control.* Chaps. I. II. 1-76. Chap. IX. pp. 255-259. McGraw-Hill.

Milne, A. (1957): The natural control of insect populations. Cen. Ent. 139(5). pp. 193-213.

———. (1957): Theories of natural control of insect populations. Cold spring Harbor Symposia on Quantitative Biology. Vol. XXII. pp. 253-271.

Mitsui, S. (1958): Isotope; Nōgyō ōyōgijutsu (*Isotopes and the technique applicable to agricultural sciences.*) pp. 243. Chijinshokan. (in Japanese.)

Morita, Y. (1944): *Analysis of Population Growth* (Jinko Zoka no Bunseki) Chap. II. pp. 65-140. Nihonhyoron. (in Japanese.)

Nagasawa, S. (1955): Probit Ho ni yoru Toryo-Hannoritsu Kyokusen no Keisan (*Calculation of Dosage-Reaction Curve by Probit Method.*) pp. 32. Kyushu Noshi Kenkyu Kikan Kyogikai. (in Japanese.)

Nitto, M. and K. Shimizu. (1954): On the chestnut varieties resistant to the chestnut gall wasp (*Dryocosmus kuriphilus* Yasumatsu). Bul. Tokyo Univ. Forests. No. 47. pp. 173-188. (in Japanese with a summary in English.)

Nohara, K. (1956): Considerations on the reproductive capacity of *Dryocosmus Kuriphilus* Yasumatsu (Hymenoptera: Cynipidae). Sci. Bul. Fac. Agr. Kyushu Univ. 15(4). pp. 441-446. (in Japanese with a summary in English.)

Okamoto, D. and Koshihara, T. (1959): The new method of application of BHC to the rice plant borer moth. Plant Protection. 13(6) pp. 5-9. (in Japanese.)

Pearl, R. (1926): *The Biology of Population Growth.* Chap. I. pp. 1-24. Williams and Norgate.

Roan, C. C. (1952): Tagging Oriental fluit flies with radioactive phosphorus for field-movement studies. Jour. Econ. Ent. 45(5) pp. 826-828.

Rodriguez, I. G. (1954): Radiophosphorus in metabolism studies in the two-spotted spider mite. Jour. Econ. Ent. 47(3) pp. 514-517.

Shinji, O. (1944): *Galls and Gall Insects* (Chuei to Chuei Konchu) pp. 580. Shunyodo. (in Japanese.)

Shiraki, T. (1954): *Classification of Insects* (konchu no Bunrui) pp. 961. (Hokuryukan. (in Japanese.)

———. (1956): A Book with References on Insect Classification. (Konchu no Kensaku). pp. 284. Hokuryukan. (in Japanese.)

Smith, K. M. (1951): *A Text Book of Agricultural Entomology.* Chap. II. pp. 6-12. Cambridge.

Tanabashi, S. (1957): Gifu-ken ka niokeru kuritamabachi no higai to tenteki niyoru bogyo. Shinrinboeki news. 6(4) pp. 87-88.

Thompson, W. R. (1951): The time factor in biological control. Canad. Ent. 83 pp. 230-240.

Torii, T. (1956): Chestnut gall wasp in Nagano Prefecture. Shin Konchu. 9(7) pp. 39-43. (in Japanese.)

———. (1956): *The Stochastic Approach in Field Population Ecology with special Reference to Field Insect Populations.* Chap. XII. Sect. III, pp. 230-243. Japan Soc. for Promotion of Sci.

———. (1957): *Statistical Methods of Prediction and Sampling Methods. Konchu Jikkenho.* Chap. XXIII. pp. 560-602. Nippon Shokubutsu Kyokai. (in Japanese.)

Townes, H. (1958): Some biological characteristics of the Ichneumonidae (Hymenoptera) in relation to biological control. Jour. Econ. Ent. 51(5) pp. 650-652.

Uetsuki, K. (1957): Okayama-ken ka niokeru kuritamabachi to sono tenteki. Shinrinboeki news. 6(4) pp. 90-91.

Wardle, R. A. (1929): *The Problems of Applied Entomology.* Chap. V. pp. 112-132. Manchester Univ. Press.

