On the Theoretical Bases for Predicting the Occurrence of the Rice Stem Borer in the First Generation*

By

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I. Introduction.

It is generally recognized that the rice stem borer, ***Chilo simplex*** BUTLER emerges twice a year, but in southern parts of Shikoku and Kyushu it is said to have three generations, while in Hokkaido and in some parts of northern Honshu the appearance is only once a year. General opinion is that the voltinism of this insect is to a large extent affected by environmental conditions, especially by the total effective temperature. Nevertheless, it is well known that in some cooler localities the moths of the first generation appear far earlier than what it should be expected when based upon the theory of thermal constant. Although this phenomenon may be purely a result of inherent racial characteristics of some groups or ecotypes, the author is of the opinion, however, that the phenomenon, at least to some extent, can be recognized when physiological process of the larvae is made clear.

The overwintering larvae of the rice stem borer seems to begin showing signs of awaking from diapause early in spring by increasing their sensibility to environmental conditions, especially to the temperature. In forecasting the time of emergence of first generation moths, a physiological study of the hibernating larvae which are in state of diapause, seems worth considering. Such a study will further bring to light the factors which check the population of the larvae during winter.

The present study was undertaken in the hope that some advance might be made toward a better understanding of the physiology of the hibernating larvae and to establish the theoretical basis by which the prediction of the occurrence of first generation rice stem borer belonging to the Western strain (FUKAYA, 1948) can be facilitated.

II. Materials and Methods.

The physiology of the hibernating larvae can be studied by several different methods, but in these experiments the following items were considered.

1. The effect of temperature during egg stage upon the induction of diapause in larvae.

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2. The fat and water contents of the hibernating larvae.
3. The seasonal changes of freezing point of body fluids.
4. The resistance of the hibernating larvae to cold.
5. The interrelation between the environmental conditions and the free or bound water in larvae.
6. The time of emergence from the diapause.

From these observation at least four problems would be expected to be solved: the first is to determine if the diapause of the larvae are induced by environmental temperature in egg stage; the second, the cold hardiness of the hibernating larvae; the third, when the diapause will be broken; and the fourth, what is the most reasonable method for forecasting the appearance of the rice stem borer in the first generation.

The rice stem borers used in this experiment were gathered in Okayama-Ken, where the diapause of the larvae is so pronounced in winter. The eggs were the so called "last night's egg", laid by females attracted to a fluorescent light in rice fields, and allowed to lay eggs on a paper in the laboratory. The larvae were reared in small glass tubes held in a saturated humidity. The test tube method was used for breeding larvae. The larvae just after hatching were cultured individually by feeding fresh stems of rice plant cut to 10 to 12 cm long usually once every two days. The tubes were set upside down to maintain a high humidity and to prevent the larvae from escaping.

Eggs and larvae were subjected to different temperatures as the experiment required.

For the effect of light, eggs were either placed in the dark or in the light.

The number of moulting was recorded at the time when the feed stems were renewed.

Larvae required for experiments during the winter were collected from the stacks of the fields; and for measuring the seasonal changes of the body elements, fresh materials were used as much as available. The larvae were exposed to temperatures by confining them, *in situ* in straw, in metal cylinders 10 cm in diameter and 5 cm in height, the two ends of which were sealed with brass nets, and placed in a thermostat controlled compartments. The humidities in the can were regulated by means of Zwölfer's method.

For the analysis of the water and the ether extractable fat, the standard method were employed.

In order to measure the amount of bound and free waters of materials the dilatometer was used.

The Beckman's method was used for determining the freezing point of the extracted body fluid. The oxygen consumption, which is said to indicate something about physiology of hibernating larvae, was measured by the Warburg's method.

III. Experimental Results.

1. *Factors inducing the diapause.*

The term diapause has been used to denote the apparent ceasation of the physiological development of an organism, independent on the external conditions. In fact, however, it is hard to determine a true diapause of an organism, as the physiological state of the organism is not easily grasped by the external appearance alone. The author has come to believe in the case of the rice stem borer, that the diapause is a state characterized by the inactiveness of the physiological processes leading to pupation.

Diapause of the borer is recognized to be induced generally in late autumn, but actually, it is commonly found in the larvae of the first generation as well as in the second when the environmental temperature is yet sufficiently high.

The younger larvae, taken from fields early in September, will easily develop until they attain the fifth instar, shortly after placing them in an incubator regulated to a temperature of from 25 to 32°C, but none of them will pupate so early as they will be expected. This may be due to the larva's state of diapause, but how such diapause has been brought about is a problem to be determined.

Thus, using the larvae reared from eggs which have previouly been subjected to different temperatures, the experiments were conducted to determine the effects of such treatments upon the percentage of pupation, length of larval period and duration of each of the instars of larvae. The results are shown in Tables 1 to 3.

Table 1. Pupation as affected by the conditions in the egg stage.

Temperatures exposed on eggs (°C)	Temperatures exposed on larvae (°C)	Number of materials tested	Percent pupation*
22.0 (Dark)	32.0	26	0
27.0 (Dark)	31.0	49	16.1
29 7 (Dark)	31.0	31	32.3
31.0 (Dark)	31.0	105	45.7
31.0 (Light)	31.0	63	42.9
33.3 (Light)	31.0	32	4.4

* Recorded at 50 days after hatching.

Table 2. Length of larval period as affected by the condition in the egg stage.

Section*	Temperatures exposed on eggs (°C)	Number of materials tested	Length of larval period (Days)
A	31.0 (Light)	5	21.6±2.0
A′	31.0 (Dark)	11	23.1±1.4
B	31.0 (Light)	17	20.5±1.2
B′	29.7 (Dark)	10	20.2±1.3
C	30.0 (Dark)	17	22.8±0.8
C′	27.0 (Dark)	8	23.5±1.4

* A, A′; B, B′ and C, C′ are one half of an egg mass, respectively.

Table 3. Instar duration as affected by the conditions in the egg stage.*

Temperatures exposed on eggs (°C)	Number of larvae tested	Number of ecdysis	Instar duration in days				
			1	2	3	4	5
31.0 (Dark)	5 (Non-pupated)	3	3.2	4.0	6.0	17.2	—
	15 (Pupated)	4	3.2	2.9	2.8	5.2	9.7
27.0 (Dark)	12 (Non-pupated)	3	2.0	4.0	6.7	15.3	—
	3 (Pupated)	4	2.7	3.3	4.0	4.7	8.0

* This table is result on surviving larvae 50 days after hatching. The breeding temperature of larvae was 31.0°C.

