An evaluation of the effectiveness of various methods and tests to measure quantitative and qualitative losses in maize due to insect infestation. Changes occurring in maize infested with the maize weevil under constant conditions of 80°F and 65±5% relative humidity were observed over 36 weeks of storage. Dry matter weight loss in infested maize was 3.7% at the end of 9 months of storage and 0.7% in non-infested maize. Numbers of insects from probe samples of 200-pound lots of maize were not a reliable indication of damage when compared to X-ray examination of samples. Non-infested lots of maize did not pick up moisture from the atmosphere under the constant environmental conditions. Large numbers of insects in infested lots of maize resulted in moisture increases; however, considerable damage occurred before moisture changes were detected. No temperature changes due to insects in the infested maize were detected. Seed germination in infested samples decreased, but whether the decrease was caused by insects or by molds was not determined. Chemical tests of maize quality did not show maize damage. Fat acidity values remained below the maximum for sound maize and glutamic acid decarboxylase activity decreased (a sign of deterioration) in sound maize. Proximate analyses (percent fiber, ash, protein, and fat) remained unchanged despite observed damage and measured loss in quantity. Research in this area is continuing.
MEASUREMENT OF MAIZE WEEVIL AND FUNGI DAMAGE TO STORED CORN

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REPORT SUMMARY

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SUMMARY STATEMENT

Changes that occurred in maize infested with the maize weevil under constant conditions of 80°F and 65±5% relative humidity were observed over 36 weeks storage. Dry matter weight loss in infested maize was 3.7% at the end of 9 months storage, and 0.7% in non-infested maize.

A major objective was to evaluate the effectiveness of various methods and tests to measure quantitative and qualitative losses in maize due to insect infestation.

Numbers of insects from probe samples of 200 pound lots of maize were not a reliable indication of damage when compared to X-ray examination of samples.

Non-infested lots of maize did not pick up moisture from the atmosphere under the constant environmental conditions. Large numbers of insects in infested lots of maize resulted in moisture increases, however, considerable damage occurred before moisture changes were detected.

No temperature changes due to insects in the infested maize were detected. Seed germination in infested samples decreased, however, whether the decrease was caused by insects or by molds was not determined.

Chemical tests of maize quality did not show maize damage. Fat acidity values remained below the maximum for sound maize and glutamic acid decarboxylase activity decreased (a sign of deterioration) in sound maize.

Proximate analyses (percent fiber, ash, protein and fat) remained unchanged despite observed damage and measured loss in quantity.

Research in this general area is continuing.
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This report has been prepared from a thesis research project carried out by Mr. Miguel A. Mora while attending Kansas State University on a University of Costa Rica scholarship. The research was supported in part under contract AID/ta-C-1162.

The research reported here is preliminary work toward developing methods for assessing damage and losses to grain in storage. Basic information developed in this study will be used to formulate plans for conducting grain loss studies in developing countries, especially at the farm level.
INTRODUCTION

Since grain production is seasonal but consumption is continuous, grain produced must be stored for variable lengths of time before used. During storage, grain is subjected to qualitative and quantitative losses due to several agents including insects, fungi, rodents, and mites.

Quantitative grain storage losses have been estimated to be from 3 to 50% (39). The figures often are not well documented but may be high in specific situations (24, 34). Estimates of annual weight loss caused by insects in all food stuff generally vary from 5 to 10% (50).

Assessment of quality loss is difficult because of the various definitions of quality (39); the concept of quality changes with specific situations. Methods for quality loss estimation are in need of improvement.

This study was to determine quantitative and qualitative losses caused by maize weevils to corn stored under controlled conditions, and to test the effectiveness of several methods to measure insect and fungi damage.
There are two main types of losses: quantitative and qualitative. Quantitative loss occurs when part of the grain actually disappears. Qualitative loss refers to lowering of commercial or nutritional quality of grain (19, 39).

**Quantitative**

Loss in weight is one of the more direct damages caused by pests. Even though insects are small, under certain conditions, large populations can cause considerable weight loss (50, 53). Production of 35,000 weevils may result in 1 kg loss of wheat (30).

