## HEAD CAPSULE MEASUREMENTS OF THE LARVAL INSTARS OF THE LARGE STRAIN SITOPHILUS ORYZAE L. AND SITOPHILUS GRAMARIUS L., IN WHEAT

by

#### EDWIN LOREN SODERSTROM

B. S., California State Polytechnic College, 1957

A THESIS

submitted in partial fulfillment of the

requirements for the degree

MASTER OF SCIENCE

Department of Entomology

MANSAS STATE COLLEGE OF AGRICULTURE AND APPLIED SCIENCE LD 2668 T4 1959 S67 C.2 Document

#### TABLE OF CONTENTS

INTRODUCTION	1
REVIEW OF LITERATURE	2
Unknown Strain Rice Weevil	2
Origin and Distribution	2
Adult Description	2
Biology	3
Large Strain Rice Weevil	3
Description	3
Biology	4
Granary Weevil	4
Origin and Distribution	4
Adult Description	4
Biology	5
Experimental Procedure	5
EQUIPMENT AND MATERIALS	6
Rearing Facilities	6
Source of Stock Cultures	7
Culture Media	7
MGTHODS	7
Media Preparation	
Stock Culture Procedure	8
Experimental Cultures	9
Installation of the Calibrated Ocular	10
Calibration of Microscope	10
Staining of Grain	10

Dissection Procedure	11
Measurement	12
RESULTS AND DISCUSSION	12
Head Capsule Width	14
Variation Within Instars	15
Development Time	16
SUMMARY	17
ACKNOWLEDGMENTS	18
LITERATURE CITED	19
APPENDIX	21

#### INTRODUCTION

Internal feeding insects have been known to occur in stored grain since 196 B. C., when Flautus first reported a weevil in stored wheat. It is not known whether he was referring to the rice weevil, <u>Sitophilus oryzae</u> L. or granary weevil, <u>Sitophilus granarius</u> L. (Cotton, 1920).

Rice and granary weevils are listed as two of the four most destructive grain damaging insects that attack grain in shipment and storage; the other two, also internal feeders, are the lesser grain borer, <u>Fizopertha dominica</u> (F.) and the Angoumois grain moth, <u>Sitotroga cerealella</u> (Oliv.). These insects make the grain subject to secondary infestations by breaking the whole kernels, thus allowing other insects to feed on the damaged kernels (U.S.D.A. Farmers Bulletin 1260, 1958).

With the increase of population and urbanization of man a need for storage of more food and food products has arisen. These foods are usually stored as whole grains, rather than as finished products. The storage of grains for long periods of time provides the internal feeding insects with an ideal environment in which to live.

In recent years, there has been a tightening of the U. S. Food and Drug Administration regulations governing the tolerance of foreign materials in food products. The regulations now read that one percent or more of insect-damaged wheat kernels, as determined by weight, constitutes a threat to human welfare and is therefore subject to seigure (Food and Drug Administration, 1957). Now, hidden infestations can be detected easily by the use of the grain X-ray inspection unit. Internal feeding insects can not be removed by screening. Heavily infested wheat must not be used for human consumption and is restricted to animal food, industrial use, or must be

discarded. The use of chemicals in the control of insects in food material is becoming more restricted due to the hazard of residues poison; and anisals.

More economical and effective ways of controlling grain weevils must be found. To develop better insecticides and other control methods, more information on the ecology and life processes of the grain weevils must be obtained. To do this, it is necessary to be able to identify the various stages of these insects. Dyar found that the head capsule width of each instar is constant within each instar and may be used in the determination of its instar (Imms, 1924).

The object of this work was to determine the head capsule measurements of the four instars of the large strain rice weevil and the granary weevil when breeding in wheat under standard conditions and to compare these with other published measurements of larval rice and granary weevil head capsule widths.

#### REVIEW OF LITERATURE

#### Unknown Strain Rice Weevil

Origin and Distribution. The rice weevil was first described by Carolus Linnaeus in 1763 and the origin of this weevil is believed to have been in India (Cotton, 1920). The weevil was transported to Europe and is now cosmopolitan, being found in the temperate, sub-tropical, and tropical zones of the world (Pang-Hwa and Yen-Nien, 1935).

Adult Description. The color of the rice weevil is opaque, reddish-brown to piceous, with four rufous spots on the elytra. The beak is slender, cylindrical, slightly dilated at the base, three-fourths as long as the thorax, with four rows of rather coarse punctures on the dorsum and a slight fovea between the eyes. The thorax is longer than wide, constricted near the apex, with the sides feebly curved, and gradually divergent to its base, the disc densely, deeply and coarsely punctured. The elytra are oblong, slightly narrowed at the tip, deeply striate, striae very coarsely and closely punctured. Intervals of the alytra are slightly convex and narrow. Overall length is 2.1 to 2.8 mm. (Elatchley and Leng, 1916).

Riclogy. The biology was studied by Cotton (1920) and Howe (1952). The time of appearance of instars of the rice weevil were recorded by Cotton (1920), Howe and Oxley (1944), Howe (1952), and O'Donnell (1956). Lathrop (1914) described the egg laying of the rice weevil.

Head capsule measurements of an unknown strain rice weevil have been recorded by Cotton (1920), and of the small strain rice weevil by O'Donnell (1956).

#### Large Strain Rice Weevil

<u>Description</u>. Birch (1946) stated that the large strain rice weevil was described by Motschulsky in 1855. Motschulsky named it <u>Calandra</u>
<u>zea-mais</u> Motsch. The identification was based on the size and sculpturing of the pronotum and elytra.

