ANAEROBIC STORAGE OF CORN WITH AND WITHOUT A DESICCANT

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By

Oghenetsavbuko Todo Edje

AN ABSTRACT OF A THESIS

Submitted to
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ABSTRACT

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The storage quality of high moisture shelled corn in closed containers with and without sawdust-salt mixtures on a laboratory scale was determined. Production of gas from each container was measured. Dried sawdust impregnated with 10% sodium chloride was used as a desiccant. In the corn, three moisture levels, 21.4%, 27%, and 30.8% wet basis, were used. Carbon dioxide prepared by the action of baking soda ($NaHCO_3$) on alum ($NH_4Al[SO_4]_2$) was used in flushing off the air in the interseed atmosphere. Viability and edibility of the corn were evaluated for 2, 4, 8, and 12-week storage periods.

All samples stored with the sawdust-salt mixtures remained clean and free flowing compared to those without the sawdust-salt mixtures, excepting in a few cases where the grains were not in close proximity with the desiccant. CO_2 production was greatly retarded by the sawdust-salt mixtures.

Germination was practically nil in all controls. Samples stored with the sawdust-salt mixture had an average of 96.67% germination during the first 2 weeks, and an average of 62.5% at the end of the 12th week. Germination decreased rapidly with increasing moisture content and time.

The edible quality rated higher with decreasing moisture content. Samples without the sawdust-salt mixture had a fermented smell but this gradually disappeared after soaking. There was no significant difference in edibility between treated and control samples.

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ANAEROBIC STORAGE OF CORN WITH AND WITHOUT A DESICCANT

1. INTRODUCTION

Corn, Zea mays, L. is one of the four most important cereal crops in the world. It is an energizing, heat-giving, and fat furnishing food for both man and livestock. Despite its importance from seed and food standpoint, several million tons of corn are lost annually in storage.

In the tropical and subtropical countries, the problem of storage is compounded by the hot humid climate. Thieme (71), in discussing the problems of storage in the less developed countries, remarked:

> . . . Most developed areas have rather hot humid climate. This creates rather unfavorable conditions for storage. The various causes of losses and the deterioration of quality of agricultural produce can be divided into two main groups: (i) moisture and temperature, (ii) insects and rodents. While the damage done by insects and rodents is more or less obvious, the influence of humidity and of temperature are indirect. They lead to biological changes by oxidation and hydrolysis, to microbiological spoilage by mold and bacteria, and to still more complicated processes as spontaneous heating, at times, combustion. In addition, excessive moisture and temperature of the

> stored produce create favorable conditions for insect activity. It goes without saying that all of these various processes are greatly activated by the high humidities and temperatures of tropical countries, and therefore, that the difficulties of storage are enormously increased in such countries. . . .

The Nigerian government outlined in her National Grain Policy in 1964 to increase the production of guinea corn (threshold grains), millet (threshold grains), maize (shelled grains), and rice (paddy) by 30.95%, 28.57%, and 46.67% respectively between 1957/58 and 1966/67 (32). It was designed to expand production because of the increase in population coupled with the rural exodus and a concentration of people in the big cities. If the government is to pursue her policy effectively, more and well preserved seeds will be needed. Produce must also be properly stored. In view of this, preservation of grains through sound storage practice is a matter of deep concern.

Although the problem of storage in the tropical and subtropical countries is mainly environmental yet other factors such as economic also enter the picture. This was well brought up by Thieme (71) when he was appraising the situation. He remarked:

> . . . chemical and technical solutions to the various storage problems can often not be applied because they are too expensive. Especially in the

> case of storage at the farmers' or village level, the first requirement for any solution is that it requires little initial capital investment and that recurring expenses are low. The farm or village level is very important because only by storage at this level can the farmer get the best price for his produce and the standard of living thus raised. . . .

The importance of sound storage practice in Nigeria cannot be overemphasized. In 1964, corn sold at $56 per ton during harvest and $126 during planting (32). This pattern was true for rice, guinea corn, millet, and yams. This two-fold increase in price was due mainly to scarcity resulting from poor storage facilities. Storing high moisture corn in sealed containers with desicants may provide a cheap and simple means of drying and preserving corn as food and seed. The experiments reported here are aimed at a method of preserving high moisture shelled corn in airtight containers on a small scale as food and seed.

2. OBJECTIVE OF THE EXPERIMENT

This experiment is based on the fact that reduced oxygen atmosphere retards the metabolic activity of micro-organisms. The corn kernels soon use up the oxygen in the storage medium until a point is reached where mold, insect development, and heating are prevented. Another fact on which this experiment is based is that of relative humidity and dehydrating agents. Chemical absorbents such as sodium chloride, calcium chloride, and a few others have great affinity for water. The object of this experiment is to determine the possibility of storing high moisture corn anaerobically, i.e. in an atmosphere of carbon dioxide and nitrogen and salt-sawdust medium for food and seeds. The practicability of such a method is measured in mold growth, dry matter loss, viability, and the edibility of the grains.

3. REVIEW OF LITERATURE

3.1 Physical Structure, and Chemical Composition of the Corn Kernel

The corn kernel is a one-seeded fruit. The seed, which is enclosed within the pericarp, consists of the embryo, endosperm, and the remnants of the seed coats and nucellus. In the mature kernel, the pericarp forms a protective cover. The seed coats beneath the pericarp are scattered noncellular remnants. There is a silk scar on the pericarp at the apical end of the kernel; at the lower end, the pericarp connects with the tip cap. Beneath the pericarp is the endosperm which is made up of cells filled with starch grains. The endosperm also consists of the aleurone layer, a thin layer of cells whose storage material is mainly protein. The endosperm forms about 85% of the kernel. The embryo, which is at the base of the kernel, is the vital part of the kernel. It consists of all the essential organs of growth, that is the plumule, the radicle, and the scutellum. The embryo forms about 10% of the kernel (41).