Real quantity of material eaten or destroyed may be obscured by changes in moisture content (28, 39). Howe (27) gave an example in which infested peanuts lost 5% in dry matter and dust weight but in commercial channels the estimated weight loss was only 0.1% because changes in moisture and dust were not considered.

**Qualitative**

Insects and fungi may reduce germination of stored grain in several ways: their activities can increase temperatures to levels which kill the germ (18), by direct consumption of germ by insects (9), by consumption of seed's reserves (21), and by fungal invasion of the embryo (18, 42). Loss of germination is especially important in storage of grain for seed purposes and in the case of barley for malting (15). Quality of grain for food or feed use is usually not materially affected.
by reduced germination, however, subsequent storability of the grain may be affected (14).

Insects and fungi lower commercial quality of grain in several ways (52); by presence of live and dead insects, and insect damaged kernels; discoloration, black germs, and musty odors caused by fungi; heat damage and damage caused by high temperatures generated by insects and/or fungi; lower density; presence of mycotoxins.

Heavy insect infestation may lower protein content especially when the damage is concentrated in the germ (33). However, an apparent protein increase may be found when only starchy content of grain is consumed or by contamination with nitrogenous compounds, as urine and saliva of insects (33).

DETERIORATIVE AGENTS

Insects

Insects are a major problem in sanitation and quality control of stored grain (22). They not only consume or contaminate large quantities of grain but also can produce conditions favorable for development of fungi by increasing the temperature and moisture of the grain. They also disseminate fungal spores and bacteria (18, 22, 31, 49).

Among stored-product insects, the weevils (Sitophilus spp.), lesser grain borer (Rhyzopertha dominica (FAB.)), and angoumois grain moth (Sitotroga cerealella (OLIV.)) are most damaging. These insects develop inside grain kernels (22). True weevils are widely distributed storage insects and include the granary weevil (Sitophilus granarius (L.)),
rice weevil, *S. oryzae* (L.), and maize weevil, *S. zeamaize* (MOTSCH.). **(22)**

Extent of insect damage to stored grain is directly related to populations which in turn are influenced by temperature, moisture, and food supply. **(15, 21)**.

Moisture content requirements vary for different insects. Weevils reproduce rapidly at 13% moisture content and higher **(22)**. Others, such as the lesser grain borer can develop populations at lower moisture contents.

Insects infesting grain lay very few eggs at temperatures below 15.6°C but at 21°C or higher can reproduce at high rates **(15, 22)**. Most storage insects stop reproducing at 35°C and at this temperature adults are short lived **(22)**.

Insect infestation is usually accompanied or followed by fungal invasion **(1, 16, 40)**. As part of their metabolic processes, insects break down food into water and carbon dioxide along with the release of energy. Moisture produced plus moisture migration due to heat liberated by insects can result in creation of environments favorable for mold development **(15, 22)**.

**Fungi**

Fungi are now considered a major cause of grain deterioration **(18)**. Fungi found on grain have been classified as field and storage fungi primarily based on moisture requirements **(19)**.

Field fungi invade grain before harvest and require moisture content in equilibrium with 90% r.h. or higher **(17, 18, 45)**. In cereals such as wheat, corn, rice, barley and sorghum, 90% r.h. results in an equilibrium moisture content (EMC) of about 20-25% **(41, 42)**.
Damage caused by field fungi has usually occurred by harvest time before moisture content is below 20-22% (18). Field fungi gradually die if grain is stored at moisture content in equilibrium with 70% r.h. or lower but in very dry grain they survive for extended periods of time.

There are two major genera of storage fungi: Aspergillus and Penicillium (17, 18, 45).

Minimum relative humidity for growth of storage fungi is 65-68% (18, 20, 41, 42) which produces an EMC of approximately 13% in wheat, corn and sorghum (18, 19). Relative humidity is considered more important as an indicator of water availability for microorganisms than moisture content (6, 32).

Optimum temperature for storage fungi varies with species but is usually about 30 to 32°C (18).

METHODS TO MEASURE STORAGE DAMAGE

In assessing quality of stored grain, information about insect infestation and fungal invasion helps to indicate potential deterioration.

Methods of detecting internal insect infestation

One of the problems in reporting the presence of insects and insect-damaged kernels is that internal infestation is not readily detected by visual examination.

