Heat as a Means of Controlling the Angoumois Grain-Moth. (I.)

By

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Introduction.

The storage of grain in Japan, and especially the storage of wheat and barley is made very difficult by the attacks of the Angoumois grain-moth (*Sitotroga cerealella* OLIV.), which causes immense damage every year. Till recently the most widespread, and the only practical method of combatting this pest was fumigation. For this purpose carbon bisulphide has generally been used, but chloropicrin, used at the rate of from half a pound to a pound per thousand cubic *shaku** over a period of two or three days is displacing it.

Recently complaints have been made that fumigation with chloropicrin carried out under these conditions was unsuccessful, and a certain agricultural expert asked the writers if there were any good method of controlling the pest besides fumigation and also if it were possible to use heat for this purpose.

Available literature on this subject of controlling the Angoumois grain-moth by the application of heat revealed that no such experiment had ever been conducted in Japan, but drying harvested wheat and barley in the sun to avoid the damage by this pest had been sometimes recommended. In foregin countries, especially in the United States of America, heat seems to have been used frequently to control the insect pests of store houses and mills. According to F. M. WEBSTER, "a temperature of 140°F.(=60°C.), continued for nine hours, literally cooks the larvae and pupae of the Angoumois grain-moth and a temperature of 130°F.(=54.4°C.) for five hours is fatal, as is also 120°F.(=49°C.) for four hours, while 110°F.(=43.3°C.) applied for six hours was only partially effective."[1] G. A. DEAN conducted experiments on the heat control of mill insects and found that a temperature of 115°F.(=46°C.) for a period of twelve hours or a temperature of 125°F.(=51.7°C.) for a period of several hours proved fatal to all the insects in their various stages.[1, 2] DENDY and ELKINGTON considered that exposure to a temperature of 145.5°F.(=63°C.) for five minutes is sufficient to kill the larvae of

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* One cubic "*shaku*" is equal to approximately one cubic foot.

Calandra oryzae and probably to sterilise the wheat completely as regards all insect life, though they did not extend their experiments to the Angoumois grain-moth.[3]

In addition to the reports cited above, there are yet other descriptions of the heat treatment of stored grain, but very few experimental investigations seem to have been carried out.[4] Recently, E. F. GROSSMAN published a report on the results of his extensive experiments, which were carried out to control insect pests in stored corn. Although he does not seem to have conducted exhaustive experiments with the Angoumois grain-moth, he makes the following statement: "Heating test showed that an exposure for 30 minutes at 49°C. killed all the adults, though 2 per cent of the adults withstood an exposure of 15 minutes. The larvae and pupae could withstand a slight increase of heat, but could not live through an hour's exposure at 50°C."[5]

Whether heat treatment of wheat or barley with the object of preventing injury by the Angoumois grain-moth is practicable under present conditions among the farmers in Japan is open to question. However, should conditions change so that it would be possible to carry out artificial drying of wheat or barley by means of heat and if it were found that the control of all the stages of the Angoumois grain-moth by heat treatment is practicable, it is probable that this method may be adopted on a large scale. Thus, the first important question to be solved is the degree of resistance of the Angoumois grain-moth to high temperature. Therefore, the writers undertook experiments in order to learn resistance to heat of the egg, the larva and pupa of the Angoumois grain-moth under conditions which are more nearly natural than those obtained in the experiments which were carried out by the earlier workers. The experiments of the writers have not yet been concluded, but the results thus far obtained are reported in the present paper as the first report.

Method of Experimentation.

Heat may be applied in two different manners; namely, either by steam or by hot air. It was feared that heating with steam might affect the vitality of the seeds since vapour at a high temperature increases their moisture content. Therefore, the writers used the latter method, namely heating by dry hot air. A simple, home-made electric incubator was used to heat the grain used in this experiment. The thermostat used for this incubator was not sensitive so that the temperature of the air inside of the incubator varied about 5°C. above and below the required temperature when the incubator was left to the automatic regulation of the thermostat. Therefore, in experiments of shorter duration of heating, the experimenter himself regulated the temperature of the incubator. In this case, the variation of the air temperature of the incubator was very much smaller, being in most cases ±0.3°C.