The foregoing Table 1 and 3 clearly indicated that the length of larval stage is affected by the temperature at the egg stage. When the eggs are kept at a temperature below 22°C, larvae that follow will enter a normal diapause. Non-dormant individuals appear for the first time when codition for the egg is 27°C, and at 31°C the percentage of them reaches a maximum; on the other hand, the condition of darkness during the egg stage seems to give a higher percentage of pupation -- the non-dormant individuals, than when light, but their effects upon eggs are not clear.

Unaffected the egg condition, there were no differences observed in the duration of larval period among those that succeeded pupation as shown in Table 2.

The duration of instar differed between the larvae that entered the diapause and those that failed; and it is apparently recognizable from Figure 1 that the larva's duration of instar upon those that entered diapause is longer after third instar than that pupated.

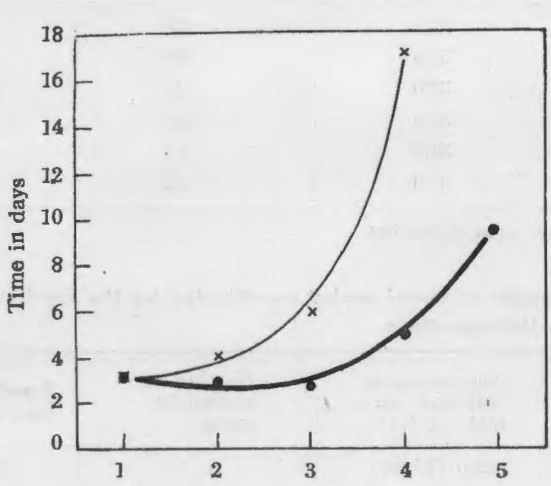

Fig. 1

Instar Duration Curve at 31°C.

× ——— Dormant larvae

• ——— Non-dormant larvae

2. *Water and fat contents of the hibernating larvae.*

(i) Seasonal fluctuation of the water and fat contents.

The results secured during three years from 1943 to 1946 are shown in Tables 4 to 6 and Figure 2,

Table 4. Seasonal fluctuation of water and fat. (1943-1944).

Date	Water (%)	Dry weight (%)	Percent fat on basis of	
			Live weight	Dry weight
Nov. 30	66.37	33.64	16.37	49.78
Dec. 15	63.98	36.02	17.77	49.33
Jan. 15	65 65	34.35	17.35	50 51
Feb. 15	67.33	32.67	17.33	53.05
Mar. 15	67.83	32.17	12.80	39.79
Apr. 15	67.83	32.17	12.11	37.64
May 15	64.93	35.07	15.22	43.40

Table 5. Seasonal fluctuation of Water and fat. (1944-1945).

Date	Water (%)	Dry weight (%)	Percent fat on basis of	
			Live weight	Dry weight
Sept. 22	72.07	27.93	13.04	46.70
Oct. 21	63.91	36.09	17.30	47.95
Nov. 6	66.29	33.71	—	—
Nov. 21	66.07	33.93	—	—
Dec. 27	66.58	33.42	—	—
Jan. 23	67.43	32.57	—	—
Feb. 23	68.11	31.89	—	—
Mar. 1	69.13	30.87	15.84	51.33
Mar. 16	67.65	32.35	15.10	46.69
Apr. 5	68.34	31.66	14.42	45.55
Apr. 16	68.11	31.89	15.32	48.04
May 16	69.18	30.82	14.50	47.06

Table 6. Seasonal fluctuation of water and fat. (1945-1946).

Date	Water (%)	Dry weight (%)	Percent fat on basis of	
			Live weight	Dry weight
Oct. 25	68.32	31.68	15.18	47.92
Dec. 4	64.32	35 68	17.10	47.87
Dec. 20	65.82	34.18	—	—
Jan. 16	67.68	32.32	—	—
Jan. 31	65.51	34.49	—	—
Feb. 15	66.32	33.68	—	—
Mar. 5	66.00	34.00	16.53	48.58
Mar. 22	65.69	34.31	—	—
May 5	67.02	32.98	—	—

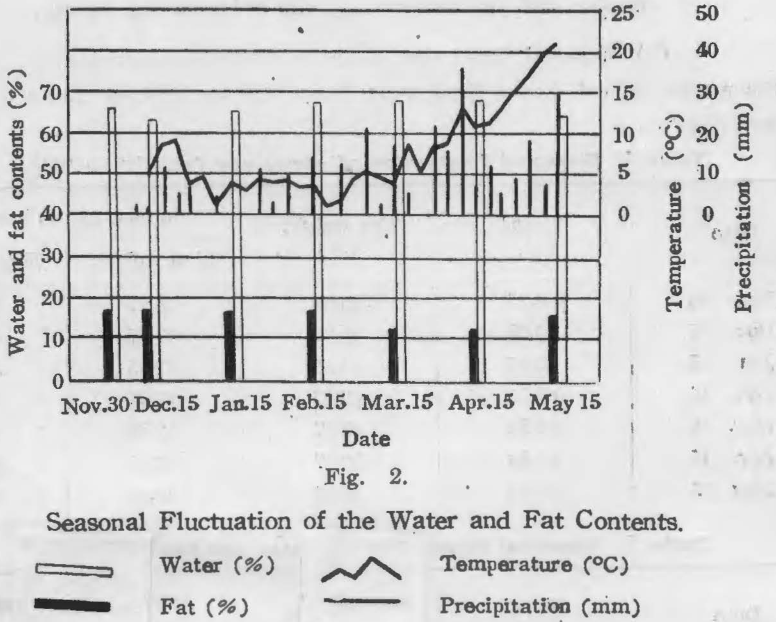

Fig. 2.

Seasonal Fluctuation of the Water and Fat Contents.

So far as the author has measured, the water content of the larvae fluctuates in amount from 64 to 72%, and high water content above 70% are shown in 2nd or 3rd instar of the larvae in general; while the 5th instar-larvae in state of hibernation maintained water about 65%.