Several techniques are used to detect internal insect infestations, and include: egg plug staining, flotation methods, cracking flotation methods, X-ray examination, ninhydrin reaction and others (4, 21, 23, 38).

X-ray technique is one of the most reliable methods but expensive (26).
Temperature and Activity of insects and molds can cause a considerable rise in temperature in grain (22). When moisture content is 15% or less, insects can cause "dry-grain heating" in which temperature may increase up to 42-43°C. At moisture content higher than 15%, microorganisms grow very well and may cause "wet-grain heating" with temperature increases up to 62-63°C (22). For this reason, any abnormally high temperature could indicate the presence of insects and/or fungi.

Fungal invasion

Abundance and kind of fungi may indicate: 1) conditions under which grain has been stored, 2) possible damage it may have suffered, and 3) potential storability (18).

Fat acidity value (FAV)

One of the best chemical indexes of grain deterioration by fungi is FAV (14). Fungal degradation of fats and oils produces fatty acids as intermediate products. The amount of fatty acids can be expressed as FAV and used as a measure of deterioration and storability of grain (18, 55).

Germination

Viability of grain may be reduced by insects and fungi. Percent germination can be used as an index of damage caused by these deteriorative agents (18, 29, 33, 44).
Glutamic acid decarboxylase activity (GADA)

Increase in storage time produces a gradual decrease in GADA values (37). This analysis is considered a good index of storage deterioration (36, 37). Linko (35) developed a rapid manometric method for GADA determination, using Sandstädt and Blish pressuremeters (46).

GADA results have shown significant high correlation with other tests such as germination and FAV (11, 12, 36, 37).
MATERIALS AND METHODS

GRAIN

Approximately 600 lb (272 kg) of yellow dent corn were obtained from the U.S. Grain Marketing Research Center, U.S. Department of Agriculture, Manhattan, Kansas. Corn was kept at -10°F (-23.3°C) for one week and later fumigated with Phostoxin® to eliminate any possible existing insect infestation.

STORAGE CONTAINERS

Three lots of corn of about 200 lb (90.7 kg) were placed in each of three 32-gallon plastic (garbage-type) drums (Fig. 1).

Lids of drums were provided with 5 holes of 4.5 cm diam., one in the center and 4 at cardinal points about 7 cm on center from the edge. Two larger holes, 12 cm diam., were placed one on each side of the lid handle. Larger holes were covered with 80-mesh brass gauze and filter paper to avoid outside infestation. The smaller holes were used for sampling and were closed with rubber stoppers. The larger holes were to allow air interchange between the corn in the drums and the outside atmosphere.

STORAGE ENVIRONMENT

Before infesting, the corn was conditioned 3 weeks in the room used during the experiment, at 65 ± 5% r.h. and 80 ± 1°F (26.7°C).

INSECTS

Two of the 3 drums were each infested with 400 adult maize
weevils, (Sitophilus zeamaize MOTSCH.) from the ASU Department of Entomology cultures.

Insects were of varying age and their sex was not determined.

The third drum was kept as a control.

**SAMPLING**

Samples were taken at the beginning of the experiment and at 4-week intervals over a period of 36 weeks.

A 100 x 3.5 cm sampling probe (Fig. 1a.) was used to take 2 samples from each sampling hole in the lid. Sections of the probe were separated with rubber stoppers to allow separation of each sample into 3 parts representing top, center, and bottom levels of the corn contained in each drum. After counting insects, samples were composited by level for each drum.

**ANALYSES**

**Temperature**

Four thermisters (48) were attached at 20-cm intervals to a metallic bag probe to form a temperature probe. One thermister was at the tip of the probe (Fig. 1b). At each sampling period the temperature probe was introduced into the corn through each sampling hole and temperatures read using a YSI, Model 42SL Tele-thermometer.

**Gross weight**

Before and after each sampling, the drums with corn were weighed using a Fairbanks, 500 lb capacity, platform scale. Accuracy of the
Fig. 1a  Upper: Drums, sampling probe, and platform scale

b  Lower: Temperature probe and telethermometer.
scale is estimated to be ± 1 lb.

Insect count

Live and dead adult weevils and moths from each probe were counted by levels within drums.