A significant difference in size between the two strains was found by Birch (1946) and the following sizes were given for the two species after seven generations in corn and wheat:

Large	St	rain	Small	St	rain
Corn		Wheat	Corn		Wheat

Length from pronotum to tip of elytra 3.23 mm. 2.87 mm. 2.72 mm. 2.42 mm. Birch (1944) gave the following results for the large strain rice weevil

bred in wheat:

		Large Strain		Small S	Strain
		Male :	Female	Male :	Female
Pronotum length		1.22	1.20	1.02	1.04
Total length Maximum width	(mm.)	2.88	2.86	0.93	0.96

Biology. The large strain rice weevil has been found to lay fewer eggs and develops slower than the small strain weevil in wheat of 10 percent moisture. The two strains are inter-sterile (Eirch, 1944). The sex ratio of the large strain rice weevil is unity (Howe, 1952). Howe found the life history of the large strain rice weevil at 25 degrees Centigrade and 70 percent relative humidity to be as follows:

	First appearance day after laying	Median days from oviposition to end of instars
Egg		6.1
Instar 1	5 days	10.9
Instar 2	9 days	15.0
Instar 3	13 days	19.4
Instar 4	16 days	27.7
Pupa	24 days	34.6
Adult	28 days	
Emergence	32 days	
rmergence	32 days	

#### Granary Weevil

Origin and Distribution. The granary weevil is thought to be the curculio known by the Romans (Cotton, 1941). Linnaeus described the granary weevil in 1758 and named it <u>Calandra granaria</u> Linn. (Elatchley and Leng, 1916).

In distribution, granary weevils are cosmopolitan and in the United States are more commonly found in the Northern States (Cotton, 1941).

Adult Description. The granary weevil is elongate-oblong, feebly convex, chestnut brown to piceous in color and moderately shining. The

beak is two-thirds as long as the thorax, slender, cylindrical and finely and sparsely punctate. The thorax is sparsely punctate, punctures coarse and on the disc more or less fusiform. Elytra are deeply striate, the striae punctured at the bottom, not serrate. The intervals of the elytra are smooth, alternately wider and more elevated, especially toward the base. The striae have a row of elongate punctures. The sternum is coarsely and less densely punctured than in the rice weevil. The length is 3-4, mm. (Mlatchley and Leng, 1916).

<u>Biology</u>. Kunike (1936) studied the life history and environmental factors as affecting the life history of the granary weevil and determined its developmental time.

Stadium	Shortest develop- mental duration in days	Total develop- mental duration in days
Egg	6	6
Larva I	7	13
Larva II	6	19
Larva III	4	23
Larva IV	5	28
Prepupa	1	29
Pupa	5	34
Weevil in Korn	3	37

Kunike (1936) described the egg laying of the granary weevil and gave measurements of the egg as 0.64 mm. long and 0.3 mm. wide. He also recorded the head capsule widths.

#### Experimental Procedure

Robinson (1926) reported that relative humidity and temperature are important environmental conditions for insects. Moisture content of grain has an influence on the water content of insects.

Other problems considered were, amount of carbon dioxide present, number

of larvae per kernel, length of oviposition and age of adults. Richards and Oxley (1943) noted that carbon dioxide buildup will cause the mid-fourth instar larvae to eject frass through a minute hole in the kernel. Two weevil larvae in one kernel provide competition and retards development of one another. Internal feeding larvae should be from a culture of a short oviposition period (Howe, 1952). In a study of Tenebrio molitor Linn., Tracey (1958) found that the age of parents affected the rate of weight increase and length of the life cycle of their larvae.

Mason fruit jars may be used as culture jars for grain weevils. A cover of muslin is desirable to keep parasites and other insects out. All media should be sterilized to kill mites and insects which may be present (Peterson, 1955).

Frankenfeld (1948) developed a method of detecting egg plugs of grain weevils by staining with acid fuchsin.

#### EQUIPMENT AND MATERIALS

#### Rearing Facilities

All stock and experimental cultures were reared under controlled conditions of temperature and humidity in the stored grain insect rearing room at Kansas State College. Temperatures were held at  $80^{\circ}$  F.  $\neq$  two degrees and at 70 percent relative humidity  $\neq$  two percent.

An automatically controlled, industrial humidifier was used to blow a mist of water throughout the room. Air circulation was provided by a continually operated fan. Heat was provided by light bulbs controlled by a thermostat. Cooling was accomplished by circulating air over a series of pipes which had cool water flowing through them.

Other equipment used is as follows: Aspirator, electronic moisture tester, dissecting microscope, forceps, freezer, microscope alides, number three pinning needles, rotomatic sifter, seed cleaner, screen lids, standard sieves, weight per bushel tester, and wide mouth quart jars.

Expendable items used are as follows: acid fuchsin stain, balsam, filter paper, and paper towels.

#### Source of Stock Cultures

Weevils used in this experiment were obtained from stock cultures at Kansas State College. Stock cultures of granary weevil were originally obtained from the United States Department of Agriculture Stored Products Insect Laboratory at Manhattan, Kansas. At varying intervals adults were added from nearby granaries. The large strain rice weevil culture was obtained from Arkansas, courtesy of Paul Boles.

#### Culture Media

The culture medium used in this experiment was Pawnee variety of hard red winter wheat. The wheat was grown in 1956 in Allen County, Kansas and stored in cold storage in Manhattan, Kansas until needed.

The corn used for the general cultures of the large strain rice weevil
was a yellow variety obtained from a local grain dealer.

#### METHODS

#### Media Preparation

Grain used in this experiment was cleaned by the use of a Farm Master Seed Cleaner, using a letter A chaff screen and a number I double screen to remove dockage and broken kernels.

The grain was divided into 25 pound lots, sealed in metal containers, and placed in a deepfreezer. After seven days at minus zero degrees Fahrenheit, the containers were removed and allowed to attain room temperature.

When the temperature reached an equilibrium of approximately 80° F., the percentage moisture was determined by the use of a model 500 G Steinlite Moisture Tester. An Chaus Weight Per Bushel Tester was used to obtain the test weight of the grain.

To bring moisture content of the grain up to 13.5 percent, the following calculation procedure was used:

#### 100 - % of present water content 100 - % of desired water content

The first digit of the quotient (always one) was dropped, leaving the remainder of the quotient as a multiple factor that was multiplied by grams of wheat to be tempered. The product was the amount of water to be added to the grain to bring the moisture content to the desired level.