The ratio of the fat to live weight of the hibernating larvae ranged between 13 and 17%, increasing at the end of autumn and remained constant during winter, but decreased rapidly in early spring. The ratio of the fat to dry weight seems to show the same fluctuation as to the live weight.

(ii) Effect of environmental humidity upon the water and fat contents.

The results of monthly measurements of the water and fat contents of the materials collected in fields in December and kept in moist (90-100%) and dry (70-80%) conditions in the laboratory are shown in Table 7. The moisture of the straw was also measured, as there are possibilities where the water content of the borer might have some correlation with the straw being a part of the habitat.

Table 7. Water and fat contents as affected by the environmental humidity.

Date	Water (%)		Dry weight (%)		Percent fat on basis of				Water of straw (%)	
					Live weight		Dry weight			
	90-100	70-80	90-100	70-80	90-100	70-80	90-100	70-80	90-100	70-80
Dec. 15	63.98		36.02		17.77		49.33		40.94	
Jan. 15	62.88	60.33	37.12	39.67	17.06	17.22	45.96	43.41	37.41	21.86
Feb. 15	64.42	64.13	35.58	35.87	—	—	—	—	37.45	18.22
Mar. 15	65.33	63.03	34.67	36.97	15.13	16.47	43.64	44.55	40.60	17.57
Apr. 15	64.24	61.23	35.76	38.77	—	—	—	—	43.78	20.17
May 15	64.96	60.44	35.04	39.56	15.21	17.65	43.41	44.62	47.56	11.95

Reviewing Table 7, it is clear that the moisture of the straw increased under the moist condition as the season progressed in contrast to the rice stem borer where no regular tendency is shown. In dry, there is also no recognizable correlation between the moisture content of the straw and that of the insect; but when the water contents of the larvae of the two humidity treatments are compared for the same period, the materials in higher humidity showed always a higher moisture than that in low. This difference is induced likely from the moisture of the straw.

The remarkable decrease of fat content in early spring is indicated independently from the environmental humidity. It is further indicative that the fat in the borer that spent in dry condition will be more than that under moist condition, when measured at the same time.

(iii) Effect of certain artificial conditions upon the water and fat contents of the hibernating rice stem borer.

Some experiment were made on the effect of certain artificial conditions created in the laboratory upon the rice stem borer which is in a state of deep diapause of hibernation.

Larvae collected from the fields in January were placed in two decicators, the relative humidities of which were regulated to between 90-100% and 70-80%. After an exposure to a temperature of 25°C for 10 days in one group and 2°C for 15 days in the other, the total weight, water and fat contents of th body were determined and compared with those before the treatments as shown in Table 8.

The results were shown that the body weight generally decreased at both temperatures, but this tendency was more marked at 25°C by losing 12-13%. On the other hand, the relative water content at 25°C indicated some 64% which is a decrease of about 1%; while at 2°C, it increased slightly. These findings apparently indicate that the relative water content is not affected by exposures to neither high or low temperatures, at least for a while in winter.

Table 8. Effect of the artificial conditions upon body weight, water and fat contents of the hibernating rice stem borer.

Temperature (°C)	Humidity (%)	Exposure in days	Body weight after treatment (%)	Water to live weight after treatment (%)	Fat to live weight after treatment (%)	Fat to dry weight after treatment (%)
25	90 - 100	10 (Jan. 15-25)	87.34	64.26	16.99	47.54
	70 - 80	10 (Jan. 15-25)	88.11	64.53	17.94	50.58
2	90 - 100	15 (Jan. 15-30)	98.31	66.31	13.97	41.47
	70 - 80	15 (Jan. 15-30)	96.21	66.81	15.20	45.80
Control*	—	—	100.00	65.65	17.35	50.51

* The materials measured Jan. 15, just after collecting.

The absolute amount of fat decreased 1.5-3.5% in both temperature groups, and the degree of decrease was more pronounced at low temperature. Under the condition of same temperature, the loss of fat was less at higher humidity. The relative amount of fat at 25°C, however, showed a decrease of 0.4% at 90-100% humidity; whereas in 70-80%, there was an increase of 0.6%. At 2°C there is a general loss of fat, and more so at the higher humidity.

It is obviously that the relative amount of fat is less affected by the higher temperature than the lower one.

The fat content compared on dried material basis shows similar tread. The loss is greater at lower temperature, and at the same temperature a higher humidity caused more loss.

3. *Seasonal fluctuation of oxygen consumption.*

From the view of physiology of hibernation, it is of significance to study the relation of seasonal fluctuation of oxygen consumption and the diapause. The results of experiments conducted from 1944 to 1945 are summarized in Table 9 and Fig. 3, indicating a great decrease in the oxygen consumption during the period from December to January.

Table 9. The seasonal fluctuation of oxygen consumption. *

Month	Body weight	Oxygen consumption (cmm/h/g)
January	82.8	446.9
April	81.2	399.6
August	70.3	985.3
October	76.5	1070.4
December	86.4	822.8

* Figures are average of five individuals.

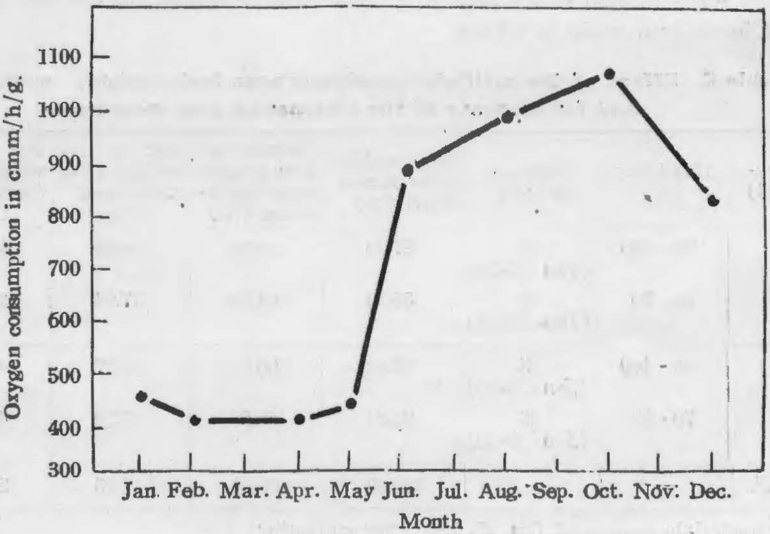

Fig. 3.