Moisture content

Samples were exposed to the laboratory atmosphere for a minimum time while insect counts were made and then placed immediately in tightly closed quart jars. Samples were then placed in plastic bags, mixed, weighed and small samples for moisture content determination placed in baby food jars. Moisture tests were made by drying 10 to 15 g of whole corn kernels in an air-oven at 103°C for 72 hours (3). Moisture percentages were calculated on a wet basis.

Dust

After removing moisture samples, the remaining corn was shaken on a No. 10, U.S.A. Standard Testing Sieve (openings 2 mm) and the material passing through expressed as grams of dust/kg.

Density

After dust was removed, density or weight per unit volume was determined. A funnel on a standard test weight apparatus was used to evenly fill a 300-ml container. After filling, excess corn was scraped from the top of the container using a standard procedure (51). Corn remaining in the container was weighed and reported as pounds/bushel.
X-ray, mold count and germination analyses were made from a common 100-g sample. A sample of approximately 125 g was separated using a Boerner grain divider and 100 g weighed and radiographed using a General Electric Grain Inspection Unit.

Radiographs were examined using an X-ray viewer and hand lens. Results were reported as insect damaged kernels/100 g.

**Germination**

Percentage of viable seeds was determined by preparing two 50-kernel replicates on wet paper towels wrapped with aluminum foil to retain moisture. The 100 kernels were obtained from the 100-g X-ray sample.

Percent germinated seeds was determined after one week at 20°C. Seeds were considered germinated if a sound root or coleoptile was produced.

**Mold counts**

Kinds and numbers of fungi invading corn samples were determined in 50 seeds. Corn was surface disinfected in 2% NaOCl for 1 minute and rinsed with sterile distilled water prior to plating them on MS4T culture medium (malt agar with 4% NaCl and 200 ppm Tergitol/1).

Plates were incubated at 25°C for one week or until fungi grew enough to be identified. Results are given in percentage of seeds invaded by each kind of fungus.

**Fat Acidity Value (FAV)**

Since tests for FAV require 10% m.c. or lower (9) corn left...
after sampling for X-ray was dried overnight at 32°C. Corn was ground
on a Thomas-Wiley Laboratory Mill, Model 4 with 1-mm sieve just before
running FAV and glutamic acid decarboxylase activity tests.

FAV's were determined in 2 ways: by titration of a benzene
extract with 0.0401 N KOH solution (1 ml 0.0356 N KOH = 2 mg KOH) (3)
and colorimetrically (3, 8).

Fatty acids from 50 g of freshly ground corn were extracted
with 50 ml of benzene by shaking for 30 minutes in a mechanical device.
The extract was filtered using suction and 15 ml of filtrate titrated
with KOH. Ten ml of the remaining filtrate were placed in a test tube
with 2 ml of cupric acetate solution, mixed, filtered, and transmittance
at 640 μm read using a Bausch & Lomb Spectronic 20. Transmittance
percent was converted to FAV by comparison with a standard curve
prepared using oleic acid (3).

Results of both methods were expressed as mg of KOH required to
neutralized free fatty acids from 100 g of dry grain.

Glutamic Acid Decarboxylase Activity (GADA)

Thirty-gram samples of freshly ground corn were placed in
Sandstedt and Blish pressuremeters (46) and 15 ml of 0.1 M glutamic
acid solution in 0.067 M phosphate buffer, added. Pressuremeters with
samples were placed in a water bath at 30°C. After an equilibration
period of 10 minutes, systems were closed and pressure produced by
carbon dioxide liberation during 30 minutes recorded. That figure
plus 100 is the reported value for GADA.

Ethyl lactate colored with crystal violet was used as manometric
fluid (35).
Proximate analyses

Proximate analyses (moisture content, crude protein, crude fat, fiber, fats, and ash) were done in the Department of Grain Science and Industry Analytical Service Laboratory according to standard methods.
RESULTS AND DISCUSSION

Data are presented graphically in the text (Figs. 2-12) and in tabular form in the Appendix (Tables 1-8).

Insect counts (Fig. 2, table 1)

No insects were expected or found in the control drum during the 36-week sampling period.