The correct amount of distilled water was then placed in the can of grain and the lid sealed with masking tape. Twice a day, the can of grain was rolled to insure the uniform moisture content of the grain. One week later, the moisture content was rechecked and the above process was repeated if the moisture content did not meet specifications of 13.5 percent £ 0.5 percent.

#### Stock Culture Procedure

Stock cultures of the rice and granary weevils were maintained under identical conditions, except that the large strain rice weevils were cultured in corn, and granary weevil in wheat. Five hundred grams of sterilized, 13.5 percent moisture grain were placed in each container. These containers were one quart, wide mouth, mason jars that had a 40-mesh screen replacing the metal cap. A piece of filter paper was placed in each lid to keep other insects from laying eggs through the screen.

Unsexed adult weevils were removed from the previous cultures by screening with a rotomatic sifter. Two hundred weevils were picked up in an aspirator and placed in each container for seven days. These cultures were placed in a rearing room and maintained at approximately 70 percent relative humidity and 80° F. At the end of the oviposition period, adults were removed from the grain by sifting the weevils through a number 10-mesh screen on a rotomatic aifter. The wheat was returned to the jar and the lid tightly put in place. The cultures were returned to the rearing room and maintained under identical conditions as the oviposition period.

#### Experimental Cultures

The experimental cultures were maintained in one quart wide mouth jars.

Metal lids were replaced with 40-mesh wire screen and a piece of filter paper.

One hundred and fifty grams of wheat were placed into each container. Next, 150 unsexed adult weevils were introduced into each jar and allowed to oviposit for three days under conditions of approximately 70 percent relative humidity and 80° F. At the end of this period, the adults were removed by hand screening. The grain was combined and thoroughly mixed, after which it was reweighed into 150 gram lots and placed back into the containers. The cultures were returned to the rearing room and maintained under identical conditions as the oviposition period.

#### Installation of the Calibrated Ocular

First, the left eyepiece was removed from the dissecting scope. Next, the lock mut was removed from the inside of the eyepiece. Inside the eyepiece above the lock mut there is a hollow cylinder. This cylinder was removed and the calibrated ocular micrometer inserted into the upper end of the cylindrical tube making sure that the micrometer numbers were right side up. A circular spring was placed on top of the micrometer to keep it from moving if the eyepiece was inverted.

The cylinder containing the micrometer was threaded into the ocular and adjusted until the lines were clear when viewed through the scope. The lock nut was threaded up until it rested against the cylinder. This eyepiece was returned to the dissecting scope.

#### Calibration of Microscope

Galibration of the microscope was accomplished by placing a calibrated slide on the stage of the scope. Then the number six objective lens was moved into place and the slide brought into sharp focus. The ocular micrometer was turned horizontal and the slide micrometer centered in the field of view.

The slide micrometer was then used to find the distance between the lines of the ocular micrometer. The slide micrometer was 2.0 mm. long and divided into 0.01 mm. On high power, number six objective lens, each division was equal to 0.0165 mm.

#### Staining of Grain

Daily, one jar of the experimental culture was removed from the rearing

room. The selected sample was thoroughly mixed and 100 kernels picked at random, omitting broken kernels. These selected kernels were placed in a strainer and immersed in water for three minutes to remove feeding material and to aid in the staining of egg plugs.

The wheat was then placed in acid fuchsin for one to five minutes, the time immersed varying with the concentration of acid fuchsin. Excess stain was rinsed off the kernels with tap water. Due to the possibility of water and stain entering the kernel in the last instar and prepupal stage, immersion time in water was reduced to 30 seconds and the time in acid fuchsin to one minute.

Egg pluge stain a deep cherry red and mechanical injuries a light pink.

Acid fuchsin stain formula is as follows:

Acid fuchsin	0.5	gm.
Glacial acetic acid	50.0	cc.
Distilled water	950.0	cc.

Glacial acetic acid was poured into the water and mixed. Next acid fuchsin was added to the glacial acetic acid and water mixture. This solution may be stored for a long period and used repeatedly until it becomes murky (Frankenfeld, 1948).

#### Dissection Procedure

Immediately after the grain was rinsed of excess acid fuchsin, the kernels were placed on a paper towal on the dissecting scope stage. Using low power, 15 kernels with egg plugs were removed from the sample and 10 larvae were removed.

Removal was accomplished by holding a kernel with forceps and cutting it open with a scalpel. In the first two instars, the larvae could be found more readily by making a small notch on one side of the egg plug, then making a cut on the opposite side of the egg plug and with a twisting motion of the scalpel, the kernels were opened. The kernels usually cracked across the larval tunnel. Latter instars could be found more easily by splitting the kernel down the crease. Each larva was placed in a gelatin capsule until measured.

Each larva was placed on a glass microscope slide and the head capsule removed. Two insect pinning needles were used to cut off the head capsule.

#### Measurement

Head capsules were mounted in balsam on a microscope slide. The head was placed with the occipital foramen down and the coronal suture vertical. Each head capsule was dissected, mounted and measured separately.

Measurements were made using a calibrated ocular in a broadfield, dissecting microscope. The scale of the ocular was horizontal and the head capsule measured in the center of the field of view. High power of 90 X was used. Each calibration mark of the ocular was determined to be equal to 0.0165 mm., and the head capsule width was estimated to quarters of a calibration or 0.0061 mm.

#### RESULTS AND DISCUSSION

In Tables 1 to 4, 1 the head capsule widths of the various instars of the granary weevil ranked from the smallest to the largest are shown. The numbers observed by replicates are included as well as the total for the whole study. Total number of larvae observed are recorded by instar for

<sup>1</sup> All tables in the Appendix.

each replicate and combined replicates.

Tables 5 to 8 show the head capsule widths of the large strain rice weevils, ranked from the smallest to the largest. The same information as in Table 1 is recorded here.