Yasumatsu, K. (1955): Some considerations on the reproductive capacity of a Kyushu race of *Anicetus ceroplastis* Ishii, an effective parasite of *Ceroplastes rubens* Maskell in Japan. Sci. Bul. Fac. Agr. Kyushu Univ. 14(1) pp. 7-15. (in Japanese with a summary in English.)

———. (1954): Noyaku de gaichu o bojo suru baai tenteki o do toriatsukaeba yoika (How should we treat natural enemies in case of chemical control of insect pests?). Noyaku 1(7) pp. 15-18. (in Japanese.)

———. (1955): Studies on the parasitic wasps on the chestnut gall wasp. (Kuritamabachi no kiseiho no kenkyu.) Shinrinboeki news 4(5) pp. 100-102. (in Japanese.)

———. (1955): The effect of insectisides on natural enemies. (Tenteki to Satchuzai). Nogyo oyobi Engei. 30(1) pp. 167-170. (in Japanese.)

———. (1958): Reflection on the control of insect pests. (in Japanese.) (Gaichu Bojo ni tsuiteno Hansei). Hayashi No. 11, pp. 5-9.

———. (1956): *A Story of Natural Enemies.* (Tenteki no hanashi.) pp. 91. Ringyo Fukyu Series. (in Japanese.)

———. (1957): A reperception of natural enemies. (Tenteki no saininshiki.) Noyakukenkyu. 4(1) pp. 54-58. (in Japanese.)

Yasumatsu, K., K. Umeda and T. Kaku. (1953): Control of pine beetles by the use of organic chemicals. Jour. Fac. Agr. Kyushu Univ. 10(2) pp. 151-162.

Yasumatsu, K. et al. (1953): *A Text Book of Applied Entomology.* Chap. 13. pp. 211-225. Asakura. (in Japanese.)

Yasumatsu, K. (1957): Advise against the misunderstanding concerning the utilization of natural parasites on the chestnut gall wasp. (Kuritamabachi no tenteki riyo ni tsuiteno chūi.) Shinrinboeki news. 6(4) pp. 83. (in Japanese.)

Yates, W. W., C. M. Gjullin and A. W. Lindquist. (1951): Treatment of mosquito larvae and adults with radioactive phosphorus. Jour. Econ. Ent. 44(1). pp. 34-37.

Yokoyama, M. and M. Kinoshita. (1951): *The chestnut Gall Wasp.* (Kuritamabachi). pp. 26. Ringyo Fukyu Series. (in Japanese.)

Yoshihara, T. (1951): *On the Logistic Curve.* pp. 95. Tokyo Fisheries College. (in Japanese.)

(Note: An asterisk denotes the paper whose original one has not yet be read.)

EXPLANATION OF PLATE I

Phot. I, a. The chestnut galls formed on a chestnut twig. An arrow A indicates the new galls formed this year, an arrow B indicating the old hardened ones formed in the preceding year.

Phot. I, b. The clippers originated with the author for clipping the twigs off the tall chestnut trees.

Phot. I, c. A large cylindrical outdoor emergence trap (50 cm. in length, 30 cm. in diameter) settled on a bundle of branches of a wild chestnut tree severely infested with the chestnut gall wasp. The trap is netted with the brass net of very minute mesh, two glass tubes, each with 2 pieces of decapitated matchsticks well soaked in honey, being attached to the upper side of it.