Test animals may be subjected to heating in two different ways; namely, (1) larvae or pupae which are taken out of the grains may be exposed to high temperature or (2) they may be heated together with the grains in which larvae or pupae are found. According to the former method, the behaviour of the test animals can be observed closely during the course of an experiment, but a serious drawback to this method is that the conditions under which experiment is carried out are far from natural for the Angoumois grain-moth. Therefore, the writers did not adopt this method. In the writers' experiments, grains with eggs or those in which larvae or pupae of the Angoumois grain-moth were found were mixed with about 15 cc. of healthy wheat, put into a small cheese-cloth bag and placed in the incubator. Several drawbacks were found to be inseparable from this method also. Two of the more serious of them are: (1) that the stage of development of the larva in grain can not be known from outside, for example, distinction between the pupa and larva can not be made; and (2) that it can not be decided whether the insect in grain is alive or dead when it is used for experiment. Notwithstanding these drawbacks to the second method, the writers adopted it, since they desired to carry on experiments under conditions which are as nearly natural as possible to this insect.

Several pairs of adults were enclosed in a glass jar with a small quantity of wheat in it. The grains were examined every morning and the eggs which were laid on wheat grains were taken out of the jar together with the wheat grains. The eggs were counted using a magnifier and 100, or in certain cases, 200 eggs were placed in a Petri-dish, together with about 15 cc. of wheat to rear the larvae that hatched.

For each experiment 100 or 200 eggs or larvae were used. In most cases nearly fullgrown larvae were used for the experiment, but a small percentage of pupae might have been mingled with them. Each time two or three experiments were conducted under different temperatures using larvae that originated from the eggs which had been laid on the same day or in a few cases on two successive days. A certain number of the test animals were always set aside as control animals for the experiments.

Although 100 or 200 eggs were placed in Petri-dish and the larvae hatching out of them were reared, the percentage of the larvae which attained to the last larval stage was by no means always the same. It varied approximately from 50 to 70 per cent in most cases. Wheat grains which contained these fullgrown or nearly fullgrown larvae could be distinguished when the grains were carefully examined from outside. Therefore, it was possible to learn approximately the number of larvae which were suitable for experiment. This was an important step for calculating the percentage of the larvae that were killed by heat treatment. Stages of growth which the larvae had been able to attain before they were used for the experiment were not necessarily the same, even among the larvae which had hatched out from the same batch of eggs. Under these circumstances, a certain error, but of incosiderable degree, might have crept into the percentage of larvae that were killed by heat treatment. This could not be avoided when the experiments were conducted by this method.

Wheat grains which had been exposed to heat were kept in Petri-dishes until the emergence of the moths was over, when they were cut open to determine the number of living larvae and pupae. A certain number of larvae remained in the larval stage even when they were fullgrown and not killed by heat treatment, and some of them may even overwinter. This could not be avoided even if the larvae were reared in an incubator after they had been treated with heat. Therefore, it was necessary to cut open the wheat grains in order to determine the result of the experiment. Dead and living eggs could be easily distinguished because the egg of the Angoumois grain-moth presents a peculiar pink colour before it hatches, and also because the dead egg becomes grayish ochreous.

Results of Experiments.

The results of experiments are given in Tables I and II.

Table I.

Effect of Heat Treatment on the Egg.

Experiment No.	Temp. (°C.)	Duration of Exposure (Minutes)	Number of Eggs Used	Per Cent of Eggs Killed	Remarks
16	60	5	100	100	Temp. variation within ±0.5°C.
17	"	7	100	100	"
9	"	10	100	100	"
18	"	"	100	100	"
4	"	20	50	100	"
10	"	"	100	100	"
12	70	5	100	100	"
13	"	10	100	100	"
14	"	15	100	100	"
Control (i)			100	4	
" (ii)			100	0	

Table II.