The corn kernel contains 61% starch, 1.4% sugars, 6% pentosan, 4% fat, and 2.3% fiber. It ranks high in total digestible nutrient and net energy, equalled only by wheat among the cereal crops. Being so rich in starch it is naturally low in protein. It contains 10% protein, mainly zein and it is deficient in tryptophane and lysine, two amino acids essential in animal nutrition (39, 42, 54).

3.2 Safe Moisture Level for Corn Storage

Several factors determine the length of time corn can be stored. Moisture content and temperature are the most critical. Corn is usually harvested when the moisture content is 25% or lower (6, 42). When shelled it can be stored for one year in unventilated bins at 13% wet basis or at 11% if it is meant for seed or to be stored for longer periods (6).

Although shelled corn can be stored at 13% moisture content wet basis for one year, it is the relative humidity of the atmosphere surrounding the grains that determines how long it can keep (6, 24, 25, 42). Dexter (24) in his experiment where grass and legume seeds were stored at various relative humidities, reported no mold growth for alfalfa

seed at 80% relative humidity for 4 weeks. During the same period mold growth was reported at 85% r.h. The grass species in this experiment came to equilibrium much faster and also molded quicker than the legume seeds. Molding was associated with the establishment of equilibrium.

3.3 Moisture Migration in Grain

Grains take up moisture quite easily when wet. The water is usually absorbed through the hilum and the seed coat. Moisture migration results from changes in temperature in the storage medium (6, 35, 42, 51).

In the United States, most grains are stored in the Fall when they are still warm. Under this condition, the air in the grain near the surface of the storage medium cools and moves down to the bottom and up the center where the air is warm. As the air moves up the center, it picks up moisture. This continues until it reaches the top where it comes in contact with cold grain and it condenses. Semeniuk et al. (68), stored yellow dent corn in bins of 1,000-2,700 bushel capacities for 3 years at Iowa. They reported that about 2½ feet of corn was spoiled at the top when it was stored over the Winter period.

If the grains are stored during the Winter, they are cold. As the season progresses into late Winter and early Spring, the temperature of the atmosphere increases. The air current along the edge of the bin gets warmer and rises. This warm air moves through the center of the bin and picks up moisture and later condenses at the bottom due to condensation.

Moisture migration and accumulation is noted in bins holding over 2,000 bushels. The effect is more pronounced with taller than shorter storage media of the same capacity (35). The effect of this moisture accumulation is molding and consequently a loss in quality and viability.

3.4 Effects of Moisture on Mold Growth

The microflora of cereal grains are made up of fungi, bacteria, and actinomycetes (2). Most of these are heterotrophs depending upon organic matter for growth. These microflora may be divided into three classes on a basis of moisture requirement for growth. They are hydrophytes when their relative vapor pressure is 90% and above, mesophytes, at 80%-90% and xerophytes, at below 80%. Molds grow at a lower relative vapor pressure than either bacteria or actinomycetes

hence they are the most important cause of deterioration in stored grains (2).

Fungi are again subdivided into field, storage, and decayed fungi (14). Field fungi invade the grains while they are still in the field at about a moisture content of 22-25% wet basis or 28-33% dry basis in the starchy cereal grains. The Fusarium, Alternaria, and Helminthosporium are the most predominant in corn fields. These soon die after harvest when the moisture content is below their requirement for growth. The storage fungi are mainly the Aspergillus species and a few of the Penicillium. These are adapted to life without free water and many can invade grains stored at 13-18% moisture content at 75-85% relative humidity. Nearly all these invade the embryo "primarily and preferentially." It is not impossible to see a whole grain with dead embryo due to an attack by internal mold. Decayed fungi resemble field fungi in their moisture requirement. Most of these are the Fusarium graminearum species (14).

3.5 Molds and Oxygen Requirement

Microorganisms such as bacteria, yeast, and fungi are divided into aerobes and anaerobes. Bacteria belong to

both classes and most fungi are strong aerobes (2). The rate of sporulation, spore germination, and the growth of the mycelium depend, among other things, on the oxygen concentration and the type of nutrient in the stored product (9).

Denny (19), showed that molds have a very wide range of oxygen requirement. Corn meal, innoculated with mycelium of Neurospora sitophila, was placed in containers of different oxygen concentrations. Mycelium growth occurred at 0.3% oxygen, by volume, after 16 hours. A 10%, by volume, of CO_2 did not retard the growth of the mycelia to any appreciable extent. At 0.3% O_2, and 32% CO_2, the growth was only slightly retarded.

The experiments of Bottomley et al. (7) and Miller et al. (50), showed that some fungi are surprisingly tolerant to low O_2 concentration. Miller et al. (50), incubated 6 mold cultures for 7 days in different O_2 concentrations and reported that Aspergillus flavus had a minimum and optimum O_2 requirement of 0.002% and 0.038% by volume for growth.

The idea that high CO_2 concentration can retard the growth and development of most molds, has prompted several investigations. Brown (9) reported that the growth, of several species of Fusarium and Alternaria, was retarded by

high CO_2 concentrations. He concluded that the effect of CO_2 was more pronounced at lower than at higher temperatures. Concentration of 17% CO_2 (43) has been reported to prevent mold on bread. In a storage medium where CO_2 concentration is high, yeast may proliferate. This has been reported by Teunisson (70) in experiments with rice. There are also reports that the effect of CO_2 may be stimulatory instead of inhibitory. This complicates the role of CO_2 in mold growth. Nevertheless the opinion that it retards growth is more widely expressed than otherwise.