In Table 9, the head capsule widths of <u>Sitophilus</u> from several sources, including known large and small strains, have been rounded off to two digits. The head capsule measurements are arranged by instar for minimum, mean and maximum widths, except where only the means were recorded.

In Table 10, the observed mean head capsule widths for each instar is compared with the calculated widths obtained by Dyar's Law for the large strain rice weevil and the granary weevil. The differences are shown in the right hand column. A constant was found by dividing each instar mean by the one preceding it. The quotients are added and the sum is divided by the number of divisions made.

Table 11 shows the variance of the head capsule measurements which were obtained by subtracting the smallest larval head capsule width from the largest width within the instar. This information is listed by instar for the small strain rice weevil, large strain rice weevil and granary weevil.

In Table 12, the largest number of head capsules observed in each instar is recorded in days from the first day that the parent stock was introduced for oviposition. The pupae are listed as the highest number observed, but the peaks may not have been reached at the termination of the experiment.

Plate I is a graphic illustration of the head capsule widths of the granary weevil larvae in Tables 1 through 4, except that the numbers were

<sup>1</sup> All Plates in the Appendix.

rounded off to two digits at the right of the decimal. The widths are plotted with the frequency of observation.

In Plate II, large strain rice weevil head capsule width data were taken from Tables 5 through 8 and plotted as in Plate I.

Plate III is a graph of the number of head capsules of the granary weevil observed by instars in days from the first day of exposure of the wheat to adult weevils for oviposition. The forms shown include egg, first, second, third and fourth instars, and pupe up to the termination of the experiment.

Plate IV shows the same information for the large strain rice weevil as in Plate III for the granary weevil.

#### Head Capsule Width

Head capsule widths of the granary and large strain rice weevils, as shown in Tables 1-3 and graphically illustrated in Plates I and II, show the divisions between the larval instars. Plates I and II do not show as wide a division between instars as Tables 1-8. This is due to the rounding off of numbers to two digits in order to be directly comparable with other published results. The divisions were made from the data in Tables 1-8, the largest break being used as the division. The break between the third and fourth instars of the large strain rice weevil does not coincide with the fewest number of head capsules (Plate II).

The number of head capsules in an instar rises to a peak, drops off sharply and then tapers off on the larger side.

Mean head capsule widths as shown in Table 9, were compared with

Dyar's Law (Imms, 1924). The largest deviation of the observed mean from
the calculated mean was 0.02 mm. in the fourth instar of the granary weevil.

Analysis of variance showed that 97 percent of the total variation was due to instar effect.

The number of granary weevil larvae observed in each instar is shown in Tables 1-4 as 154, 127, 128, and 261 respectively for instar one through four. Differences in the numbers observed in the four instars were found to be due to the differences in the length of each instar and the overlapping of instars, thus several instars being found on the same day. For example, on the 17th day there were found to be one 1st, eight 2nd, twenty 3rd and one 4th instar larvae present.

The length of time of each instar was 11, 15, 19 and 16/ days for instar one through four respectively (Plate III). Due to the termination of the head capsule measurements on the 33rd day, the fourth instar appears to be shorter than the third. If the fourth instar were extrapolated to completion of all larval development, it would undoubtedly be the longest instar.

Since a few of the specimens develop very slowly, the head capsule measurements were terminated when a low level of larvae was found.

In Tables 5-8, the number of large strain rice weevils observed in each instar is shown as 162, 129, 137 and 258 for instars one through four respectively. Flate IV of the large strain rice weevil gives the following days in each instar for instars one through four respectively: 10, 7, 9, and 11\(\frac{1}{2}\). As with the granary weevil, the differences are mainly due to the overlapping of other instars and length of instars.

#### Variation Within Instars

Head capsule width variation within the instars of the granary weevil is least in the first instar and increased through the remaining three instars. The large strain rice weevil instar variation is least in the second instar, being greater in the first, third and fourth instars respectively, as shown by Table 12. A similar order of variation within instars was also obtained for the small strain rice weevil by O'Donnell (1956).

#### Development Time

Development of the large strain rice weevil from egg to pupa was much shorter than the development of the granary weevil (Plates III and IV). This may be due to the time of the year the insects were reared. It is possible that weevil development slows down in the winter even though conditions remain constant in the rearing room. Granary weevils were reared November 23 through March 5, 1957, while the large strain rice weevils were reared March 13 through May 17, 1958. Table 12 shows that differences in developmental time between the two weevils increased with each successive larval instar and pupa.

The number of larvae in any instar rises abruptly to a peak, drops off sharply and then tapers off gradually in the latter days as shown by Plates III and IV. This gradual tapering off may be the result of natural variability of the weevils, some abnormality within wheat kernels, or the three day oviposition period.

Overlapping of instars is greatest during the fourth instar, followed by the third, second, and first instars respectively. This overlapping may be due to one or to a combination of the following possible reasons: the three day oviposition period, an inherent variation in the weevils, or variations in the mutritive values of the wheat kernels.

#### SIDMARY

This experiment was designed to obtain head capsule width measurements and times of development of the four larval instars of the granary and large strain rice weevils.

The granary and large strain rice weevils were allowed to oviposit for three days on 150 grams of sterile Pawnee hard red winter wheat of 13.5 percent moisture. The weevils were removed at the end of the oviposition period and the cultures were maintained in the rearing room at  $80^{\circ}$  F. and a relative humidity of 70 percent until needed.

Each day, a random sample of weevile was removed from the rearing room. The sample was stained with acid fuchsin and 15 infested kernels removed. Ten kernels were dissected and the larvae removed. The head capsules were removed and mounted on a microscope slide in balsam. The head capsule widths were recorded.

The measurements were grouped by obvious divisions present. Instar mean head capsule widths compared favorably with Dyar's Law. Statistically 97 percent of the variation was due to instar effect. Variation within instars was least in the second instar of the large strain rice weevil and increased through the first, third and fourth instars, respectively. The granary weevil increased in variation from the first through the fourth instar.