Effect of Heat Treatment on the Larva and Pupa.

Experiment No.	Temp. (°C.)	Duration of Exposure (Minutes)	Number of Individuals	Per Cent Killed	Corrected* Percentage of Kill	Remarks**
23	45	180	81	0	0	Temp. variation within ±5.0°C.
24	"	300	62	4.8		
25	"	480	73	28.7	12.9	"
Control			63	18.1		

Table II. (Continued.)

Experiment No.	Temp. (°C.)	Duration of Exposure (Minutes)	Number of Individuals	Per Cent Killed	Corrected* Percentage of Kill	Remarks**
68	50	120	73	27.3	.....	Temp. variation within ±5.0°C.
69	”	240	69	86.9	86.9	”
Control			90	0		
52	55	120	99	91.9	87.8	Temp. variation within ±5.0°C.
53	”	180	125	97.6	96.3	”
50	”	240	58	100	100	”
51	”	420	49	”	”	”
Control (i)			90	34.1		
” (ii)			197			
15	60	40	100	100	.....	In the former half of larval stage. Temper. variation ±1.0°C.
17	”	50	100	96		”
B.10	”	”	109	52.2	37.8	Temp. variation within ±1.0°C.
Control (i)			51	23.1		
” (ii)			78			
18	60	60	100	100	.....	In the middle of larval stage. Temp. variation as in the preceding experiment.
35	”	”	38	98.1	97.2	
44	”	”	70			
Control (i)			64	31.7		
” (ii)			84			
54	60	90	80	97.8	96.9	Temp. variation within ±1.0°C.
55	”	”	87			
45	”	”	64			
Control (i)			84	28.6		
” (ii)			108			
29	65	30	62	93.2	91.5	
33	”	”	57			
19	”	”	100	100	.....	In the middle of larval stage.
Control (i)			81	19.3		
” (ii)			64			

Table II. (Continued.)

Experiment No.	Temp. (°C.)	Duration of Exposure (Minutes)	Number of Individuals	Per Cent Killed	Corrected* Percentage of Kill	Remarks**
20	65	40	80	97.6	96.4	
34	″	″	45			
Control (i)			87	32.4		
″ (ii)			64			
21	65	50	100	86.7	82.8	August 23.
66	″	″	66			October 6.
Control (i)			87	22.3		
″ (ii)			52			
9	70	10	84	63.1	58.1	
26	″	″	88			
30	″	″	97			
Control (i)			92	11.8		
″ (ii)			81			
″ (iii)			64			
27	70	20	58	96.0	95.1	
31	″	″	59			
B. 13	″	″	62			
Control (i)			81	19.3		
″ (ii)			64			
28	70	30	67	100	100	
56	″	″	38			
57	″	″	78			
Control (i)			81	7.7		
″ (ii)			52			
16	75	10	29			In the middle of larval stage. August 12th.
37	″	″	61	83.4	80.0	
40	″	″	66			
Control (i)			76	21.8		
″ (ii)			66			

Table II. (Continued.)

Experiment No.	Temp. (°C.)	Duration of Exposure (Minutes)	Number of Individuals	Per Cent Killed	Corrected* Percentage of Kill	Remarks**
38	75	15	45	90.4	87.7	
42	”	”	80			
Control (i)			76	21.8		
” (ii)			66			
41	75	20	70	93.1	92.5	
62	”	”	62	.....	.....	October 6th.
Control (i)			66	7.6		
” (ii)			52			
48	80	10	70	93.1	90.7	
60	”	”	52			
61	”	”	53			
Control (i)			90	25.3		
” (ii)			52			
49	80	15	51	99.3	99.0	
58	”	”	66			
59	”	”	45			
Control (i)			90	25.3		
” (ii)			52			
64	80	20	44	100	100	
65	”	”	76			
Control			52	5.7		

* Correction was made according to the formula $\frac{(x-y)\,100}{100-y}$, where x represents the percentage of insects killed which was not corrected and y the percentage of dead individuals in the control.