3.6 Deteriorative Effects of Mold

The invasion of grains and grain products by microflora lowers the viability, storage quality, nutritive value, edibility, industrial usefulness, and increases the "off odor" and disease incidence to livestock and humans.

3.6.1 Loss of Viability

In storage media where molds are permitted to proliferate, seeds lose viability due to mold and heating. The storage fungi (14) attack the embryo. This causes death since it is the center of metabolic activity.

The loss of viability due to mold is well documented. Tuite et al. (72) brought this point out clearly, when they inoculated Mindum wheat variety, of moisture content (15.4 to 16.3) with Aspergillus ruber. At the end of 4 months, control seed that were treated with sodium hypochlorite had 82% germination and those inoculated, only 15%.

Christensen et al. (17) also reported that with increasing moisture content, there was an increase in the number of storage fungi. Rough rice stored at 14.5% and 15% moisture content had 48% and 0% germination after 4.29 and 420 days respectively. This was due to damage of the embryo by mold. Similar cases have been reported by Qasem et al. (64, 65) on corn.

"Sick" wheat is characteristic of toxic materials exhibited by mold especially of the Aspergillus species. This destroys the embryo showing dull and darkened appearance (53). Increase in the population of microflora increases heating. This can increase the fat acidity and lower the germination. This creates a problem when it is realized that about 30% of the embryo of a kernel is made up of oils and fats. Fat acidity has been used as an index of soundness of grains (73, 75).

Peterson (62) indicated that high O_2 concentration encourages mold growth resulting in loss in viability and germ damage. Samples of hard red Spring wheat which had 88% germination were conditioned to 18% moisture content and stored at 30°C for 16 days in 2 sets of gas mixtures. One was a mixture of O_2 and N_2 containing 0.2-21% O_2 and the other containing 21% O_2 with varying amounts of CO_2 and N_2 to provide CO_2 levels from 0.02-79% by volume. At 0.5% O_2, the viability was 58% and germ damage was 60%. At 20% CO_2, viability was 22% while germ damage was 52%. In the presence of 21% O_2, CO_2 had little effect on respiration until the concentration exceeded 13.8-18.6%. At 50-70% CO_2, the viability was high and there was little or no germ damage. Similar results of loss of viability and increase in germ damage in the presence of high O_2 concentrations have been reported by Nagel et al. (55) and Swanson (69).

3.6.2 Loss of Nutritive Value and Quality

Chemical changes which have effect on nutritive values are continually taking place in all grains and milled products of grains no matter how they are stored. Most of these changes are detrimental.

Bottomley et al. (7, 8) have demonstrated a marked disappearance of non-reducing sugars in corn stored under conditions that favored deterioration (7). They stored yellow corn at 30°C at 4 different moisture contents between 19 -31% for 12 days both in sealed and aerated containers. They reported that there was a decrease in non-reducing sugar with increase in moisture content and mold count with aerated samples. The growth of Aspergillus glaucus appeared to cause a rapid loss in non-reducing sugars and little increase in fat acidity.

In the wet milling of corn, it has been noted that it is more difficult to obtain a good separation of the starch from the other constituents of the grain when the corn has been stored under unfavorable conditions (2).

Cox et al. (18) reported that heat damaged corn yielded only 45% as much starch as normal kernels. The viscosity of suspensions of the starch in water was low, and in many instances the starch granules were split into wedge-shaped fragments.

Although poor storage may have adverse effects on the quality of corn from nutritional standpoint, the picture is different for rice. Fresh rice contains active alpha-amylase. This causes rice to be sticky when cooked. To

avoid this, it must be inactivated during storage. Jones et al. (40) studied the changes in the physical and chemical properties of the proteins of corn. Samples of Yellow Dent corn, whole kernels and ground kernels, were stored over a period of 24 months at a relative humidity of 55%. They reported a decrease in solubility of protein of about 60% less than that of fresh corn. There was also a decrease in digestibility test when rations were made from these samples and fed to young albino rats. The loss in nutritive value was not due to mold growth since only one sample had an indication of mold. This must have resulted from denaturation of the proteins.

Fats break down rapidly during storage causing a rise in fat acidity. This results from the action of lipase on fat breaking it into fatty acids and glycerol. Fat hydrolysis is much faster than either carbohydrate or protein hydrolysis. Hence the free fat acidity is an index of the soundness of most grains.

3.6.3 Toxicity of Molded Grains

Molded grains are usually fed with caution because they are sometimes harmful to humans and livestock.

Christensen et al. (14) innoculated barley with different fungi to produce blighted barley. These were then fed to pigs. Barley that were naturally infected by fungi were also fed in this experiment. Barley blighted by Helminthosporium and Alternaria was not toxic to pigs even when it contained up to 31% of the blighted kernels. Pigs which were fed with barley blighted by Fusarium had Fusarium poisoning. They lost appetite, became lifeless, weak, vomited, and even died. In the same experiment, corn blighted by Fusarium graminearum was made into a ration. Ten parts of this corn to 90% basal food mixture was fed to pigs that have been fasting for 24 hours. With reluctance they ate ¼ of a pound during the first 15 minutes. About 10 minutes later they ceased eating. Toxic syndrome was observed and they vomited 8 times in 90 minutes.

Christensen et al. (17) also reported that swine fed corn infected with mold, produced strong extrogenic symptoms in them. Tumefaction of the vulva was also reported. Pigs have also been reported to vomit grains carrying excessive amounts of Gibberella zeae (2).

Christensen et al. (17) innoculated corn with various fungi and fed them to rats. Rats fed this ration had an average uterus weight of 49 mg. while those on control had 29.6 mg.

Poultry fed moldy grains have been reported to develop aspergillosis. Corn infected with Gibberella zeae has been reported to cause dizziness, headache, and exhaustion to man.