Infested drums were expected to give results similar to one another, however, a secondary infestation of Angoumois grain moth (Sitotroga cerealella (O.)) in drum II may have affected the results.

Very few insects were recovered by probe sampling before 20 weeks of storage. At this point, numbers of insects began to increase, especially in the bottom portion of infested drums. At 24 weeks there were about 60 insects/kg in the bottom samples of each infested drum. No further increase appeared in drum I but insect counts in the bottom of drum II continued to increase until infestation reached 130 insects/kg at 36 weeks. No moths were found in bottom samples and only a few in the top and center samples of drum II.

Total numbers of insects in top and center samples were also higher in drum II than drum I. At 36 weeks, insect counts were 8 and 18/kg in top and center of drum I and 28 and 45 insects/kg in top and center of drum II, respectively.

Pingale (39), in a review of literature, mentioned that Sitophilus spp. need at least 12% m.c. for reproduction and that rate of oviposition increases as relative humidity increases from 70%. The optimum range for development was found to be 13.5 to 17.6% m.c. with 14.75% m.c. as optimum.
Number of insects in corn infested with *Sitophilus zeamaize* stored at 68% r.h. and 80°F (26.7°C) during 36 weeks.
Drums were maintained in a room at 80°F (26.7°C), a temperature appropriate for insect development and optimum for weevils (22). It is assumed that the relatively low starting moisture content (12.3 to 12.6%) and natural low initial rate of population increase were, in part, responsible for the small numbers of insects recorded in the first 16 weeks of sampling. After 16 weeks, moisture content began to increase but never exceeded 14.8% during the experiment.

Even though initial infestation was about 5 insects/kg (400 weevils in 90.7 kg), lower concentrations were found in the samples up to 8 and 12 weeks storage. This indicated that probe sampling may fail to give representative sampling for insects.

**X-ray (Fig. 3, table 2)**

X-ray results do not coincide with insect counts. Although insect counts (Fig. 2) did not increase much before 16 weeks, X rays showed that weevils were doing considerable damage at 8 and 12 weeks.

Number of insect-damaged kernels, at 36 weeks, detected by X ray, tend to be similar (61 to 70 damaged kernels/100 g) in infested drums at all levels. As mentioned, numbers of insects at 36 weeks varied with infested drums and levels.

Number of insects counted per kilogram did not increase much in drum I after 24 weeks but X-ray results showed a steady increase in number of damaged kernels during the entire experiment.

In drum II, insect counts at 36 weeks were greater at the bottom (130/kg) than at the top (28/kg) and center (45/kg). X-ray results showed approximately the same degree of damage in the 3 levels at 36 weeks.
Fig. 3. X-ray results of corn infested with *Sitophilus zeamaize* stored at 68% r.h. and 80°F (26.7°C) during 36 weeks.
Results indicated that insect counts are not good indicators of the actual damage done in grain. X-ray results are better.

**Moisture content (Fig. 4, table 3)**

Moisture content of control samples remained essentially unchanged except for an increase of 0.4% in the top sample after 36 weeks. This was probably due to moisture absorption from the atmosphere.

The moisture increase of less than 0.5% after 36 weeks indicated that if shelled corn is reasonably dry prior to storage, moisture absorption from the atmosphere is not likely to be great under constant temperature conditions. Day/night temperature changes in most storage situations might induce air circulation within the drums which could alter this condition.

Moisture in infested drums began to increase after 16 weeks. Bottom samples in drums I and II had increased by 2.4 and 2.0% m.c., respectively, at 36 weeks. Top and center samples of infested drums increased by 1.2 and 1.4% m.c. in drum I and 1.1 and 1.3% in drum II, respectively.

Increase in moisture content was expected (1, 2, 16) due to water release during metabolic activities of insects. Moisture content increased more in bottom samples where insect and mold counts were higher. Since "hot spots" did not develop, measurable moisture migration was not expected to occur.

Increased moisture content might be used to detect heavy insect infestation, however, under actual conditions, grain is generally exposed to variable relative humidities and temperatures both of which can cause changes in moisture content.
Fig. 4. Moisture content of corn infested with Sitophilus zeamaize stored at 68% r.h. and 80°F (26.7°C) during 36 weeks.
Temperature

No appreciable changes in grain temperature (from $80^\circ F$) were recorded during 36 weeks' observation. The insect populations probably were not large enough to produce heat that could not dissipate from the bulk of grain.