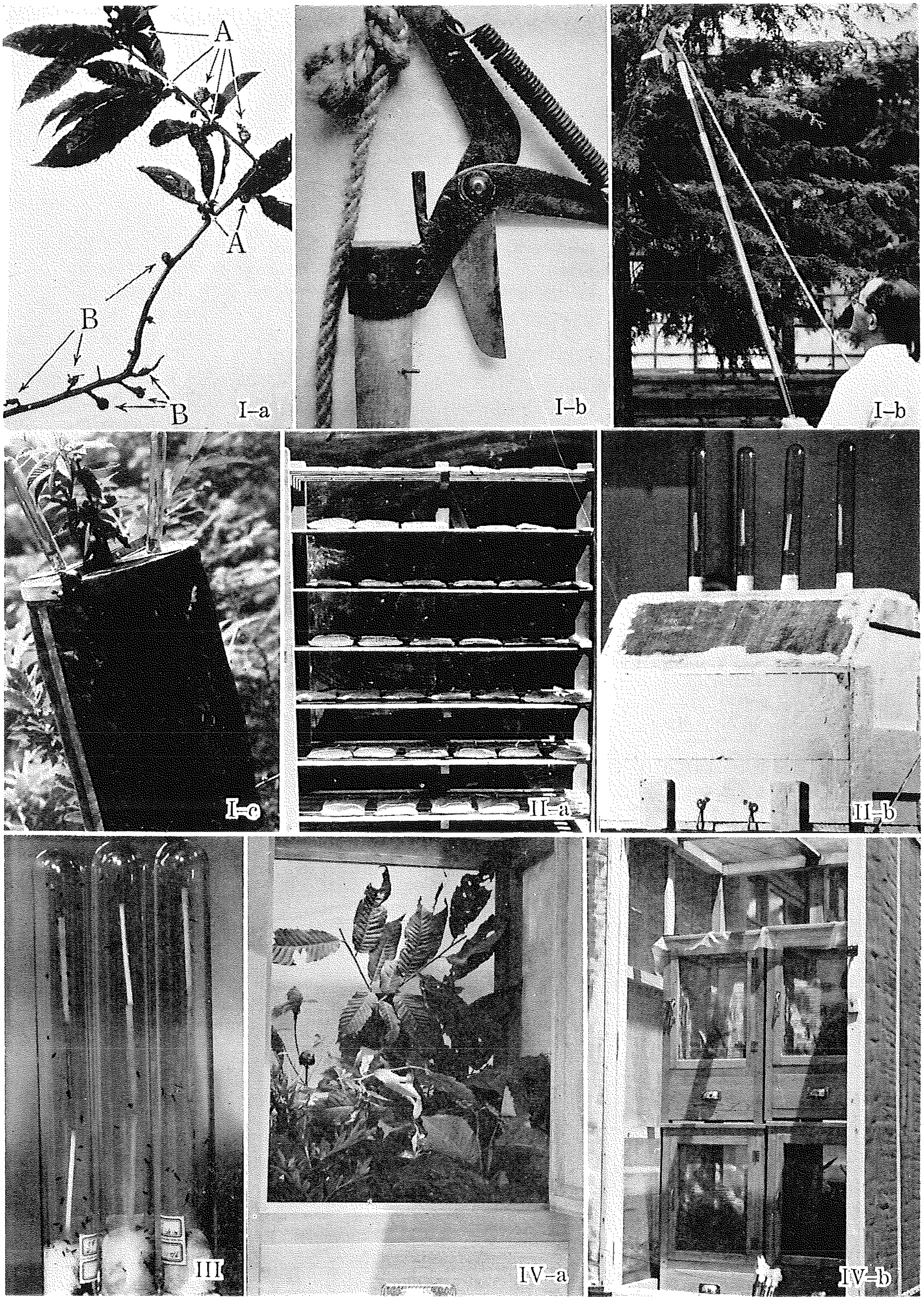
Phot. II, a. The rack for storing the paper bag in which the collected chestnut galls were kept.

Phot. II, b. An indoor emergence trap (ca. 1/10 in size) for parasitic natural enemies emerging from the chestnut galls which were kept within it. Four glass tubes are attached to the top of the trap, each with 2 pieces of decapitated matchsticks well soaked in honey. After emergence, parasitic wasps can easily be observed coming up into them.

Phot. III. A glass tube for feeding the parasitic wasps fresh from the chestnut galls, each with 3 pieces of decapitated matchsticks well soaked in honey.

Phot. IV, a. A small cage for release use, the four sides and the ceiling of which are netted with the brass net of very minute mesh.

Phot. IV, b. A large outdoor cage in which four small cages for release use are placed.


A
A
B
B
I–a
I–b
I–b
I–c
II–a
II–b
III
IV–a
IV–b

EXPLANATION OF PLATE II

Phots. V–X. The macro-autoradiographs taken in 1958. Compare with the left-hand side plant specimens which were caged the P^{32}-labelled parasitic wasps released, and note the position of the typically sensitized portions, which corresponds to that of the galls and of the buds of young terminal shoots. In these autoradiographs, a white portion indicates the portion sensitized by radioactive P^{32}. Autoradiograph time, 145 days.