While fungi have been stated to be toxic to man and livestock, they have also been reported to be rich in protein and in vitamins. Gorcica et al. (34) cultured Aspergillus sydowi in a medium so that any vitamins found in the medium shall have been synthesized by the organism. The mycelia were put in cans and frozen. They were ground and incorporated wet into a ration. The ration was fed to rats and chicken. The reported that the addition of 10% of the mycelium to the ration was able to support the growth of rats whose ration was low in vitamin B. It was also able to protect chicken against polyneuritis. Pellagra was prevented by the addition of 1% of the mold in the ration.

3.7 Respiration

Respiration is an oxidative process that occurs in all living cells. This provides the protoplasm with the energy needed for its metabolic activities. The gaseous exchange involving oxygen is aerobic respiration.

Respiration in which CO_2 is evolved and O_2 is not absorbed but may occur in its presence is called anaerobic respiration. A typical equation for aerobic respiration is (48):

$$C_6H_{12}O_6 + 6O_2 = 6CO_2 + 6H_2O + 673 \text{ kg-Cal.}$$

or

$$C_6H_{12}O_6 + 6O_2 = 6CO_2 + 6H_2O + 673 \text{ kg-Cal.}$$

180 lbs.+ 6x360 cu. ft. = 6x360 cu. ft. + 6x18 lbs.(26)

In the above equations, the volume of CO_2 produced is equal to the volume of O_2 consumed. A complete hexose oxidation has a respiratory quotient of $CO_2=O_2$ of 1, while fats is 0.7. Anaerobic respiration also takes place in every living cell. The end product is CO_2 and a number of relatively simple organic compounds. A typical equation is (26):

$$C_6H_{12}O_6 = 2C_2H_5OH + 2CO_2 + 22.0 \text{ Cal.}$$

180 lbs. = 2x46 lbs. + 2x360 cu. ft.

The "pound molecular volume" (26) is very useful in predicting or calculating loss of dry matter in closed storage medium.

3.7.1 Effects of Moisture Content on Respiration

The amount of moisture in the grain within certain limits is one of the determining factors in the rate of respiration. One of the reasons why increase in moisture content increases respiration is that the colloids of the cell imbibe water to a gel-like substance. With increase in water

content, more water is absorbed until the gel becomes less viscous. Diffusion of gas through such media is quite rapid. Since the rate of respiration is a function of the rate of gaseous diffusion and absorption, the less viscous, the greater the rate of respiration (49).

Olafson et al. (56) have shown that the respiration increases with increase in moisture content. They conditioned White Dent corn to six moisture contents ranging from 12.9-17.0%. These were stored at 30°C for 14 days. At the end of the experiment, corn stored at 14.5% had a respiration rate of 0.90 mg. CO_2/100 gm. dry matter for 24 hours. A difference of 2.5% m.c. resulted in a 60-fold increase in the rate of respiration. Similar results have been reported by Ragai et al. (66) on corn and by Milner et al. (51) on soybeans.

3.7.2 Effect of Gas Content on Viability

The type and volume of gas in a storage medium has effect on the rate of respiration of stored grains and the rate and degree of deterioration.

Padua (58) stored rough rice in different gas proportions in airtight containers at 18%, 22%, and 26% m.c. for 60 days. He reported a poor germination where initial

O_2 concentration was high. Sayre (67) also reported a loss in germination when corn was stored in a high concentration of O_2.

3.8 Chemical Absorbents

The principal method of preserving grains has been drying. This old method is based on the fact that the reduction in the moisture content of the stored product renders destructive organisms inactive. Construction of driers can be expensive and this has led to the use of less expensive material in accomplishing the same purpose.

Hurst and Humphries (37) tested the practicability of silica in drying grains. Soybeans, wheat, corn, and rice whose initial moisture content ranged from 8.9 to 14.6 were conditionted to 20%. The drier, silica, equivalent of four times the weight of the water added, was then placed in the container and mixed with the grain by shaking and tumbling. The container was then sealed and allowed to stand for a desired length of time. In 24 hours, corn that was 20% had 13.4% moisture content. Similar decrease was reported for other crops in the same experiment. They remarked that although calcium chloride and sodium chloride have great

affinity for water, they would doubtlessly have an injurious effect on stored grains.

This idea of chemical absorbents was later developed on a larger scale by Dexter et al. (29). They obtained blocks of wood that have been sawn into about ½ to 1 inch thick and about 1 inch in cross section. These were placed in boiling calcium chloride solution, with a specific gravity of 1.20, that was acidified slightly to remove troublesome carbonates. This was boiled for about 15 minutes. After being submerged for about 15 hours, they were dried at about 102°C. These blocks were then used in drying soybeans and oats. One thousand pounds of soybeans at 16.25% moisture content were placed in a tight bin. Forty pounds of the dehydrator was mixed with the soybeans to remove about 20 pounds of water. After 36 days the moisture content of the soybeans indicated that the blocks had absorbed 18.4 pounds of water. In the same experiment 1000 lbs. of freshly harvested oats with 18.5% moisture were placed in a bin together with 100 pounds of dehydrator blocks. At the end of 11 days, the blocks were removed and found to weigh 146 pounds. The oat at this time had 12.5% moisture content.

Another experiment that has been conducted by Dexter is the conditioning of beans with sawdust-sodium chloride

mixture. The result is yet to be published. Beans with 22% moisture content were mixed with the "drier." A few days later the moisture content was 16.5%. No damage due to mold or insect has been reported. The experiment is over 6 years now and the beans are still free flowing (27). Hall (35) has also used calcium chloride in keeping the moisture content of corn in 3200 bushel bin below 14%.