With the quantity of grain and level of infestation reported here it was not possible to detect insect infestation based on changes of temperature even though considerable damage and loss was experienced.

Dust (Fig. 5, table 4)

After 12 weeks' storage and at 4-week intervals, material passing through No. 10 U.S.A. Standard Testing Sieve was weighed from composite samples and reported as "dust". Most of this fine material was found in bottom portions of infested drums and increased with time of storage and insect population. Maximum dust found was 3.3 and 4.1 g/kg in bottoms of drums I and II, respectively. Maximum dust in top and center of drum I was 0.8 g/kg and in drum II, 1.2 and 1.3 g/kg for top and center samples, respectively.

No measurable amounts of dust were obtained from control samples.

Dust in the bottom of infested drums seems to have created a better environment for insect development since it was here that the majority of insects were found. Bronswijk and Sinha (14) also found more dust in bottom portions of storage containers, but in their study dust concentration became so high that insects migrated from the bottom to top and lateral sections of containers.

Dust could be used as an indication of insect presence but in order to be meaningful, representative samples would have to be taken.
Fig. 5. Dust weight in corn infested with *Sitophilus zeamais*, stored at 68% r.h. and 80°F (26.7°C) during 36 weeks.
all the way to the bottom of the container. It is difficult to introduce the sampling probe in grain with large quantities of dust. In practical situations, this may lead to non-representative samples obtained from only the upper portions of containers.

Presence of dust in the sampling probe was one of the first signs of insect activity noticed.

Density (Fig. 6, table 5)

Weight per unit volume results were quite variable, probably due to the relatively small samples (about 225g) used to determine "test weight" (pounds/bushel). In general, however, density of control samples remained unchanged and density of samples from infested drums decreased with increased insect damage.

Average decrease in density of infested samples, at 36 weeks, was 2.25 lb/bu (range 2.0 to 2.5 lb/bu).

Calculated loss of weight based on density, was null in the control, 3.7% in drum I and 3.4% in drum II at 36 weeks. These results approximate weight losses found by direct weighing of drums (Fig. 12).

Density determined on successive samples from a given lot of corn, could be used to measure grain loss if: 1) samples are large enough to give a good estimate of density, 2) moisture changes are compensated and 3) insect infestation is heavy.

Germination (Fig. 7, table 6)

Seed viability of control samples remained almost unchanged for 36 weeks. Viability of infested samples began to decrease after 16 weeks. Germination of bottom samples decreased from over 90% to
Fig. 6. Density (test weight) of corn infested with Sitophilus zeamaize stored at 68% r.h. and 80°F (26.7°C) during 36 weeks.
Fig. 7. Germination of corn infested with Sitophilus zeamaize stored at 68% r.h. and 80°F (26.7°C) during 36 weeks.
5 and 54% in drums I and II, respectively. Top and center samples of infested drums decreased from over 90 to 80% germination.

Germination results followed more closely changes in insect counts (Fig. 2) than actual damage revealed by X-ray (Fig. 3). Viability also seems to follow changes in Aspergillus glaucus counts (Fig. 10) but not counts of other fungi.

L-Lysine acid decarboxylase activity (GADA) (Fig. 8, table 7)

GADA values remained constant at about 260-265 during 20 weeks in the control, 16 weeks in drum I and 12 weeks in drum II. After 20 weeks, GADA decreased slowly in all levels of the control to an average of 221 at 36 weeks. In drum I GADA values decreased to an average of 178 at 36 weeks with only a slightly lower value in the bottom sample. GADA decrease in drum II was similar to that in drum I except that the bottom sample at 36 weeks was considerably lower (117 mm) than top (185 mm) or center (183 mm) samples.

The GADA test was not a good test to detect insect damage, especially with low levels of infestation.

Fat acidity value (FAV)

With methods used to determine FAV, no values higher than 22 were found. FAV below 22 is considered normal for sound corn (9).