A: Chestnut leaf blotches caused by a leaf miner moth, *Tischeria complanella* Hübner, and a red ball gall on *Quercus acutissima* leaf, cased by an unidentified gall insect.

B, D, and H: Young terminal shoot of the chestnut twig with chestnut galls caused by a gall wasp, *Dryocosmus kuriphilus* Yasumatsu.

C: Cherry leaf aphid burns, caused by an aphid, *Phomyrus sasaki* Mats.

E: Oak knot galls, caused by a kind of leaf roller moth, *Pelataea bicolor* Walsingham.

F: Warty galls on mugwort leaf, caused by a gall fly, *Rhopalomyia foliorum* Kieffer.

G: Bowl-like galls on mugwort stem, caused by a gall fly, *Rhopalomyia tubifex* Kieffer.

I: Chestnut galls on chestnut twig, caused by a gall wasp, *Dryocosmus kuriphilus* Yasumatsu.

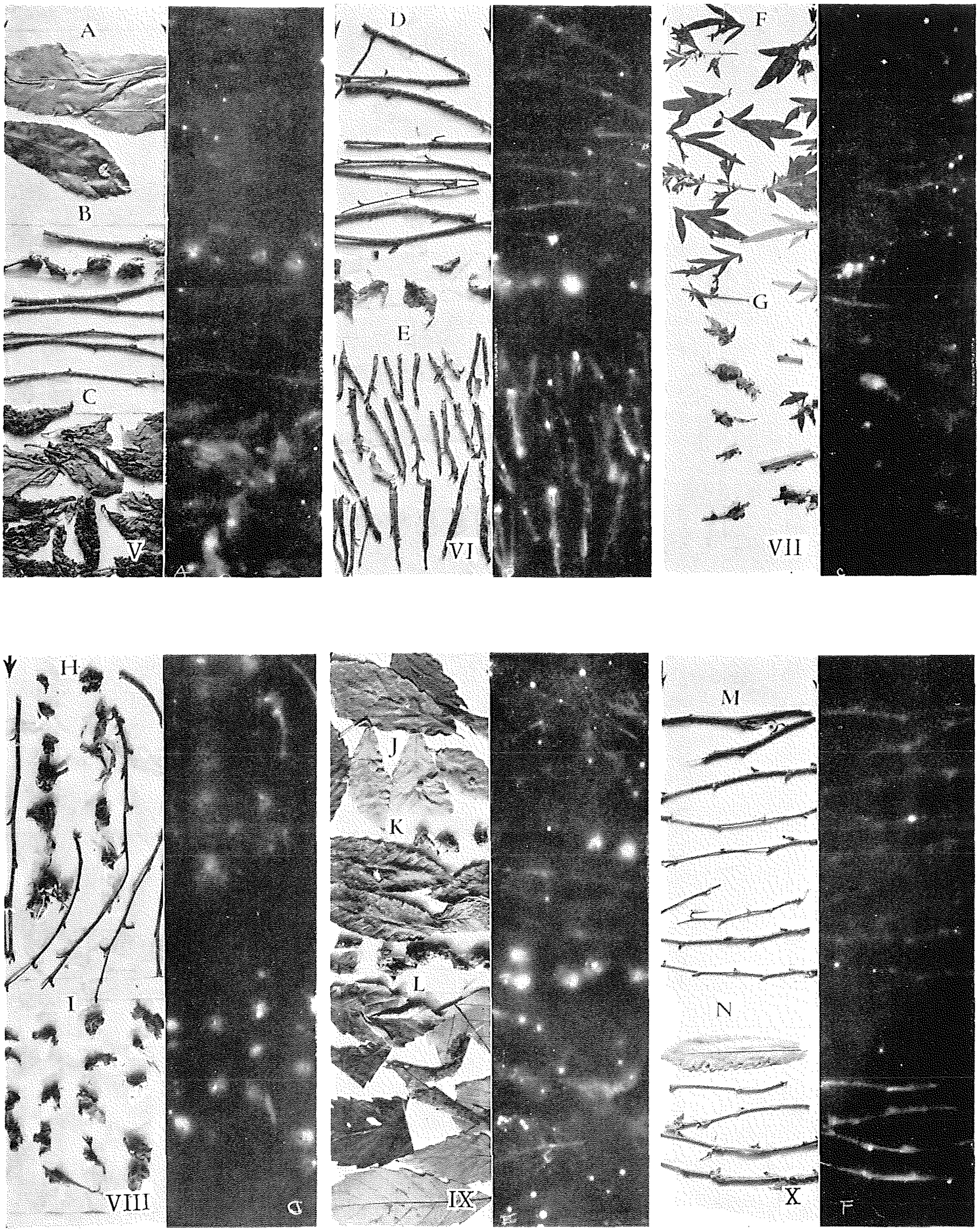
J: Ball galls on oak leaf vein, caused by an unindentified gall wasp, oak leaf rolls caused by an aphid, *Phyllaphia korarae* Shin. and chestnut serpentine leaf miner winding trails caused by an unidentified serpentine leaf miner maggot.