** When no specification as to the stage of larva is given, the experiment was carried out in the last larval stage.

Discussion.

Grossman concludes from the results of his experiments that the adult of the Angoumois grain-moth was less resistant to high temperature than the larva or pupa and that all the adults were killed by exposure for 30 minutes at 49°C. The present writers have not yet studied the effect of high temperature on the adult, but the adult seems to be rather susceptible to high temperatures judging from experience in rearing this insect.

According to the data in Table I, 100 per cent of eggs were killed by exposure for 5 minutes to 60°C. The writers have not conducted any experiment at temperatures lower than 60°C. so that it was not known whether the egg is killed by a lower temperature than 60°C. In the writers' experiments eggs were heated together with about 15 cc. of wheat so that a certain period of time must have been necessary in order that every wheat grain reached 60°C. The fact that 100 per cent were killed by exposure for 5 minutes to a temperature of 60°C. indicates that the resistance of the egg to high temperatures is rather small.

The results of the experiments conducted to show the effect of high temperatures on the larva and pupa are shown in Table II. The results may be briefly summarized as follows: Exposure for 8 hours at 45°C. killed only 12.9 per cent of the larvae and pupae. Exposure for 2 hours at 50°C. killed 27.3 per cent. When the time of heating was increased to 4 hours, 86.9 per cent were killed. At 55°C., 87.8 per cent were killed by 2 hours' exposure, and 96.3 per cent were killed by 3 hours' exposure. When the duration of exposure was increased to 4 hours, 100 per cent were killed. At 60°C., all of the young larvae and those which were in the middle of the larval stage were killed by an exposure of 40 minutes, but 97.2 per cent were killed by exposure for one hour to a temperature of 60°C. in the case of nearly fullgrown larvae. Even when the time of exposure was increased to 1.5 hours, the percentage of insects killed did not increase. At 65°C., exposure for 40 minutes killed 96.4 per cent while exposure for 50 minutes did not seem to produce a higher percentage of kill. Exposure for 20 minutes to 70°C. killed 95.1 per cent and exposure for 30 minutes to this temperature killed 100 per cent. At 80°C., exposure for 15 minutes killed 99 per cent, while 20 minutes' exposure to this temperature killed 100 per cent.