A more recent experiment is that of Chaves (10). He used sawdust-salt mixture in storing corn. Salt was dissolved in water to make a saturated solution. A sawdust-salt drier was prepared by mixing 1 part by weight of the solution to 3 parts by weight of sawdust. Equal volumes of corn and sawdust were placed in jars and sealed. Ten days after the corn with 20% moisture had 12.24% m.c. The drier had absorbed 8.85 gm. of water.

This relative humidity approach to drying and storing farm products has been used in conditioning popcorn for the best popping (20). By storing popcorn with sawdust-salt mixtures, Dexter (20) was able to pop popcorn to 26 times the original volume of the popcorn, with 98% of them popping.

4. METHODS AND MATERIALS

This experiment was carried out on a laboratory scale using 300 cc flasks. A three-way hybrid corn, WFGMS x MS206 x B8, with large round grains obtained from Dr. E. C. Rossman of Michigan State University, East Lansing, were used. The original moisture content was 7.6%. Germination test was 95.9%. Each treatment condition was replicated twice and a code was designed to identify each treatment.

Example: (i) (27- OC- OS- 2-1) means

Moisture content.....27%
Flask not flushed with CO_2.....OC
Stored without sawdust salt mixtures...OS
Storage period in weeks.....2
Replication number.....1

(ii) (27- C- S- 4-2) means

Moisture content.....27%
Flask flushed with CO_2.....C
Stored with sawdust salt mixture.....S
Storage period in weeks.....4
Replication number.....2

4.1 Preparation of the "Dehydrator"

A "dehydrator" in this text is defined as a dried mixture of sawdust moistened with sodium chloride (rock salt)

solution or a dried mixture of sawdust and calcium chloride solution as prepared in the manner described below.

The sawdust used was a combination of several trees. It was screened through 8/64 round screen. The sawdust on the top of the screen was used. It was decided to use this portion of the sawdust because the discarded portion was very powdery and this might adhere to the grains at the end of the experiment and thereby create a problem of separating the "dehydrator" from the grains. Sawdust was used instead of "wood blocks" because in an earlier trial by the author, it was observed that grains nearest or in close proximity with the "wood blocks" were drier than those at the top of the sample. This was expected since the moisture from the grain moved by diffusion. The longer the distance, the slower the rate of diffusion, and the greater the chances of spoilage. Rock salt was used instead of table salt as a result of an earlier experiment by Chaves (10). A concentrated solution of rock salt was made up thus; three parts of rock salt was dissolved in 10 parts of water by weight. This solution was mixed with the sawdust to give a proportion by weight of one part rock salt in nine parts of sawdust. This was dried in the oven at 82^{o}C for 24 hours.

4.2 Moisture Content of Corn Grains

Three moisture levels, 21.4%, 27%, and 30.8% wet basis were used. (These moisture levels hereinafter referred to as 21%, 27%, and 31%.) The grains were remoistened to the desired moisture levels by using this equation:

$$M = \frac{NxP}{Q}$$

where:

M = Weight after remoistening
N = Weight before remoistening
P = Initial dry matter percent
Q = Desired dry matter percent.

Example:

100 grams of corn at an initial moisture content of 7.6% (dry matter 92.4%) would weigh M grams at 21% moisture content (dry matter 79%).

where:

$$M = \frac{100 \times 92.4}{79} = 116.96 \text{ grams.}$$

4.3 Storage Period and Temperature

Four storage periods of 2, 4, 8, and 12 weeks were scheduled. The samples were not stored at a definite temperature. The time, June to September, was chosen because temperatures at this time reflected those in most tropical and subtropical countries. During the experiment, the

average room temperature was 87.3°F. The lowest was 78°F, highest, 96°F, and a range of 18°F.

4.4 Filling the Flasks

The flasks that were stored without a "dehydrator" were filled with corn of about 200-225 grams each. Treatments in which the interseed gas was not flushed off with CO_2 had a one-holed size six rubber stopper carrying an inverted "V" shaped glass tube which was connected to a graduated test tube by a 36-39 inch rubber tubing. Treatments in which the interseed gas was flushed off with CO_2 had a two-holed size six rubber stopper carrying an inverted "V" shaped, and a long inverted "L" shaped glass tube. The former tube carried a 36-39 inch rubber tubing connected to a test tube. The latter tube was connected to a CO_2 "tank" by a short rubber tubing. This tubing also had a clamp. Samples stored with a "dehydrator" were filled by a mixture of 90 grams of corn and 30 grams of the "dehydrator"(10). The rubber stoppers for these were of the manner described above.

4.5 CO_2 Preparation and Flushing off of Interseed Air

The CO_2 was prepared by the action of baking soda ($NaHCO_3$) on alum according to the equation:

$$6NaHCO_3 + 2NH_4ALCSO_4)_2 = 2AL(OH)_3 + NaSO_4 + (NH_4)_2SO_4 + 6CO_2.$$

This was collected over water in a "CO_2 tank." Interseed atmosphere was flushed off by pouring 130 cc. and 200 cc. of water through a funnel into the tank connected with a rubber to those stored with and without the "dehydrator" respectively. This water dispaced equal volume of CO_2 into the flasks. The flushed air escaped through the shorter rubber tubing. It was then tied and clamped.

4.6 Gas Collection

The gas produced by each sample during the respiration was collected over water in graduated test tubes. These were refilled when necessary. Gas collection was stopped on the 11th day as a result of an earlier trial.

4.7 Final Moisture Content

At the end of each treatment period, samples stored with the "dehydrator" were poured into a 12/64 round screen.

The "dehydrator" was screened from the grains and weighed. The amount of moisture absorbed was calculated using this equation:

$$A = B - C$$

where:

A = Weight of moisture absorbed
B = Final weight of "dehydrator"
C = Initial weight of "dehydrator."

In order to avoid loss of the "dehydrator," it was weighed directly from the pan beneath the screen.