K: Chestnut galls on chestnut twig, caused by a gall wasp, *Dryocosmus kuriphilus* Yasumatsu and chestnut leaf pitted spottings caused by a tree louse, *Trizoa querci* Shin.

L: Oak leaf rolls caused by an aphid, *Phyllaphia korarae* Shin. and oak leaf blotches caused by a leaf miner moth, *Tischeria complanella* Hübner.

M: Young terminal shoot of the chestnut twig without chestnut galls caused by a gall wasp, *Dryocosmus kuriphilus* Yasumatsu.

N: Young terminal shoot of *Quercus acutissima* and minute, pitted spottings on *Quetcus acutissima* leaf.


A
B
C
V
D
E
VI
F
G
VII
H
I
VIII
J
K
L
IX
M
N
X

EXPLANATION OF PLATE III–IV

Phots. XI–XIV. The macro-autoradiographs taken in 1959. In these autoradiographs, a black portion indicates the portion sensitized by radioactive P^{32}. Autoradiograph time, 30 days.

Marks a, b, c, and d indicate a set of plant specimens caged together with the P^{32}-labeled parasitic wasps as the following combination:

a: the specimens caged with *O. nigritibialis.*

b: the specimens caged with *Megastigmus* spp.

c: the specimens caged with *A. amoenus japonicus.*

d: the specimens caged with *E. rosae* plus *P. ferrierei.*

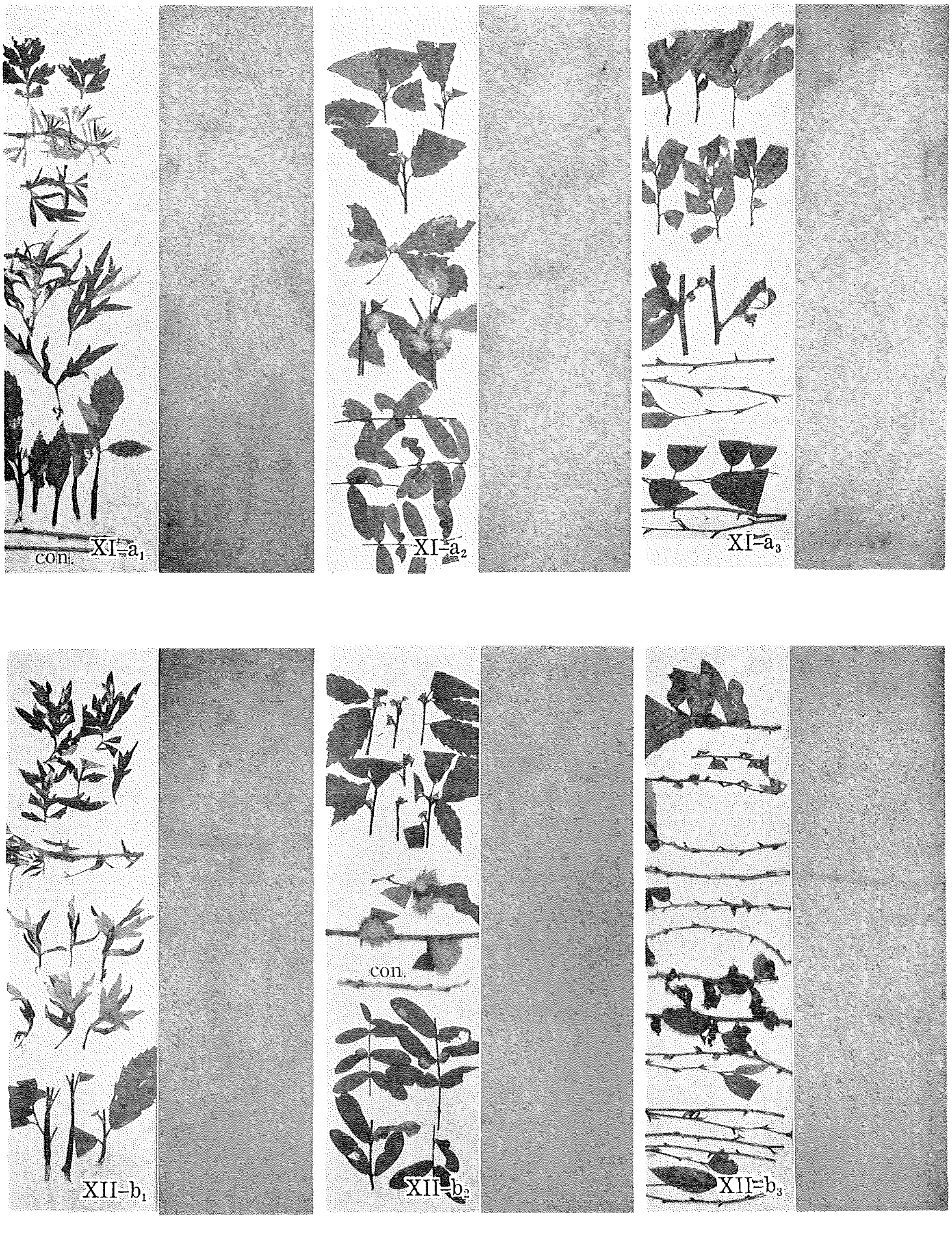
A set of specimens classified with the marks suffixed with arabic figures 1, 2, and 3 are arranged in each set in such an order from top to bottom as follows:

1: Warty galls on mugwort leaf, bowl-like galls on mugwort stem, mugwort leaf aphid burns, oak knot galls,

2: Oak scale galls, oak burry galls, bowl-like galls on willow leaf,

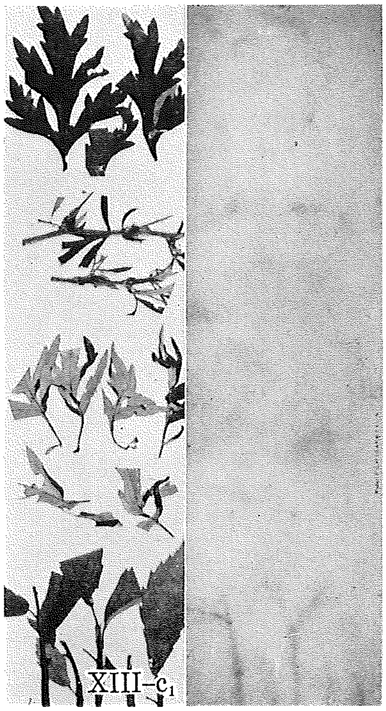
3: Galls on the twig of *Q. acutissima* plus its young terminal shoots, *Q. acutissima* young terminal shoots without galls, chestnut galls, chestnut young terminal shoots susceptible to the chestnut gall wasp, chestnut young terminal shoots resistant to the chestnut gall wasp.

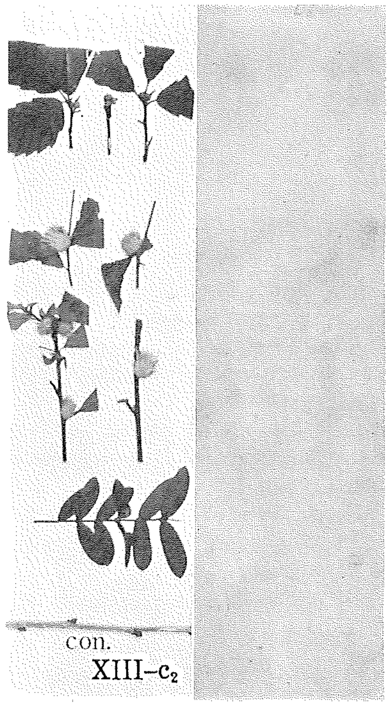
Con.: P^{32}-free control specimen.