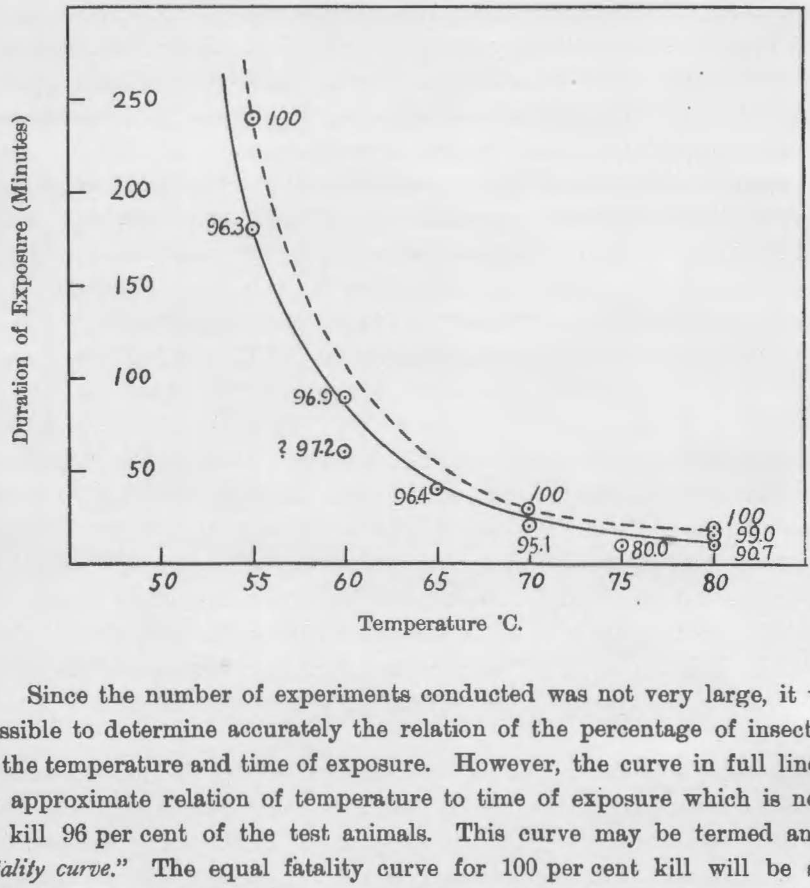
An examination of the data shows that the results of experiments which were carried out two or three times under the same conditions did not always agree closely. There must have been various causes for such apparently inconsistent results. The following seem to be important causes for the fluctuation of the percentage of insects killed: Although precaution was taken to use for experiment test animals which were considered to be in the last larval stage or in the pupal stage judging from the fact that two or three moths had begun to emerge, they were not by any means in the same stage of development. Besides, the number of larvae that were completely fullgrown was not the same in different cultures, varying from approximately 40 per cent to 80 or 90 per cent of the individuals which had hatched out of 100 or 200 eggs. Thus, the number of individuals which were used varied considerably in different experiments. The younger larvae were less resistant to high temperatures as is evident from the results of *Experiment Nos. 15, 17, 18 and 19* recorded in Table II, in which larvae in the middle of the larval stage or in still younger stages were used. The time of year when experiments were conducted seemed also to have slight influence on the rate of insects killed. When experiments were carried out at the end of September or in October, the percentage of kill was slightly lower than in the earlier experiments. Thus, in *Experiment No. 21*, which was carried out on August 23rd, 100 per cent were killed by exposure for 50 minutes to 65°C. while in *Experiment No. 66* which was carried out on October 6th approximately 66.6 per cent were killed by the same treatment. There were other such instances besides the two experiments just cited. These circumstances seem to have caused the percentage of insects killed to vary in different experiments even when temperature and duration of exposure were the same.

The results of the writers' experiments varied slightly as has been pointed out above, but the variation was comparatively small when only such experiments in which larvae in their last stage of development were used are considered. Therefore, we may be justified in summarizing the results as described above, according to the average value obtained from two or three experiments.

Dean reported that heating for several hours at 51.7°C. killed all insect pests in mill and Grossman stated that the larvae and pupae of the Angoumois grain-moth were killed by exposure for one hour to 50°C. In the writers' experiments, exposure for 2 hours to 50°C. killed only 27.3 per cent and when the duration of exposure was doubled, 86.9 per cent were killed. In order to kill 100 per cent of larvae and pupae, it was necessary to expose for more than 3 or 4 hours to a temperature of 55°C. In the writers' experiments conducted at 50° and 55°C., the temperature did not remain constant throughout the course of an experiment, but it varied approximately from 5°C. below, to 5°C. above the temperature which was required. This fluctuation of temperature might have been one of the causes that made the time of exposure in the writers' experiments longer than in Dean's or in Grossman's.

In the other experiments which were carried out by the present writers under temperatures higher than 55°C., the fluctuation was always less than $\pm$1.0°C., being approximately $\pm$0.3°C. in most cases. In these experiments, too, the time of exposure which was necessary to kill 100 per cent was much longer in the writers' experiments than in Dean's or in Grossman's. That longer exposure was required in the writers' experiments seems to be accounted for chiefly by the difference in the method of experiment. As has been already stated, in the writers' experiments, larvae or pupae which were in the wheat grains were heated together with about 15 cc. of uninfested grains, while in Dean's or in Grossman's experiments test animals were taken out of the grains in which they were found and were exposed to high temperature. No doubt, it took a little time to heat 15 cc. of wheat up to the temperature required, although the writers did not determine the time which was necessary. This must have been the chief cause which made the time of exposure longer in the present writers' experiments. In regard to this point, the writers hope to carry out further experiments.