The final moisture contents were determined by obtaining samples of 30 gms. of corn and putting them into a vacuum oven. A preliminary trial showed that after heating the oven at 100°C for about 96 hours, there was no significant loss in weight. All samples were, therefore, heated at 100°C for four days.

4.8 Viability Test

Thirty "sound" looking seeds were selected from each sample. These were rolled in moistened paper towels. Both ends were tied. The paper towels were placed in trays and were placed in a germinator at a temperature of 84-88°F. A previous trial indicated that all viable seeds would germinate

in 6 days, and that seeds that did not germinate within this period would not germinate even after they had been dried on a laboratory shelf 12 days after remaining in the germinator. Germination count was, therefore, taken 6 days after sowing.

4.9 Edibility

About 100 gms. made of both replicates, were soaked in water. These samples were kept in the laboratory for a period of 90-96 hours (about 4 days). At the end of the first two days, they were washed, and washed again at the end of the fourth day. These were then ground in a "wet mill." At first a coarse plate was used. A finer one was used in grinding them thrice. Each time the finer plate was tightened to have a more thorough grinding. After the first grinding, some of the membranous, fibrous pericarps were removed by hand after soaking them in water, shaking them and carefully removing them by hand. After grinding, the ground mass was filtered through cheese cloth one layer thick. The decantate was allowed to set for about one hour. A meal was then prepared by adding 1 tablespoonful of damp settled starch to 1 tablespoonful of water. This mixture was then poured into about 150 cc. of boiling water. This was vigorously

stirred until a semi-gelatinous meal was done. This was poured into a dish, cooled off and eaten. No sugar was added. The meal was graded on a point system, thus:

Very good.....1
Good.....2
Fair.....3
Poor.....4
Very.....5

The grading was based on appearance, smell, and taste.

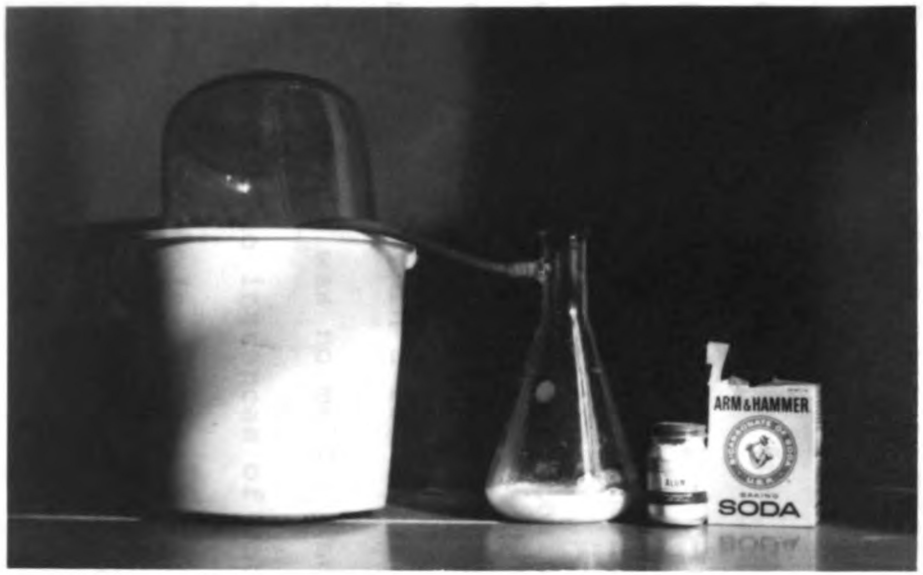

Fig. 1.--Preparation of CO_2 by the action of baking soda on alum.

Fig. 2.--The set-up of the experiment. Gas was collected through rubber tubing connection between flasks and graduated test-tubes.

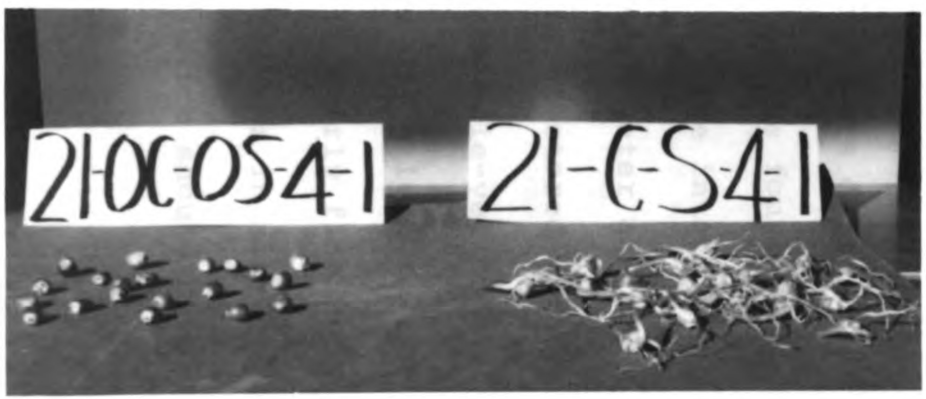

Fig. 3.--Effects of storing corn at 21.4% moisture content with the "dehydrator" and in an atmosphere of CO_2 on germination.

5. RESULTS AND DISCUSSION

5.1 Physical Appearance of the Grains

All samples stored with the "dehydrator" at 21- and 27% moisture content were free flowing and clean except in flasks where there were cases of isolated mold growth. The grains rattled on the screen as they were being separated from the "dehydrator." Samples stored at 31% moisture content were not as free flowing as those described above. These also had a mild fermented smell.

Samples stored without "dehydrator" were dull in appearance, and not as free flowing. After the fourth week, samples 31-OC-OS- and 31 - C-OS- were wet. The interior of the flasks were wet and almost dripping with water as they were being emptied. Sectioning of the grains stored without the "dehydrator" showed that the germs were pale-yellow to ochre. Except in a few cases where there was a leak in the flask, there was no mold. The grains felt fairly firm and there were no instances of rotting or moldiness in air-tight flasks. Very few grains had brown germs which should have

resulted from heat damage. These samples had a fermented smell as compared to those stored with the "dehydrator."