There was considerable overlapping of the instars in the period of development.

#### ACKNOWLEDGMENTS

Grateful acknowledgment is made of the assistance and guidance of Professor Donald A. Wilbur in the conduct and execution of this research problem. Thanks are also due to Gary F. Krause for his assistance in the statistical analysis of the problem.

#### LITERATURE CITED

Elatchley, W. S. and C. W. Leng. Rhynchophora or weevils of north eastern America. Indianapolis: Nature Publishing Company, 1916.

Birch, L. C. Two strains of <u>Calandra oryzae</u> L. (Coleoptera). Australian Jour. Expt. Biol. and Med. Sci. 22:271-275. 1944.

The influence of food on the size of two strains of Calandra oryzae L. Australian Jour. Expt. Biol. and Med. Sci. 24:123-125. 1946.

Cotton, R. T. Rice weevil, (Calandra) <u>Sitophilus orygae</u>. Jour. Agr. Res. 20(6):409-422. December 15, 1920.

Insect pests of stored grain and grain products. Minneapolis, Minnesota: Burgess Publishing Company, 1941.

Frankenfeld, J. C. Stain methods for detecting weevil infestation in grain. U. S. Dept. Agr. Bul. ET-256. July, 1948.

Food and Drug Administration.
Grain sanitation program of the Food and Drug Administration.
U. S. Dept. of Health, Education and Welfare, U. S. Govt. Prin.
Office, April, 1957.

Howe, R. W. The biology of the rice weevil <u>Calandra orygae</u> L. Ann. of Appl. Biol. 39(2):168-180. June, 1952.

Hows, R. W. and T. A. Oxley. The use of CO<sub>2</sub> production as a measure of infestation of grain by insects. Rul. Ent. Res. 35(1):11-22. 1944.

Imms, A. D. A general textbook of entomology. New York: E. P. Dutton and Co., 1924.

Kunike, Georg.
Beitrage zur lebensweise und Bekampfung des kornkafers, <u>Calandra granaria</u> L. (Curculionidae). Zeitschrift Fur Angewandte Entomologie 23(2):303-326. Juli 1936.

Lathrop, Frank H.

Egg-laying of the rice weevil, <u>Calandra oryzae</u> Linn. Ohio
Naturalist 14(7):321-327. May, 1914.

O'Donnell, Albert.

The determination of larval instars of the rice weevil <u>Sitophilus</u> orygae L., in wheat. Unpublished M.S. thesis, Kansas State College, Manhattan, Kansas, 1956.

Pang-Hwa, Tsai and Chang Yen-Nien.

Experimental studies regarding the influence of temperature and relative humidity on the oviposition of the rice weevil (Calandra Orygae L.). Agr. Sinica 1(6):175-188. May. 1935.

Peterson, Alvah.

A manual of entomological techniques. Ann Arbor, Michigan: Edwards Brothers, 1955.

Richards, O. W. and T. A. Oxley.

The ejection of frass by larvae of <u>Calandra</u> (Col. Curculionidae) under the influence of CO<sub>2</sub>. Proc. R. Ent. Soc. London. May, 1943.

Robinson, William.

Low temperature and moisture as factors in the ecology of the rice weevil, <u>Sitophilus oryzae</u> L. and the granary weevil, <u>Sitophilus granarius</u> L. Univ. Minn. Agr. Expt. Sta. Tech. Bul. 41. September, 1926.

Stored Grain Pests.

U. S. Dept. Agr. Farmers Bul. 1260. 1958.

Tracey, Kathleen M.

Effects of parental age on the life cycle of the mealworm, <u>Tenebrio molitor</u> Linnaeus. Ann. Ent. Soc. Amer. 51(5):1429-432. September, 1958.

APPENDIX

Table 1. Head capsule widths of first instar granary weevil larvae.

Head capsule width (mm.)	Replicate	Replicate 2	Replicate: 3	Replicate	: Total
0.172		1	-		1
0.176	-	1	-	-	1
0.213	1	-	-	-	1
0.221	3	1	-	-	4
0.223	_	-	4	1	5
0.225	1	-	-	-	1
0.230	4	2	-	-	6
0.231	_	-	5	2	7
0.234	-	-	1	-	1
0.235	-	tesp	-	3	3
0.238	4	4	-	-	8
0.239	400	des	3	2 5	1 3 8 5
0.243	-	-	6	5	11
0.246	10	8	-	-	18
0.248	-	-	7	6	13
0.252	-	-	3	10	13
0.254	4	6	-	-	10
0.256	-	1	7 3	5	13
0.260	-	-	3	1	4
0.262	-	3	-	-	3
0.264	-	-	9	9	18
0.268	-	-	-	1	1
0.271		1	-	-	1
0.272	-		1	1	2
0.276	-	-	-	2	1 2 2 1
0.281	200	-	-	1	1
0.285	- (	-	1		1
Total	27	28	50	49	154

Table 2. Head capsule widths of second instar gramary weevil larvae.

Head capsule width (mm.)	Replicate 1	Replicate 2	Eeplicate: 3	Replicate 4	Total
0.303	1	-	-	-	1
0.305	-	40	2	-	2
0.309	00	-	3	40	2 3 1 2
0.310	1	400	olas	Gar.	1
0.312	1 2	1		000	2
0.320	2	2	-	***	4
0.322	-	-	1	-	1
0.326	***	-	2	-	2
0.328	5	5	-	-	10
0.330	**		7 2	2 - 2	8
0.334		**	2	2	
0.336	3				3
0.338		-	3 2	2	5
0.342		-	2	-	2
0.344	3	2		-	3 5 2 5
0.347	-	-	4	4	8
0.351	-	-	4	4	10
0.353	3	1	-		
0.355			4 2	2 3	4654734711
0.359	-	**	2	3	5
0.361	2	2		-	L
0.363			4	3	7
0.367			4	3 2	3
0.369	1	3	-	-	4
0.371	44	-	1	6	7
0.375	-	-	-	1	1
0.377	-	1		40	1
0.380	-		1	3	4
0.388	60	-	1	3	2
0.392	-	-	600	1	1
0.394	1	1	-	-	2
0.402		1	-		2 1 2 1 1
0.408	1.4	-	**	1	
0.417	90		**	1	1
0.423	-	-	2	-	1
0.425	40	440		1	1
Total	23	19	47	38	127

Table 3. Head capsule widths of third instar granary weevil larvae.