Seasonal Fluctuation of Oxygen Consumption.

Table 10. Effect of the environmental temperatures upon oxygen consumption.*

Temperature (°C)	January		April	
	cmm/h/g	μ	cmm/h/g	μ
10	77.8			
15	159.2	23242.8	148.5	
20	274.0	18392.4	233.1	15301.5
25	409.4	13999.5	373.1	16415.7
30	446.9	3213.5	399.6	2525.3
Average body weight of 5 individuals	82.8mg		81.2mg	

The oxygen consumption at temperatures between 10 and 30°C in January and April is given in Table 10 and Fig. 4. As shown in graphic form, there are little differences between the amount of oxygen consumed in both months when measured under same condition, but the μ value of the curve in January becomes steeper as the temperature rises, unlike in April of which value remains comparatively constant.

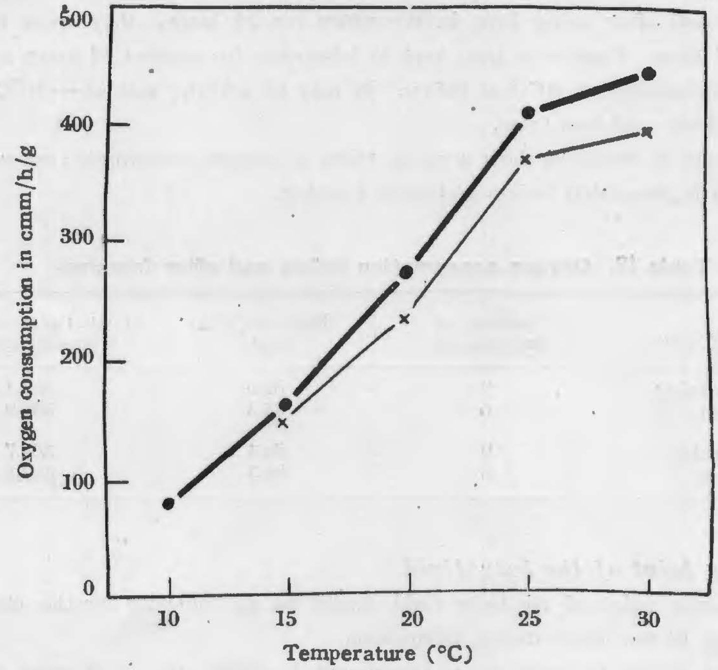

Fig. 4.

Oxygen Consumption at different Temperatures in January and April.

● ——— January
× ——— April

Table 11 summarizes the tendency of change of R. Q. increasing with the development of larvae leading to pupation.

Table 11. Seasonal fluctuation of R.Q.

Date	Number of materials	R. Q.	Body weight (mg)
Mar. 8	5	0.64	96.8
Apr. 26	10	0.78	101.5
May 13	5	0.89	116.8
May 17	5	0.89	88.5

4. *Effect of the freezing upon oxygen consumption.*

Subject of freezing should be an extraordinary phenomenon for a living organism as it should give a striking effect upon the physiology of the rice stem borer. With this purpose the effect of freezing upon the respirative metabolism of the borer was examined.

The oxygen consumption of fresh larvae collected from the fields were measured under 20°C and after being kept in laboratory for 24 hours, they were frozen at −10°C for 1 hour. They were then kept in laboratory for another 24 hours and made a second measurement at 20°C as before. It may be added, that at −10°C, about 60% of the body moisture froze.

The results in Table 12 show a mean value of oxygen consumption where almost no difference is detectable before and after freezing.

Table 12. Oxygen consumption before and after freezing.

Item	Number of individuals	Body weight (mg)	Oxygen consumption (cmm/h/g)
Before freezing	9	83.0	307.1
Control	6	84.3	389.9
After freezing	9	80.4	303.7
Control	6	84.3	429.3

5. *Freezing point of the body fluid.*

The freezing point of the body fluid should be an indicator for the changes in the physiology of the insect during hibernation.

From the results of three years experiment in Table 13, it is quite apparent that the freezing point rises rapidly from March to April. Since such a change is independent on the fluctuation in the water content, it could be that some substancial metabolism occurred during this critical period.

Table 13. Freezing point of the body fluid of larvae.

Year	Date	Freezing point (°C)	Under cooling temperature (°C)
1944	Feb. 3	−1.88	−3.28
	Mar. 3	−1.80	−3.26
	Apr. 3	−1.30	?
1945	Jan. 18	−2.50	?
	Mar. 1	−2.18	?
	Apr. 5	−1.40	−2.57
1946	Jan. 28	−2.38	−4.36
	Mar. 7	−2.21	−2.24
	Apr. 2	−1.70	−1.96

6. *Bound and free waters in the body.*

The bound and free waters, determined in 1944 by the method of SACHAROV (1930), are summarized in Table 14 and 15.

Table 14. Percentage of free water in the body of larvae.

Source of larvae	Date	Exposed temperature (°C)	Water content (%)	Free water to total water (%)	Bound water (%)
Kurashiki, Okayama	Feb. 3	−10.0	65.38	68.63	31.37
〃	Feb. 3	−17.0	65.38	82.04	17.96
〃	Mar. 15	−10.6	67.83	73.01	26.99
Kogane, Yamagata	Feb. 10	−10.0	60.94	40.17	59.83
〃	Feb. 10	−20.0	60.94	72.58	27.42

Tale 15. Effect of environmental conditions upon the amount of free water in the larvae, collected at Kurashiki, Okayama.

Date	Exposed temperature, (°C)	Water content (%)	Free water to total (%)	Bound water to total (%)	Condition before freezing
Feb. 21	−10.0	61.15	59.88	40.12	25°C, 90-100% for 10 days.
Feb. 21	−10.0	62.28	43.85	56.15	25°C, 70-80% for 10 days.
Feb. 23	−10.0	64.36	63.21	36.79	2°C, 90-100% for 15 days.
Feb. 23	−10.0	64.42	66.15	33.85	2°C, 70-80% for 15 days.