5.2 Appearance of Mold

Anderson (2) and Brown (9) have shown that most fungi are strong aerobes. Their growth is retarded by high concentration of CO_2.

In the Spring of 1965, the author filled several 300 cc. flasks with shelled corn. Two flasks were half filled with corn at 16% m.c. and another two, filled 2/3 with corn at 25% m.c. After 12 days of storage, the former had molded extensively. The latter did not mold until the 17th day and then only slightly. At the end of the experiment (21 days), those stored at 25% m.c. had produced three times more CO_2 than those stored at 16%. Apparently the rate and total volume of CO_2 produced by the 16% moisture corn was not enough to flush out the interseed atmosphere and this trapped air must have been responsible for the mold growth. At the end of the experiment, 1.35 gm and 1.95 gm dry matter had been lost at 16% and 25% m.c. respectively.

In this present experiment, no samples stored without the "dehydrator" were moldy excepting two samples stored

at 21% and 27% m.c. that were slightly moldy 12 and 23 days later. The absence of mold was due to the volume of CO_2 produced that flushed out the interseed atmosphere (see Fig. 5a).

No treatments coded 21- OC- S- for the entire 3 months were moldy excepting in 2 instances of either slight or isolated mold. This was due mainly to grains not being in close proximity to the "dehydrator." No treatments coded 27-OC-S- were moldy excepting air leakage in a few flasks that caused slight molding near the stopper.

5.3 Moisture Absorbed by "Dehydrator" and Final Moisture Content

Figs. 4a, 4b, 4c, and Table 1 show the effect of the various treatments on the final moisture content. Samples stored at 21% m.c. without the "dehydrator" were 21.10% and 21.7% at the end of 2 and 12 weeks respectively. Samples stored at 21% m.c. with the "dehydrator" were 13.84% and 15.25% at the end of the above mentioned periods. Table 1 shows that the "dehydrator" was highly significant in reducing the moisture content especially in the first two weeks. Fig. 4a shows that flushing out the interseed atmosphere reduced the moisture content. The effect of this, however, was less than that of the "dehydrator." Samples stored at

21% m.c. without the "dehydrator" but had the interseed atmosphere flushed had 20.9% and 21.25% at the end of 2 and 12 weeks respectively; while those stored at the same moisture with the "dehydrator" and the interseed air flushed, had 11.99% and 13.75% at the end of the same period. Figs. 4b and 4c show the final moisture content of samples stored at 27% and 31% m.c. From these graphs, it is evident, excepting in 27-OC-S-, that the grains became wetter after the first 2 weeks. That is the longer the grains were stored, the wetter they become. In view of this, the "dehydrator" should have been screened off at the end of 2 weeks or earlier, and replaced with a new "dehydrator." The increase in moisture content with time indicates that moisture (salt solution) was being reabsorbed and this could cause salt injury resulting in poor germination.

The amount of moisture absorbed by the "dehydrator" was determined by either weighing the grains or the "dehydrator" at the end of the experiment. The least amount of moisture absorbed was 20.4% and the highest, 41.83% of the "dehydrator's" original weight. These figures were in agreement with Chaves (10). In an earlier trial in which one inch cube "dehydrators" were used, the moisture absorbed was 57.29% from a sample that was originally 29.05% m.c. The one-inch cube "dehydrators"