Head capsule width (mm.)	Replicate	Heplicate 2	Replicate 3	Replicate	Total
				-	_
0.446	-	-	1	1	1 2
0.450		***	*	1	1
0.451		2	-	-	2
0.454	-	•	1	-	1
0.458	-	-	1	-	1 4 3 4 1 6 3 1 2
0.459	1	3	-	**	4
0.462	-		2 1 2 3 3 3	-	3
0.467	3	1	-		L
0.470	_		1	-	3
0.476	1	5	-	-	4
	-	9	0	1	0
0.479	-		2		3
0.483		-	1		1
0.484	1	1		-	2
0.487		- 1	3	1	4
0.491	-	-	3		la
0.492	7	3		**	10
0.495			2	1	3 4
0.499	-	2	-	4	4
0.500	2	2	-		la
0.503		-	2 2	2	L
0.507			2	1	3
0.512			2	5	7
0.516			2	2 1 5 3	- 6
	4	ī		2	-
0.517		7	-	3	3
0.520	-			1	7
0.524	ī	-	T.	2	4
0.525	1	1	3	3 2	- 4
0.528	440	-	3	3	6
0.532		-	-	2	2
0.536	-	-	don	1	1
0.540	-	100	1		2
0.543	ī	1	-	40	2
0.545		1	-	4	4
0.549	-		1	1	2
0.557		1	40	1 1	1
0.558		3	-	-	1
0.561	-	-	-	1	1
0.501			1	1	2
0.565	-		î	-	1
0.573		-	2	_	44375512262122421121212124112
0.578		-	~	-	3
0.582	1		1		3
0.586	-	-	1	-	-
0.590	1	-	1		2
0.594	**	00	1	3	4
0.598	1	-	-	-	1
0.599	1	**	-	-	1
0.606		tie	2	-	2
Total	25	21.	41	42	1.28

Table 4. Head capsule widths of fourth instar granary weevil larvae.

Head capsule width (mm.)	Replicate	Replicate 2	Replicate	Replicate	Total
0,615		2	-		2
0.623		-	_	1	î
0.627	_	-	_	2	-
0.631	-	1		2	2 2 5 5 6
	3		2 2	3	26
0.635	-	-	4	3	2
0.639	-	-	2	3	2
0.640	3	3	-	-	0
0.642	1	-	-	-	1
0.644	-	-	2 2	3	13
0.648	3 2	5	2		
0.650			-	1	2
0.652	-	-	3	1 .	4
0.656	4	4	35   5431453572222   33   5   221212	1	14
0.658		1		-	1
0.660	-	-	5	6	11
0.664	3	1	4	4	12
0.668		-	3	4 2	5
0.672	4	4	1	ĩ	10
0.677		46	-	4	
	3		4	5 8	9
0.681	2	4	5	8	22
0.685	-	-	3	6	9
0.689	2	1	5	9	17
0.693	-	400	7	4	11
0.697	3	3	2	1	9
0.701	-	2	2	4 6	6
0.705	1	2	2	6	11
0.710	-	-	2	5	7
0.713	2	2		5 2	h
0.724		-	3	2	5
0.718			3	i i	4
0.722	2	2	2		10
	4.	3	-	3	8
0.726	ī	2	>	2	4
0.730	1		-	1 5 3 1 2 1	
0.734	ī	-	2	2	4
0.738	1	-	2	1	lo.
0.743	-	-	1		3
0.751	-	-	2	-	2
0.755	-	-	1	ī	1
0.759	-	000	2		3 2 1 3 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1
0.763	1	-	1	-	1
0.771	-	•••	1	-	1
0.776	-		1	-	1
0.780	-	-	1	-	1
0.792	40		•	1	1
0.813			1	-	1
0.821	_	-	-	1	î
O.DKT	-				-
					261

Table 5. Head capsule widths of first instar large strain rice weevil larvae.

Hend capsule width (mm.)	Replicate:	Replicate: 2	Replicate 3	Replicate	! Total
0.182	1	-	-	-	1
0.194	2	400	**	2	. 4
0.198	2	2	1	2	. 7
0.202	2	3	la la	1	10
0.206	1	3	1	3	8
0.210	1	2	ds	4	11
0.215	10	14	5	2	31
0.219	A,	h	7	7	22
0.223	-	1	5	5	10 8 11 31 22 11 15
0.227	4	1.	4	6	15
0.231	8	4	la .	3	14
0.235	2	2	-	1	?
0.239	2	1	-	1	44
0.243	2		-	7	2
0.252	2	7	-	-	2
0.256	7	-	_	7	
0.264	1	-	1	1	2
0.268	1	2	-	-	3
0.272	-	-	1	-	1
Total	46	40	37	39	162

Table 6. Head capsule widths of second instar large strain rice weevil larvas.

Head capsule width (ma.)	Replicate	Replicate: 2	Replicate 3	Replicate	Total
0.276	-	1		-	1
0.281	-	400	1	-	1
0.285	-	2	1	1	4
0.289	-	1	1	1	3
0.293	2	1	2	2	7
0.297	da	3	1	1	9
0.301	1	2	2	4	9
0.305	-	5		3	8
0.309	3	2	3	2	10
0.314	5	la .	3	4	16
0.318	2	2	1	2	7
0.322	1	4	1	2	8
0.326	2	4	2	4	12
0.330	3	1	5	3	12
0.334	1	1	-	-	2
0.338	1	2	-	2	5
0.342	2	1	-	1	3
0.347	1	2	1	-	4
0.351	-	1	1	-	2
0.355	1	00	-	-	1
0.363	-	-	2	3	>
Total	28	39	27	35	129

Table 7. Head capeule widths of third instar large strain rice weevil larvae.