It is general that, the lower the freezing temperature, the more greater will be the ratio of the water to be frozen; and this ratio is said to be so affected by the environmental conditions to which the materials were exposed previously. According to the author's results, however, the ratio of frozen water of the rice stem borer subjected to a higher temperature and forced it to lose a part of its hardiness to cold, appeared to be less than for that in the lower temperature.

The author is of opinion that the relation between the cold hardiness and the amount of bound water in the insect requires in future to be reexamined.

7. *Cold resistance of the rice stem borer.*

It is generally understood that the insects distributed over the temperate zone show a more or less cold resistance in winter whereby it is believed to protect themselves from freezing to death. In the case of the rice stem borer too, there is a remarkable resistance of the hibernating larvae to cold, as they will survive the winter in northern district of Japan, where there are days with a temperature of 20°C or lower.

To show the extent of cold resistance of this insect, the author kept some larvae under various low temperatures for a while, then forced them to develop. The ability of the borers to survive low temperatures is shown in Table 16.

Table 16. Effect of low temperatures upon the mortality of larvae.

Temperature exposed (°C)	Hours exposed	Number of larvae	Mortality (%)
− 6.6±0.4	1	20	0
− 6.8±0.7	3	20	0
−10.0±0.2	1	20	0
−14.2±0.2	0.5	20	0
−14.1±0.2	1	20	0
Cont.	—	20	0

* Mortality was recorded at 24 hours after treament.

As many investigators have pointed out, the cold resistance of the insect should be affected by environmental conditions prior to the exposure to cold. The author also attempted to determine the relation between the environment and the cold resistance to cold using the rice stem borer. The data obtained are given in Table 17.

Table 17. Relation between environmental conditions and the cold resistance in the rice stem borer. February 1944.

Condition before treatment	Temperature exposed	Hours exposed	Number of larvae	Mortality (%)
25°C, 90 - 100% for 10 days.	−14.6±0.8	3	30	100
2°C, 90 - 100% for 15 days.	−14.0±1.0	5	30	0
In field	−14.6±.08	3	15	0
In field	−14.0±1.0	5	15	0

From Table 17, a temperature of −14°C for 5 hours gave 100% death of larvae when transferred from a high temperature; while those that were placed at 2°C for 15 days seemed not to have recieved any critical effect by the sudden exposure to the low temperature.

8. *Mortality of the hibernating larvae.*

From an economic point of view a more precise knowledge of the mortality of the hibernating larvae, is of utmost importance, since the mortality during the hibernation has a direct correlation with the number of imagoes emerging in the first generation. It is generally believed that the main factor which causes the death of the hibernating larvae is the extreme cold in winter; whereas according to HARUKAWA (1934) the mortality of the larvae is not very high until May or the approach of warm season.

The results obtained by the author is shown in Table 18 and Fig. 5, from which the view of HARUKAWA is supported. From Fig. 5 we can see the seasonal mortality curve of the larvae rising rapidly in May, and a higher mortality indicated in the moist condition. Such a high mortality may be attributed partly to the agencies of the parasitic Hymenoptera, and still to a larger extent to the fungus, ***Beauveria (Botrytis) bassiana*** BALS.

Table 18. Mortality of the hibernating larvae. (1944).

Date	Humidity (%)	Number of larvae	Mortality (%)
Jan. 15	90 - 100	496	7.1
	70 - 80	227	7.1
Feb. 15	90 - 100	376	4.3
	70 - 80	322	0.9
Mar. 15	90 - 100	386	14.3
	70 - 80	246	18.3
Apr. 15	90 - 100	144	24.3
	70 - 80	45	11.1
May 15	90 - 100	230	22.6
	70 - 80	154	15.6
July 15	90 - 100	248	98.4
	70 - 80	236	90.7

9. *Time of emergence from the diapause.*

In order to forecast the time of appearance of the imagoes in the first generation, it is important to determine the time when the diapause of the hibernating larvae is first broken. with this purpose, the hibernating larvae were exposed to a comparatively high temperature of 27°C for some period during every month from end of autumn to early summer. By this method, one can see the critical time when the insect begins to react to the environmental temperature. Results of the observation are summarized in Table 19 and Fig 6. They show that the exposure to a high temperature during the period from November to January gives little effect upon the development of the larvae as compared to the control, while the larvae treated after February are affected by the treatment.

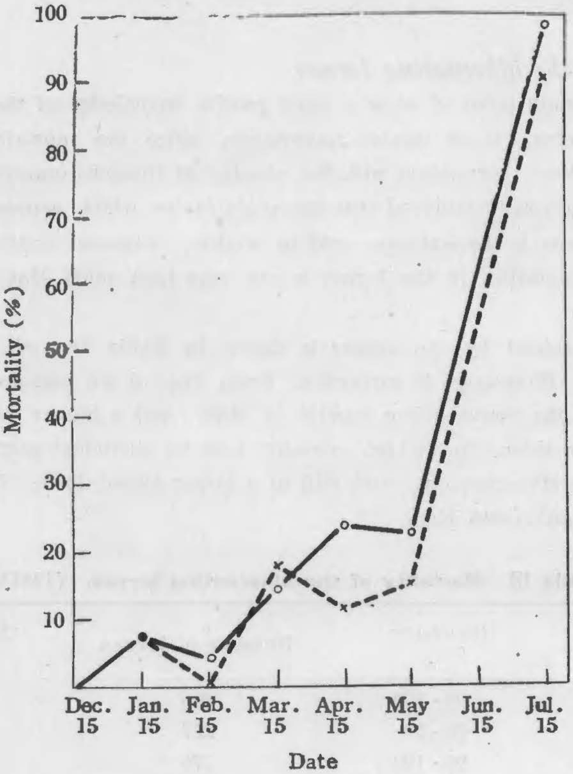

Fig. 5.

Seasonal Mortality Curve of Larvae.

○ —— Moist (90-100%)
× - - - - Dry (70-80%)

Table 19. Effect of the high temperature in winter upon the time of emergence. (1947-1948).