The curves* in the accompanying figure were constructed according to the data in Table II taking the time of exposure on ordinates and temperature on abscissae. Their point of intersection corresponds to the percentage of insects killed which is obtained by exposure of the duration and temperature represented by the ordinate and abscissa, respectively.



Since the number of experiments conducted was not very large, it was not possible to determine accurately the relation of the percentage of insects killed to the temperature and time of exposure. However, the curve in full line shows an approximate relation of temperature to time of exposure which is necessary to kill 96 per cent of the test animals. This curve may be termed an "*equal fatality curve.*" The equal fatality curve for 100 per cent kill will be situated slightly above the 96% curve and almost parallel to the latter. The curve shown in broken line was drawn in such a manner as to pass through three points which correspond to 100 per cent kill. This curve shows an approximate relation of temperature to the time of exposure which is required to kill 100 per cent. In order to obtain an equal fatality curve which shows the relation of time and temperature more accurately, further experiments are necessary.

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* The curves in this figure are essentially the same as those which the senior writer published in *Nôgaku-Kenkyû* Vol. 22, p. 284. The only differences are that the scale of the ordinate was somewhat modified and also that the curves in this figure are based upon the percentage of kill which was corrected by taking into consideration the percentage of dead individuals in control while in the earlier paper the curves were constructed using uncorrected data.

Summarizing what has been stated above, the following conclusions may be drawn: Larvae and pupae of the Angoumois grain-moth can be almost completely killed by exposing them for one hour to a constant temperature of 60°C., but as the equal fatality curve indicates, much longer exposure is required when the temperature is 50°C. or lower. Whether it is practicable to utilize such high temperatures as 60° or 70°C. for the purpose of controlling the Angoumois grain-moth is another important problem and should be investigated from various points of view. Admitting that heat treatment is practicable, an important question is whether, in treating a large quantity of wheat, it is more convenient to expose for a long period of time to a comparatively lower temperature or to expose for a short duration to a higher temperature. At present the writers consider that it would seem more convenient to use a short period of exposure under a fairly high temperature.

Effect of Heat on the Germination of Wheat and Barley.

If heat treatment is effective and practicable as a method of controlling the Angoumois grain-moth, it is necessary to study the effect of high temperature on the germination of wheat and barley before proceeding to apply heat treatment for the purpose of killing the Angoumois grain-moth.

According to Kondō, wheat seeds are highly resistant to high temperature. For example, if wheat seeds are fully mature, the germination is not affected by exposing them to 70°C. for a week or by drying them at 80°C. until their weight becomes constant. The drier the seeds, the more resistant they are to high temperature.[6)]

In order to learn the effect of such high temperatures as the writers used for the purpose of killing the Angoumois grain-moth on the germination of wheat and barley, the writers conducted some germination tests. The moisture of the wheat and barley which were used for heat treatment was not determined, but it is believed that they were fairly dry since these seeds were preserved in a dry state after having been dried well when they were harvested.

The method of exposing wheat and barley to high temperature was quite the same as in the experiments which were carried out to kill the Angoumois grain-moth. If a germination test of wheat and barley is made before the *after-ripening* is completed, the percentage of germinating seeds is usually very low. For example, when germination tests were made at the beginning of July using *Hatakeda* (a variety of wheat) and *Chinko-hadaka* (a variety of barley) the percentage of germinating seeds was very low. It seemed that the percentage was lower in the control (i. e., seeds that were not exposed to high temperature) than in the treated seeds. Therefore, the second germination test was conducted from the end of September to the earlier part of October. The variety of barley which was used for the test was *Chinko-hadaka* and that of wheat, *Shirodaruma*. The results of the germination tests are shown in Table III.

Table III.

Effect of High Temperature on the Germination of Wheat and Barley.