Head capsule width (mm.)	1 1	Replicate 1	2 2	Replicate 2	:	Replicate 3	: :	Replicate 4	2 2	Tota:
0.380		-		1		-		-		1
0.388		-		_		1		-		1
0.392		-		-		-		1		1
0.396		-		1				1		2 2 3 2
0.400		-		-		2		-		2
0.413		1		-		1		1		3
0.416		1		1		-		-		
0.425		-		1		-		-		1
0.429		-		2		3		-		5
0.433		2		-		3		1		5 4 5 3 3 8
0.437		1		1		7		2 2		5
0.441		-		1		00		2		3
0.446		1				1		1		3
0.450		1		3		1		3		
0.454		1		1		-		2		4
0.458		3		3				1		7
0.462				2		3		1		7
0.466		2		2		3 3 1		1		6
0.470		-		3		1		-		4
0.474		2				-		1		40
0.479		4		3		3 3		3		11
0.483		1		-		3		1		5
0.487		2		-		3		1		6
0.491		1		1		-		-		2
0.495		3		1		2		3		9
0.499		1		1		1				2
0.503		-		-		2		1		3
0.507		1		1		1		7		3
0.516		7		T		4		1		5629233352231
0.520		2		-		4		-		2
0.524		-		2		_		_		2
0.528		_		1		2		_		3
0.532		_		4		2		-		3
0.536		1						-		î
0.545						1		-		ī
0.557		1		-		-		-		î
0.561		-		-		-		1		î
0.565		-		-		1		2		3
Total		33		31		40		33		137

Table 8. Head capsule widths of fourth instar large strain rice weevil larvae.

0.578 0.582 0.586 0.590 0.594 0.598 0.602 0.602 0.611 0.615 0.619	1 4 1 2 2 3	1 1 1 5 1 - 1 4 1 1 - 1	1 3 2 1	3 1 1 1 1 1 1 2	422647338
0.582 0.586 0.590 0.594 0.598 0.602 0.606 0.611	1 2 3	1 1 5 1 - 1 4 1	1 3 2 1	-	2 2 6 4 7 3
0.586 0.590 0.594 0.598 0.602 0.606 0.611 0.615	1 2 3	1 4 1	1 3 2 1	-	2 6 4 7 3
0.594 0.598 0.602 0.606 0.611 0.615 0.619	1 2 3	1 4 1	3 2 1	-	6 4 7 3
0.594 0.598 0.602 0.606 0.611 0.615 0.619	1 2 3	1 4 1	3 2 1	-	7 3
0.598 0.602 0.606 0.611 0.615 0.619	2 3	1 4 1	3 2 1	-	7 3
0.602 0.606 0.611 0.615 0.619	2 3	1	1	1	3
0.611 0.615 0.619	2 3	4	1	1	
0.611 0.615 0.619	3	4	en .	2	3
0.615	3	1		6	8
0.619			As .	5	10
0.623			-		3
	-		2	2	4
0.627	2	3 1 2	4	4	11
0.631	2	3	2	4	11
0.635	2	í	2	4 2	7
0.639	2	2	1	2	7
0.664	4	3	2212225555122211122	3 4	12
0.648	2	3 2	2	4	10
0.652	2	-	2		4
0.656	2	2	5	3	13
0.660	-	l.	3	7	14
0.664	6	5 3 1 3	3	3 7 1	14
0.668	5 2	3	í	-	6
0.572	2	1	2	2	7
0.677	8	3	2	4	17
0.681	2	í	2	2	7
0,685		600		-	1
0.689	5	2	1	3	11
0.693	6	3	1	1	11
0.697	_		2	3	5
0.701	-	-	2	-	2
0.705	1	2	3	2	8
0.710		2 2	3 4	-	6
0.714	2	1		-	3
0.718		-		1	1
0.722	-	1		-	1
0.726	-	1	-	2	528631122321111
0.734	1		1	-	2
0.743	î			2	3
0.747	-		1	1	2
0.751	-		44	1	1
0.759		-	1		1
0.763		1			1
0.796	-	-	ī	-	1
Total	66	58	63	71	258

Table 9. Width of head capsules of larval Sitonhilus spp. (in millimaters).

	ls.	t Insta	Let Instar 2nd Instar 3rd Instar 4th Instar	2nd	Instar	IEX.	3r	Instal	The state of	Man	Instar	Hax
Small Stradn S. Orrza O'Dounell '56 Wheat	1	0.30	0.16 0.20 0.22 0.25 0.28 0.29 0.34 0.39 0.43 0.49 0.54 0.64	0.25	0.28	0.29	0.3%	0.39	0.43	0.49	0.54	0.64
Large Strain S. orras Wheat	0.18	0.22	0-18 0.22 0.27 0.28 0.32 0.36 0.38 0.47 0.57 0.58 0.66 0.80	0.28	0.32	0.36	0.38	0.47	0.57	0.58	99.0	0.80
S. gronarine	0.17	0.25	0.17 0.25 0.29 0.30 0.35 0.43 0.45 0.51 0.61 0.62 0.68	0.30	0.35	0.43	54.0	0.51	19.0	0.62	99.0	0.82
S. oryan Cotton '20 Corn		0.22			0.32			8700			0.64	
S. granariue Kunike '36 Kheat		0.3			0.32			94.0			79.0	

Table 10. A comparison of the calculated and observed mean head capsule widths (in millimeters) for the four instars of the large strain rice weevil and the granary weevil using Dyar's Law.