Month	Number of larvae	Percentage of emergence	Date of emergence in June	σ
December	170	8.2	27.3±1.5	±8.0
January	232	15.9	29.3±0.7	±5.6
February	186	27 6	22.3±1.0	±5.8
March	184	17.7	25.4±1.5	±7.7
April	167	15.2	24.4±1.2	±5.7
May	197	13.2	28.2±1.3	±6.8
Control (Non-treated)	233	13.7	5.8±1.4 (in July)	±6.2

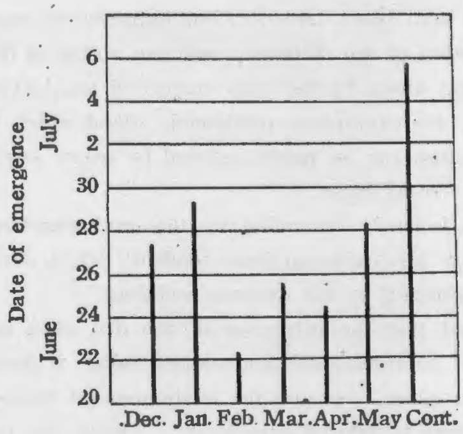

Fig. 6.

Effect of the high Temperature in Winter upon the Time of Emergence, 1943 - 1944.

Since the exposed mean temperature during the colder season of December to March is same, and the fact that the exposure to high temperature in February induced a rapid development of the insect, should be understood as a significance indicating the diapause of the larvae being broken this month. However, the temperature in February is generally so low that it can hardly believe to cause the development of the insect. The environmental temperature after March, therefore seems to be the practical problem. Thus, the author has come to the conclusion that the time of emergence of the imagoes should have some correlation with the environmental temperature after March. Results obtained by Ishikura in his statistical studies seems to support the authors's conviction.

IV. Discussion.

The results of experiments bring to a conclusion that the physiological conditions producting the diapause is initiated early in the development of the insect rather than the sole condition of the fourth or fifth instar which coincides with the entrance into diapause. A similar result, proving that the temperature at a certain stage in the development of an insect affects the subsequent physiological state, has been obtained by Kogure (1933) in the silk worm. He assumed that there exists a certain substance inducing the diapause and named it the hibernating substance. This substance is considered to increase in amount during pupal period by means of high temperature, but distoroyed after egg stage. Babcock (1927) pointed out that the diapause in the corn borer will not be initiated by a lowering of the temperature, while Kozhanchikov (1938) succeeded in inducing the diapause of this borer solely by low temperature.

In the case of the rice stem borer, however, low temperature could be the definite factor controlling the induction of the diapause, and the author is of the opinion that the diapause may be brought about by the consequence of the inactiveness or activeness shown in some organ, for example a prothoracic gland which is so well known in silk warm. So the diapause can be partly induced by every environmental condition which will affect this central organ.

Although the diapause is partly controlled by the environmental condition, one can not deny that there may also exists an inner tendency which decides the physiologial state of an animal unaffected by the external codition.

But it can be concluded that the offsprings of the rice stem borer moth which appear in September when environmental temperature falls, it should already enter the diapause without an exception, because the environmental temperature is so low that the diapause of the larvae is induced completely. Also in the cooler areas where the second generation eggs in the summer are naturally exposed to a temperature below 27°C, prevents the occurrence of the imagoes more than twice a year.
The hibernating larvae are generally in state of diapause as stated above, yet they are considered to be in an unusual condition relative to the cold hardiness. Relation between the amount of water and fat in hibernating larvae and cold hardiness has often been discussed and several authors pointed out that the hardiness to cold is acquired gradually by means of reduction of water or free water of the body and an increase of fat; but in the case of the rice stem borer the body water seems to be so variable to the extent of 5% depending upon the environmental moisture. So the total amount of water content in the body does not appear to be of significance in deciding the cold hardiness. On the other hand, the fat, which is regarded not to take any part in the formation of the tissue, seems to have an important physiological role during the cold season.

According to Sacharov (1930), who studied the relation between the fat and cold resistance of the insect, the cold hardiness correlates with the percentage of fat contained. In fact, there are evidences where a part of the fat in the cutworm has been utilized in the building of resistance against cold. It is easily recognizable that the fat should play an important part in protecting the inner organs of an insect from cooling, being a poor conductor of heat, and since it is distributed between various organs. In the rice stem borer too the fat content increases with the advance of the season from autumn to winter and decreases rapidly in early spring when the environmental tempertaure rises.

Having taken all the above into consideration the author has come to believe that the fluctuation of the fat during winter has some significance upon the cold hardiness.

As Robinsom has shown, it is generally recognized that an increase of bound water in insects coincides with the fall in temperature, and that it is this bound water which protects them during the winter. But it is hard to give any conclusive argument about the cold hardiness basing merely upon the quantity of the bound water, because, so far as the rice stem borer is concerned, the freezing of the body does not mean an immediate death of the insect. According to Kozhanchikov (1938) freezing of the protoplasmic water causes the death of an insect only in the absence of thermostable respiration. The author is of the opinion that the cold hardiness is more closely connected with the type of respiration than with the state of water in the body. In fact, the remarked decrease in the oxygen consumption at the beginning of the winter seems to indicate something about the cold hardiness of the rice stem borer. But the date obtained here are still insufficient for solving this problem. The subjects of bound water and the respiration in winter are still left for further investigation.

Unexpected fact of mortality of the larvae being low in winter and suddenly rising in early summer, teaches us at least the increase of thermostability of the body in winter and that of the environmental resistance especilly of the parasitic agencies just before the pupation.

The diapause of the larvae, being broken gradually in early spring as shown by different sighns in the physiology, can be observed in this critical period, Table 19.

Thus a high correlation between the average date of emergence and the mean temperature in March to May or in April to May is substantiated by the experiments, since the effect of environmental factors affecting physiology of the dormant larvae can be neglected.

Based upon author's investigation the annual forecast of the time of emergence in southern districts of Okayama-Ken can be made by the following formula derived statistically from the data compiled by the Economic Division of Okayama-Ken:

$Y = -5.09X + 94.64$ (Y: date of emergence in June; X: mean temperature during April to May).

On the other hand, the number of imagoes emerging in the first generation, can be calculated from the negative correlation, $r = -0.69$, between the number of imagoes which emerge and the total amount of precipitation in May when the mean temperature of May is higher than usual; and likewise, $r = -0.59$, between the mean temperature when it is lower than usual.

The reason for the above interrelation can easily be explained from the fact that most larvae just before pupation are killed by ***Beauveria (Botrytis) bassiana*** Bals. and other natural enemies, which multiply during high temperature and humidity conditions.