FAV results do not correlate well with insect damage (10) unless this damage is very high and accompanied by high mold counts (34). Aspergillus glaucus counts (Fig. 10) were high in some 36-week samples, however, A. glaucus invades grain at relatively low moisture content (ca 14%) and does not have a good correlation with FAV (13).
Fig. 8. Glutamic acid decarboxylase activity (GADA) of corn infested with Sitophilus zeamaize stored at 68% r.h. and 80°F (26.7°C) during 36 weeks.
FAV should not be considered a good indicator to determine insect damage in grain stored under the test conditions.

**Proximate analyses (Table 8)**

No differences in proximate analysis results were found with time, between drums or between levels of drums.

Average chemical composition of corn for the length of the experiment was: crude protein 10.3%, crude fiber 2.2%, crude fat 3.6%, and ash 1.5%.

*Sitophilus* spp. feed almost exclusively in the endosperm and only slight changes in the relative composition of corn might be expected with infestation levels reported here. Numbers of damaged kernels were probably not large enough to influence proportional composition of samples. Even though relative composition of corn did not change, quantitative losses were produced by insects (Fig. 12).

Proximate analysis is not a good method to reveal insect infestation or grain deterioration under the test conditions.

**Mold counts (Figs. 9, 10, and 11)**

Most abundant field fungi were *Cephalosporium* and *Fusarium* spp. At the beginning of the experiment 58 to 72% of the kernels were invaded by field fungi. As storage time increased, field fungi decreased until only 6 to 14% were invaded at 36 weeks.

No major differences in numbers of kernels invaded with field fungi were found between levels or drums.

*Penicillium* and *Aspergillus glaucus* groups were the most common fungi found during the experiment with only a few kernels invaded with
Field fungi (principally Cephalosporium and Fusarium) in corn infested with Sitophilus zeamaize stored at 68% r.h. and 80°F (26.7°C) during 36 weeks.
Fig. 10. Aspergillus glaucus counts in corn infested with Sitophilus zeamaize stored at 68% r.h. and 80°F (26.7°C) during 36 weeks.
Fig. 11. Penicillium spp. counts in corn infested with Sitophilus 
graminearum, stored at 68% r.h. and 80°F (26.7°C) during 36 weeks.
A. flavus, A. niger, and A. candidus.

In general, Penicillium counts (Fig. 11) were 20% or less and except for bottom samples of infested drums there was a tendency for a decrease with time. Bottom samples of infested drums tended to maintain a constant level of Penicillium invasion probably because the moisture content here was higher than in the other levels.

A. glaucus counts (Fig. 10) were never higher than 6% in the control samples. After 24 weeks A. glaucus increased in the bottom portions of infested drums. A. glaucus counts remained at 15 and 20% in bottom samples of drums I and II respectively, at 24, 28 and 32 weeks of storage. At 36 weeks invaded kernels increased to 64 and 54% respectively. In center samples A. glaucus remained below 10% for 32 weeks and then increased to 24 and 22% in drum I and II, respectively. There was no increase of invaded kernels in top samples of infested drums.

At the beginning of the experiment, a few seeds (less than 4%) were invaded by A. flavus, A. niger, and A. candidus. As storage time increased A. flavus and A. niger died but A. candidus was isolated from 8 to 12% of seeds from bottom samples of infested drums at 36 weeks. At this time A. candidus was also found in top and center samples of infested drums (less than 4%).

The fact that some Penicillium, A. flavus, A. niger, and A. candidus were found at the beginning of the experiment could indicate that the corn may have been stored at a moisture content above 16% or that seeds were invaded in the field.

Penicillium, A. flavus, and A. niger counts might be expected to decrease because they all require over 15% m.c. to develop.
and average moisture contents in the tests were always below 14.7%.

*A. candidus* in corn needs a minimum of 15% m.c. to grow (20), however it was found in samples with averages below 13.2% and 14.6% m.c. Probably individual kernels, especially those infested with insects, had much higher moisture content than the average for the sample.

*A. glaucus* requires a minimum moisture content of 14% in corn (20), thus the high *A. glaucus* counts found here.

Mold counts cannot be used to evaluate direct insect damage but give useful information about secondary effects of insect activity, such as increased moisture content and temperature which create conditions for fungal deterioration of grain.