Instar	Dyar's Law calculation		Observed	Difference:
		Large Strain Rice W	<u>leevil</u>	
1		-	0.22	0.0
2	1.44 X 0.2	2 0.32	0.32	0.0
3	1.44 X 0.3	2 0.46	0.47	0.01
4	1.44 X 0.4	6 0.66	0.66	0.0
		Granary Weevil		
1			0.25	0.0
2	1.40 X 0.2	5 0.35	0.35	0.0
3	1.40 X 0.3	5 0.49	0.51	0.02
4	1.40 X 0.4	9 0.69	0.68	0.01

Table 11. Variance of head capsule measurements between the largest and smallest individuals within an inster (in sm.).

	lst instar	2nd inetar	3rd inster	Ath instar
Small strain rice weevil	0.06	0.04	0.09	0.15
Large strain rice weevil	0.09	0.08	0.19	0.22
Granary weevil	0.12	0.13	0.16	0.20

Table 12. Peak numbers of head capsules observed in each instar and pupa (in days).

	instar i	2nd instar	3rd instar	4th instar	Pupa
large strain rice weevil	10	12	15	19	25/
Granary weevil	9	24	17	25	33/
Difference	1	-2	-2	-6	-84

## EXPLANATION OF PLATE I

Head capsule widths of granary weevil by instar and number observed. Data used were from Tables 1 through 4. The data were rounded to two figures to the right of the decimal.

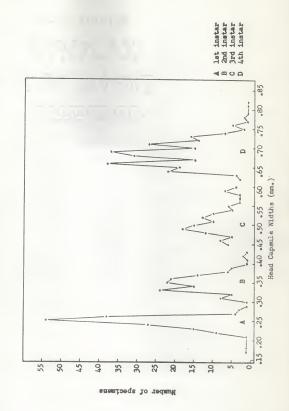


PLATE I

# EXPLANATION OF PLATE II

Data used were taken from Tables 5 through 8. The data were rounded to two figures Head capsule widths of large strain rice weevil by instar and number observed. to the right of the decimal.

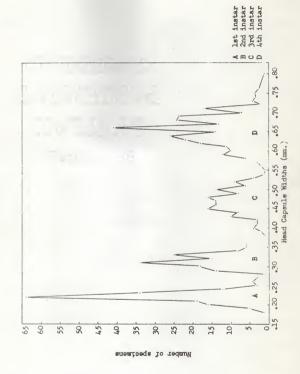
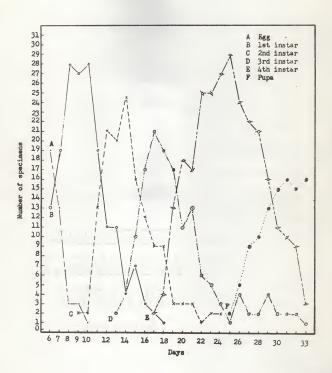


PLATE II

#### EXPLANATION OF PLATE III

Number of granary weevils observed in egg, lst, 2nd, 3rd, 4th instars and pupa in days from first day of exposure of the wheat to the adult weevils for oviposition.

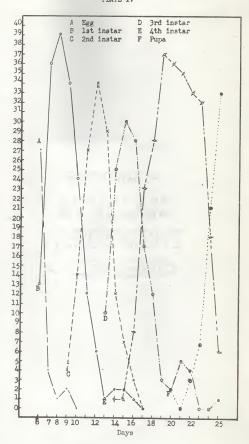
PLATE III



#### EXPLANATION OF PLATE IV

Number of large strain rice weevils observed in egg, instar lst, 2nd, 3rd, 4th, and pupa in days from first day of exposure of the wheat to the adult weevils for oviposition.

PLATE IV



Number of specimens

### HEAD CAPSULE MEASUREMENTS OF THE LARVAL INSTARS OF THE LARGE STRAIN SITOPHILUS ORYZAE L. AND SITOPHILUS GRANARIUS L., IN WHEAT

by

#### EDWIN LOREN SODERSTROM

B. S., California State Polytechnic College, 1957

AN ABSTRACT OF A THESIS

submitted in partial fulfillment of the

requirements for the degree

MASTER OF SCIENCE

Department of Entomology

KANSAS STATE COLLEGE
OF AGRICULTURE AND APPLIED SCIENCE

The object of this work was to determine the head capsule measurements of the four instars of the large strain rice weevils <u>Sitophilus oryzae</u> L., and granary weevils <u>Sitophilus granarius</u> L., breeding in wheat under standard conditions and to compare these with other published measurements of larval rice and granary weevil head capsule widths.

Grain used in this experiment was cleaned, sterilized and tempered.

One hundred and fifty grams of 13.5 percent moisture wheat were placed into each container. One hundred and fifty unsexed adult weevils were allowed to oviposit for three days, after which they were removed and the cultures returned to the rearing room. The rearing room was maintained at approximately 70 percent relative humidity and 80° F.

Daily, one jar of the experimental culture was removed from the rearing room and a sample of 100 kernels was removed and stained in acid fuchsin.

After staining, 15 kernels with egg plugs were removed from the sample and 10 larvae dissected out.

Each larva was placed on a glass microscope slide and the head capsule removed. The head capsules were mounted in balsam, on a microscope slide, with the occipital foramen down and the coronal suture vertical.

Measurements were made using a calibrated ocular in a broadview, dissecting microscope. Under high power of 90 % enlargement, head capsules were measured to 0.0165 mm. and estimated to 0.0041 mm.

For comparison, the minimum, mean and maximum head capsule widths by instars are shown in tabular form. Mean head capsule widths were compared with Dyar's Law and found to be comparable, the largest deviation being 0.02 mm. Analysis of variance showed that 97 percent of the total variation was due to instar effect.

Head capsule width variation within the instars of the granary weevil

was found to be least in the first instar and increased through the remaining three instars. The large strain rice weevil instar variation was least in the second instar, increasing in the first, third and fourth instars respectively. A similar order of variation within instars, as found in the large strain rice weevil was also recorded for the small strain rice weevil by O'Donnell (1956).

The developmental period of the large strain rice weevil from egg to pupe was shorter than that of the granary weevil.