On the Shonai strain of Tohoku and Hokuriku districts, which begins reviving from diapause in late autumn, the larval development is more affected by the environmental temperature in the spring, and the mortality just before the pupation being less, perhaps owing to the prevailing low environmental temperature. The number of imagoes that appear in the first generation in these districts are, therefore, assumed from the population density of winter larvae.

V. Summary.

1. The diapause of the first generation larvae is induced by temperatures below 27°C during egg stage. Non-dormant individuals appeared 45% from larvae reared from eggs which were exposed to a high temperature of about 30°C.

2. The sighn of diapause can be detected by the duration of larval instar being

prolonged after third instar, in general.

3. The water contents of the hibernating rice borer were apparently affected by the degree of environmental humidity; and of the temperature, it is of secondary in significance.

4. The ratio of fat to live weight of the hibernating larvae increased rapidly at the end of autumn and remained constant during winter, but decreased rapidly in early spring.

5. Oxygen consumption decreased gradually from October, and showed a minimum in January.

6. R. Q. increased with the development of larvae leading to pupation.

7. There was no noticiable difference in the mean value of the oxygen consumption of larvae before and after freezing.

8. Freezing point of the body fluid rose from March to April rapidly.

9. Ratio of frozen water to total water content was greater after contacting a low temperature than a high temperature.

10. The cold hardiness of the rice stem borer was affected by the environmental condition to which it has been placed.

11. The highest mortality of hibernating larvae was shown in May or June, and this may be attributed mostly to the parasitization by a fungus.

12. It appeared that the diapause of larvae was broken in February for the western strain.

13. From experiments, it may be concluded that the time of emergence of imagoes can be forecasted by the formula;

$Y = -5.09X + 94.64$ (Y: date of emergence in June, X: mean temperature during April to May).

14. The number of imagoes which will emerge in the first generation can be expected, to a certain extent, from the negative correlation ($r = -0.69$) between the number of imagoes and the total amount of precipitation in May when the mean temperature of May is higher than usual; and between the mean temperature in May and number of the moth when it is lower than usual, $r = -0.59$.

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APPENDIX

Strains of the Rice stem borer in Japan

By Masatsugu Fukaya

The riec stem borer (*Chilo simplex* BUTLER), being distributed throughout Japan with the exception of some parts of Hokkaido, is considered one of the most serious pests of rice plant, and from which a large part of investigations concerning this insect has been to date centred about the relation to ecological factors. The question of whether there exists such inherent differences among the insect in the forms of distinct strains is still left undetermined.

It is known that in some localities the moths of the first generation appear earlier than what it should be expected when based upon the theory of thermal constant. Although this phenomenon has been explained, in general, as a result of combined effects of certain environmental conditions, the author is of the opinion, however, as based upon evidences, that the phenomenon should be the result of a inherent racial characteristic of some groups, being not so much affected by climatic conditions.

According to the author's investigations, there are at least two distinct groups having different natures. The first group, which may be called the Shonai strain, is distributed in such prefectures as Yamagata, Niigata, Toyama and Ishikawa facing Japan Sea and has a comparatively shorter life sycle than the second group, which is called the Saigoku strain, when they are placed and compared under an identical conditions.

The diapause of the borer belonging to Shonai strain is so slight that, when it is subjected to a temperature of about 25°C in winter (December), the pupation normally follows after 18 days; while on the Saigoku strain, the larvae fails to pupate within 60 days. Furthermore, there is a distinct difference in the movement of the hibernating larvae when observed under a low temperature and analyze the curves obtained according to the probit method (BLISS, 1935), substantiating the physiological differences between the two strains. Thus, the author came to believe that the same control measure that K. OKAZAKI (1947) announced for the boror in Yamagata

* BLISS, C. I. (1935) Ann. App. Biol. 22.
* OKAZAKI, K. (1947) Agric. Hortic. 22.

prefecture could be extended to other distincts where the Shonai strain prevails.

There will be an increase in the numbei of generations (3-4 times) a year if the Shonai strain is transferred from its native habitat to Kurashiki area (Okayama prefecture) where the Saigoku strain occurs with a normal two generations per year.

A more rapid development of the larvae in summer is also observed in Shonai strain as compared to the Saigoku strain under the laboratory conditions in Kurashiki -- the larval period being about 29 days as compared to about 40 days.

In a cross between the Shonai female and Saigoku male, the F_1 progeny resembles the paternal parent. Its segrigation in the F_2 was scarcely observed to occur.

The foregoing results indicate how an inherent physiological characters, such as the larval development in the Shonai strain, are correlated closely with the environmental temperature. It is reasonable there, that the time of emergence of the Shonai strain can be forecasted by correlating with the environmental temperature. The bounding areas confining these two strains have not been determined definitely as yet, but the borer of the neighbouring districts appear to exhibit, in general, the characters intermediate of the two.

Description of the "Figure of Water Qualities in Akita Prefecture."