Temperature (°C.)	Duration of Exposure (Minutes)	Percentage of Seeds that germinated	
		Barley	Wheat
80	10	97.3	81.6
"	15	96.3	88.3
"	20	95.0	87.0
75	10	97.3	87.6
"	20	98.3	88.3
"	30	97.6	90.6
70	10	98.0	91.0
"	20	98.3	88.3
"	30	99.0	90.0
65	30	99.0	87.3
"	40	96.0	92.0
"	60	98.3	88.6
60	60	93.6	93.0
"	90	95.6	88.3
55	240	96.0	88.6
"	360	98.3	88.3
"	480	96.6	86.3
Control	Not exposed to high temperature	98.1	87.6

According to the data in Table III, the percentage of seeds that germinated was always quite high, especially in barley, so far as exposure remained between 8 hours at 55°C. and 20 minutes at 80°C. When the percentage of the treated seeds which germinated is compared with that of the control, the difference is very small. Besides, the percentage of germinating seeds has no definite relation to the temperature to which the seeds were exposed or to the duration of exposure. Therefore, it can not be concluded that the heat treatment affected the germination of treated seeds. When these results are compared with what KONDŌ states as to the resistance of wheat seed to heat, it may be concluded that heat treatment of from 8 hours' exposure to 55°C. to 20 minutes' exposure to 80°C. does not affect the germination of wheat and barley.

Summary and Conclusion.

No experiment has yet been conducted in Japan as to whether the Angoumois grain-moth in all the stages of its development can be killed by heat. The writers, therefore, carried on some experiments to study the resistance of the egg, larva and pupa of this insect to high temperatures. According to the results of the experiments, the egg is highly susceptible to high temperatures and can be killed by exposure for 5 minutes to 60°C. The larva and pupa are more resistant than the egg and 100 per cent can not be killed unless they are exposed to 60°C. for 1 to 1.5 hours. As the temperature is raised, the duration of exposure which is required becomes gradually shorter, and at 80°C., only 15 to 20 minutes' exposure is required to kill almost 100 per cent.

In the writers' experiments, larvae and pupae which were present in wheat grains were subjected to heat treatment together with approximately 15 cc. of healthy wheat. In these conditions, heating for a certain period of time seemed necessary in order that heat might penetrate to every portion of the material. Consequently, even at such high temperatures as 70° or 80°C., 100 per cent could not be killed when the duration of heating was 10 minutes or less.

In order to kill 100 per cent of the larvae and pupae, a longer exposure and a higher temperature seem to be necessary than those which the previous investigators considered to be sufficient. When heat treatment of stored grain is undertaken on a practical scale, satisfactory results would not be obtained unless the grain is subjected to treatment in which the temperature and time of exposure are at least nearly the same as those which the writers found to be sufficient for killing 100 per cent of the test animals.

Germination tests carried out with wheat and barley which were subjected to heat treatment indicate that no ill effect will be produced if the seeds are well mature and dry, so long as the temperature and the time of exposure remain within the range used by the writers.

The results of the present experiments show simply how resistant the Angoumois grain-moth is to high temperature. Whether heat treatment can be successfully used as a practical means of controlling the Angoumois grain-moth remains to be determined by further experiments. However, it would not be impossible to control this insect by heat if improvement is made in the method of artificial drying of grain and also in the method of storage.

Literature Cited.

1) DEAN, G. A., Heat as a means of controlling mill insects. Jour. Econ. Ent. Vol. 4: 142—158, 1911.

2) Ditto. Further data on heat as a means of controlling mill insects. Jour. Econ. Ent. Vol. 6: 40—53, 1913.

3) DENDY, A. and ELKINGTON, H. D., Temperature limits for the life of insects. Reports of the grain pests (war) committee, No. 7 : 39—45, 1920.

4) BACK, E. A. and COTTON, R. T., Stored grain pests. U. S Department of Agriculture, Farmers' Bull. No. 1260 : 46, 1922 (Revised edition, 1925).

5) GROSSMAN, E. F., Heat treatment for controlling the insect pests of stored corn. Florida Agric. Exper. Sta. Bull. No. 239, 1931.

6) KONDŌ, M. (近藤萬太郎), 日本農林種子學. 前編. 第 128 頁, 昭和 8 年 (1933).