**Loss of weight (Fig. 12)**

Data for weight loss are somewhat variable possibly due to the type and accuracy of scale used to weigh the drums.

Dry matter weight loss at 36 weeks was calculated to be 0.7% in the control, 4.4% in drum I and 3.0% in drum II.

This is one of the most direct methods to measure quantitative losses due to insect infestation. However, it would usually not be practical in actual farm storage.

In commercial storage, where grains are received and delivered on a weight basis and records are maintained, estimates of storage losses can be made if records are properly interpreted, i.e. if moisture, dust and other factors are considered.
Fig. 12. Dry matter weight loss of corn infested with *Sitophilus zeamaize* stored at 68% r.h. and 80°F (26.7°C) during 36 weeks.
SUMMARY

Changes produced in corn infested with the maize weevil (Sitophilus zeamaize MUSCH.) and stored at 80°F (26.7°C) and 65 ± 5% r.h. were monitored during 36 weeks storage. A major objective was to estimate the effectiveness of various tests to measure quantitative and qualitative losses in corn due to insect damage.

Number of insect-damaged kernels, as revealed by X ray after 36 weeks of storage, were similar at top, center and bottom sampling levels in infested drums. Most insects in probe samples were found in bottom portions of infested drums. There were more insects in center than top sections of these drums. In early sampling, few insects were detected, however, X ray showed damage present. Thus insect counts from probe sampling were not enough to estimate insect damage. A measure of damaged kernels would be more meaningful.

Corn in equilibrium with the test environment would have approximately 14% m.c., however, moisture content of control samples remained almost unchanged (average of 12.4%) during the 36 weeks. As a result of insect activity, moisture content increased to an average of 13.5, 13.9 and 14.7% in top, center and bottom portions of infested drums, respectively, after 36 weeks. Moisture content itself did not indicate the presence of deteriorative factors, however, considerable damage can occur before measurable changes in moisture content can be detected.
Temperature of infested and control corn during the 36 weeks of storage remained similar to that of the storage room (80°F). Under conditions used here temperature readings failed to show insect activity. Dust (throughs of No. 10 sieve) accumulated mainly in bottom portions of infested drums. Presence of dust may indicate insect activity, however, the weight of dust probably would not be a good indicator of insect population or damage.

Density results (pounds/bushel) were variable but a steady decrease can be noticed in infested samples. Test weight may be a good method to measure quantitative insect damage if a series of samples are taken from a lot of grain.

Viability of control samples decreased only slightly during the experiment. Bottom samples of infested corn decreased from over 90% to about 50% germination in 36 weeks. Germination decreased as insect and Aspergillus glaucus counts increased, however, whether the germination decrease was caused by insects or molds was not determined.

Glutamic acid decarboxylase activity (GADA) decreased in control and infested drums. Final GADA values of infested corn were lower than values of control. GADA does not seem to be a good test to measure insect damage.

Since fat acidity values (FAV) were always below 22 (maximum for sound corn) the test failed to show any damage caused by insects or fungi.

Proximate analyses (fiber, ash, protein and fat) also remained unchanged despite the observed damage.

Field fungi decreased with increasing storage time. Penicilli...
samples of infested drums but decreased in all other samples. _A. glaucus_ counts were closely related to moisture content, increasing mainly in bottom samples of infested drums where moisture was higher.

Mold counts did not indicate insect damage but gave useful information about the general condition of the grain.

2. AGRAWAL, N. S., C. M. CHRISTENSEN, and A. C. HODSON. 1957. Grain storage fungi associated with the granary weevil. J. Econ. Ent. 50:659 - 663.


APPENDIX
Table 1. Insect counts in corn infested with *Sitophilus zeamaize* MOTSCH. and stored at 68% relative humidity and 26.7°C for 36 weeks.

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* Total number of insects (live and dead)/kg
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Table 3. Moisture content (percent, wet basis) of stored grain X species

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Table 4. Dust weight (throughs of No. 10 sieve): Grams/kg

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### Table 7. Glutamic acid decarboxylase activity (GADA) in control and storage of Cyclommatus americanus

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* mm of CO₂/30g/30 minutes ± 100
Table 8. Proximate analysis: Percent. Moisture free basis.

Average of the three sampling levels.

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