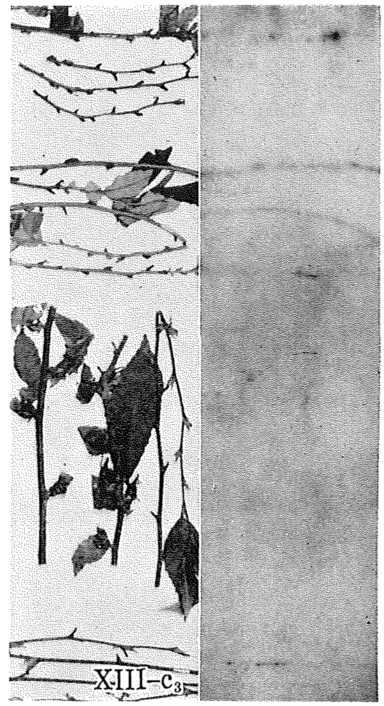

con.
XI-a1
XI-a2
XI-a3
XII-b1
con.
XII-b2
XII-b3

EXPLANATION OF PLATE IV

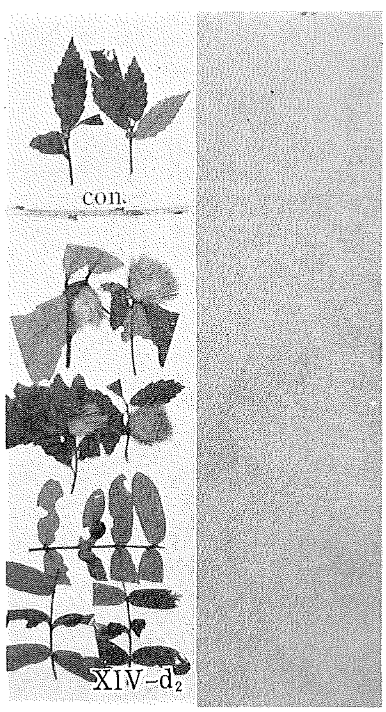
Cf. the explanation of plate III.

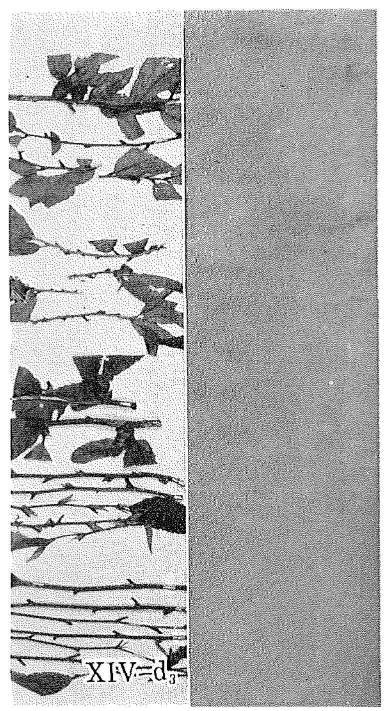

XIII–c₁


con.
XIII–c₂


XIII–c₃


XIV–d₁


con.
XIV–d₂


XIV–d₃

EXPLANATION OF PLATE V

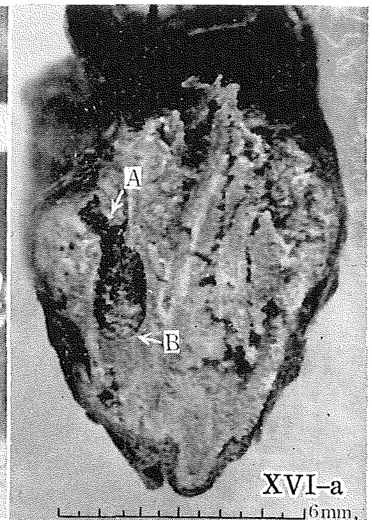
Phot. XV. Three jars, each being about 1/3 in size, for the use of observing the parasitic wasps emerging from the chestnut galls treated with γ-BHC, each with a cap netted with the brass net of 50 mesh. From left to right: Galls sprayed with γ-BHC wettable powder suspension, galls dusted with γ-BHC powder, and γ-BHC-free control galls.