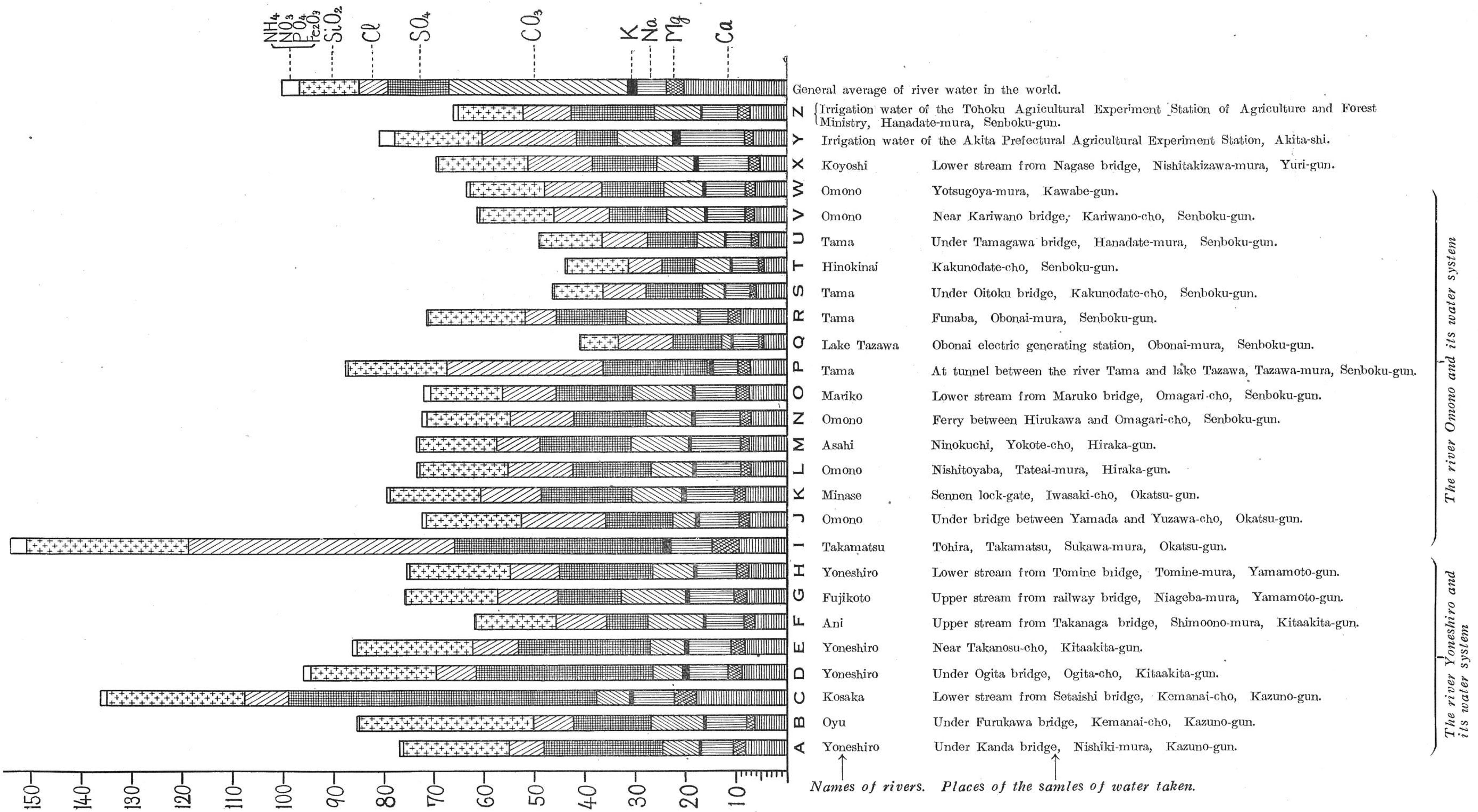
The mineral salts in the river water are composed of four basic radicles (Ca, Mg, Na, K) and three acid radicles (CO_3, SO_4, Cl). To those SiO_2 included, the eight are the principal inorganic components dissolved in the water. The figure shows the concentration of these components in milligrams in a litre.

Compared with the average composition of the river waters in the world, given at the right end in the figure, the rivers in Akita prefecture show: as the basic radicles, the small presence of calcium and the larger presence of sodium; and as acid radicles, the small quantity of carbonic acid and the predominance of sulfuric acid and chlorine.

Accordingly, calcium carbonate, the alkaline component in the river water is short, and calcium sulfate, sodium chloride, etc. are great. This is the characteristics common to every river in Akita prefecture.

The so-called "poisonous waters" as the rivers Tama (P) and Takamatu (I), are the mineral acid rivers which contain hydrochloric and sulfuric acids. The rivers Kosaka (C) and Yoneshiro (D, E) contain abnormally much sulfate because of the mines. These are all unsuitable waters for irrigation.

The most diluted waters are lake Tazawa and the river Hinokinai, which are parts of the river system of Tama.



NH4
NO3
PO4
Fe2O3
SiO2
Cl
SO4
CO3
K
Na
Mg
Ca
General average of river water in the world.
Z {Irrigation water of the Tohoku Agricultural Experiment Station of Agriculture and Forest Ministry, Hanadate-mura, Senboku-gun.
Y Irrigation water of the Akita Prefectural Agricultural Experiment Station, Akita-shi.
X Koyoshi Lower stream from Nagase bridge, Nishitakizawa-mura, Yuri-gun.
W Omono Yotsugoya-mura, Kawabe-gun.
V Omono Near Kariwano bridge, Kariwano-cho, Senboku-gun.
U Tama Under Tamagawa bridge, Hanadate-mura, Senboku-gun.
T Hinokinai Kakunodate-cho, Senboku-gun.
S Tama Under Oitoku bridge, Kakunodate-cho, Senboku-gun.
R Tama Funaba, Obonai-mura, Senboku-gun.
Q Lake Tazawa Obonai electric generating station, Obonai-mura, Senboku-gun.
P Tama At tunnel between the river Tama and lake Tazawa, Tazawa-mura, Senboku-gun.
O Mariko Lower stream from Maruko bridge, Omagari-cho, Senboku-gun.
N Omono Ferry between Hirukawa and Omagari-cho, Senboku-gun.
M Asahi Ninokuchi, Yokote-cho, Hiraka-gun.
L Omono Nishitoyaba, Tateai-mura, Hiraka-gun.
K Minase Sennen lock-gate, Iwasaki-cho, Okatsu-gun.
J Omono Under bridge between Yamada and Yuzawa-cho, Okatsu-gun.
I Takamatsu Tohira, Takamatsu, Sukawa-mura, Okatsu-gun.
H Yoneshiro Lower stream from Tomine bridge, Tomine-mura, Yamamoto-gun.
G Fujikoto Upper stream from railway bridge, Niageba-mura, Yamamoto-gun.
F Ani Upper stream from Takanaga bridge, Shimoono-mura, Kitaakita-gun.
E Yoneshiro Near Takanosu-cho, Kitaakita-gun.
D Yoneshiro Under Ogita bridge, Ogita-cho, Kitaakita-gun.
C Kosaka Lower stream from Setaishi bridge, Kemanai-cho, Kazuno-gun.
B Oyu Under Furukawa bridge, Kemanai-cho, Kazuno-gun.
A Yoneshiro Under Kanda bridge, Nishiki-mura, Kazuno-gun.
The river Omono and its water system
The river Yoneshiro and its water system
150 140 130 120 110 100 90 80 70 60 50 40 30 20 10
Names of rivers. Places of the samles of water taken.