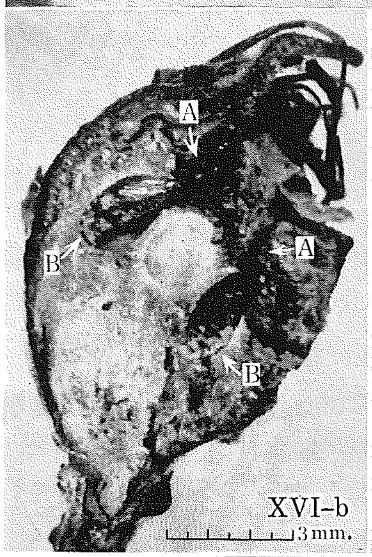
Phot. XVI. A cross-sectional view of the chestnut gall which was treated with γ-BHC in the early season (on 18th June). As regards the details see the text.

An arrow A: An exit hole left unfinished.

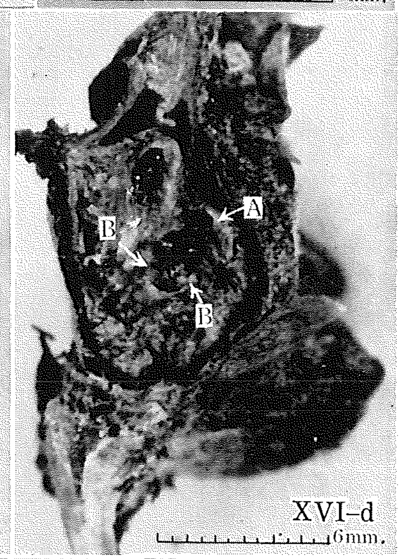
An arrow B: Frass or woody refuse produced by the adult wasp as a result of its burrowing an exit hole.

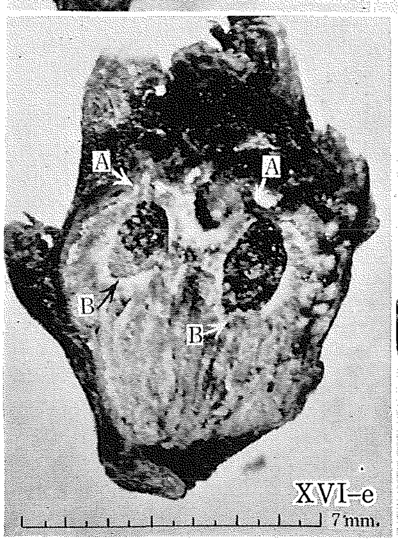

No.5
No.1
XV

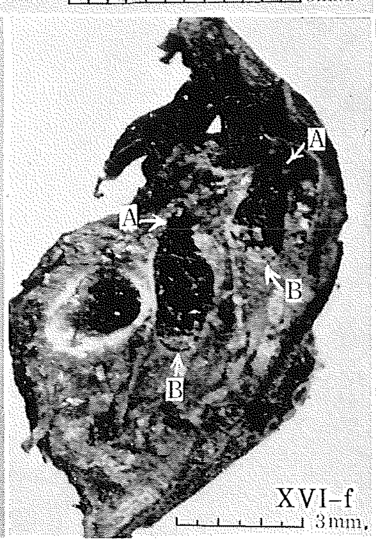

A
B
XVI-a
6mm.

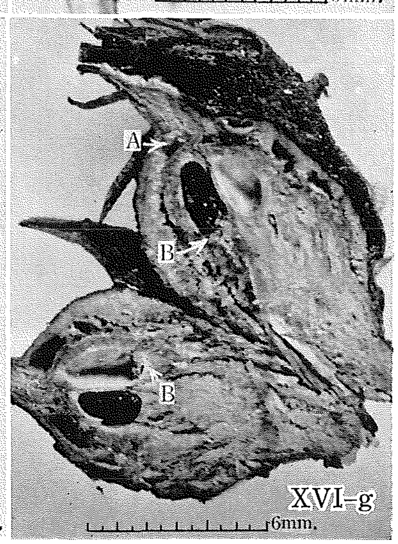

A
A
B
B
XVI-b
3mm.


A
B
XVI-c
6mm.


A
B
B
XVI-d
6mm.


A
A
B
B
XVI-e
7mm.


A
A
B
B
XVI-f
3mm.


A
B
B
XVI-g
6mm.