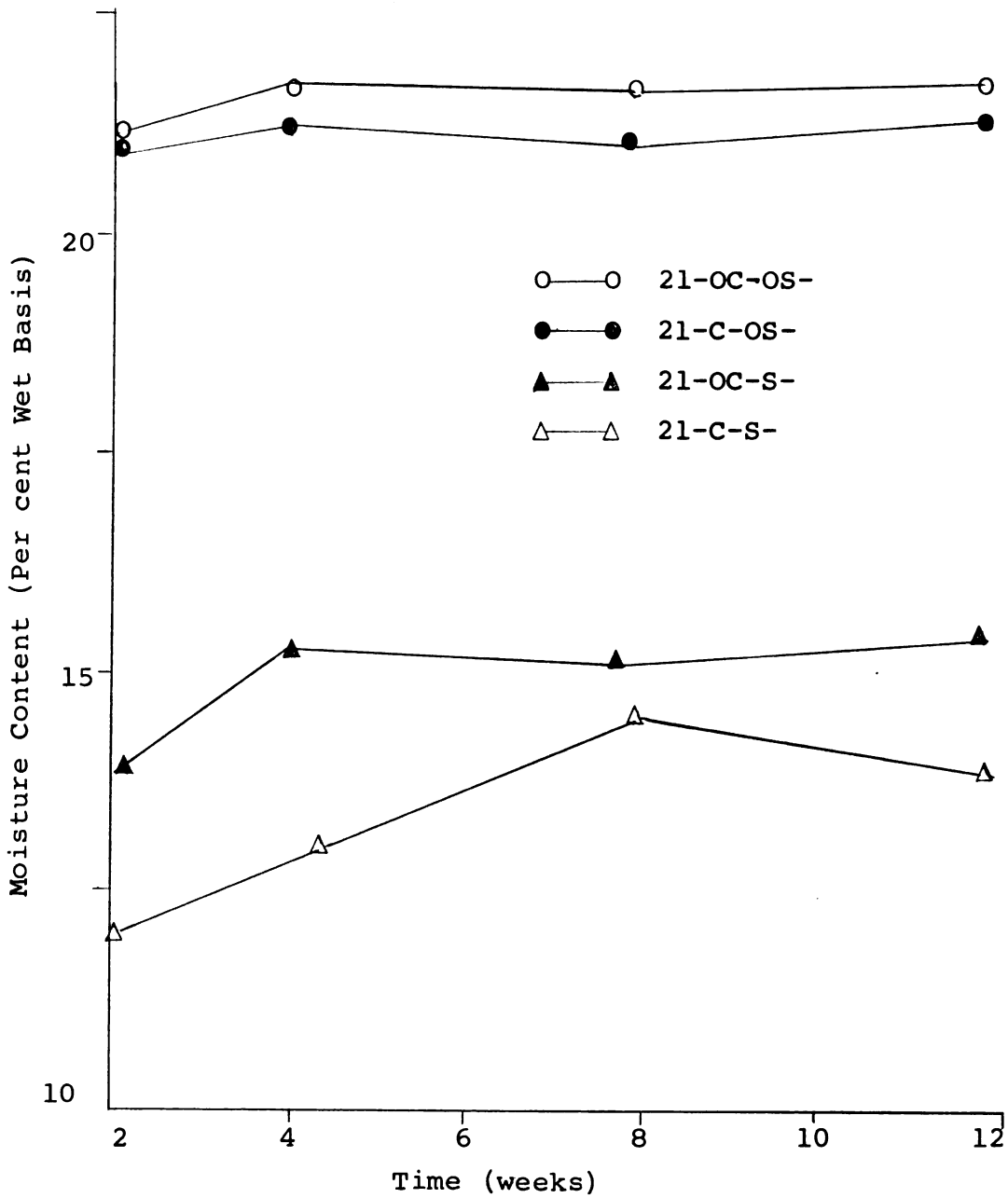

Fig. 4a.--Final moisture content of all treatments stored at 21.4% m.c.

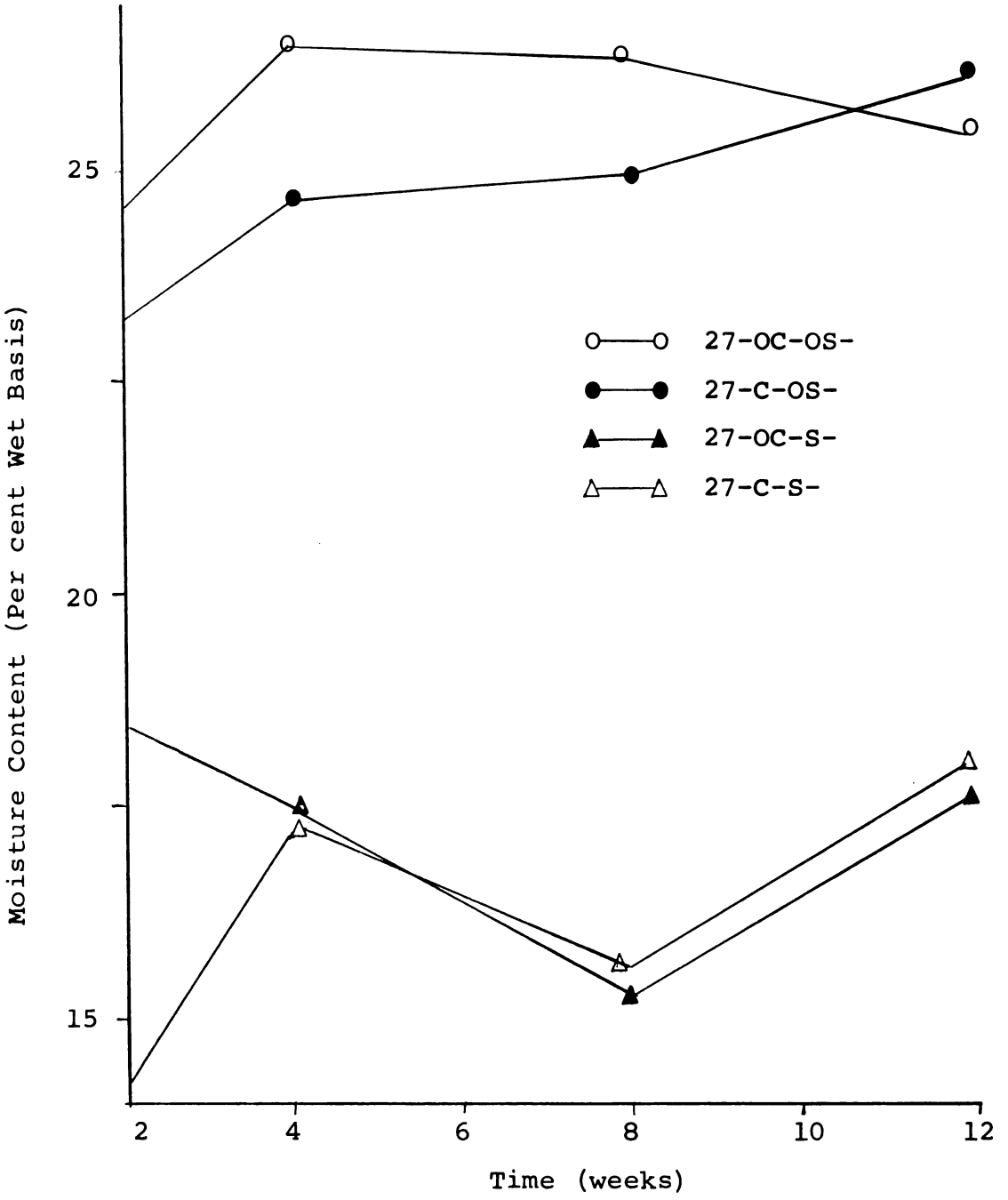

Fig. 4b.--Final moisture content of all treatments stored at 27% m.c.

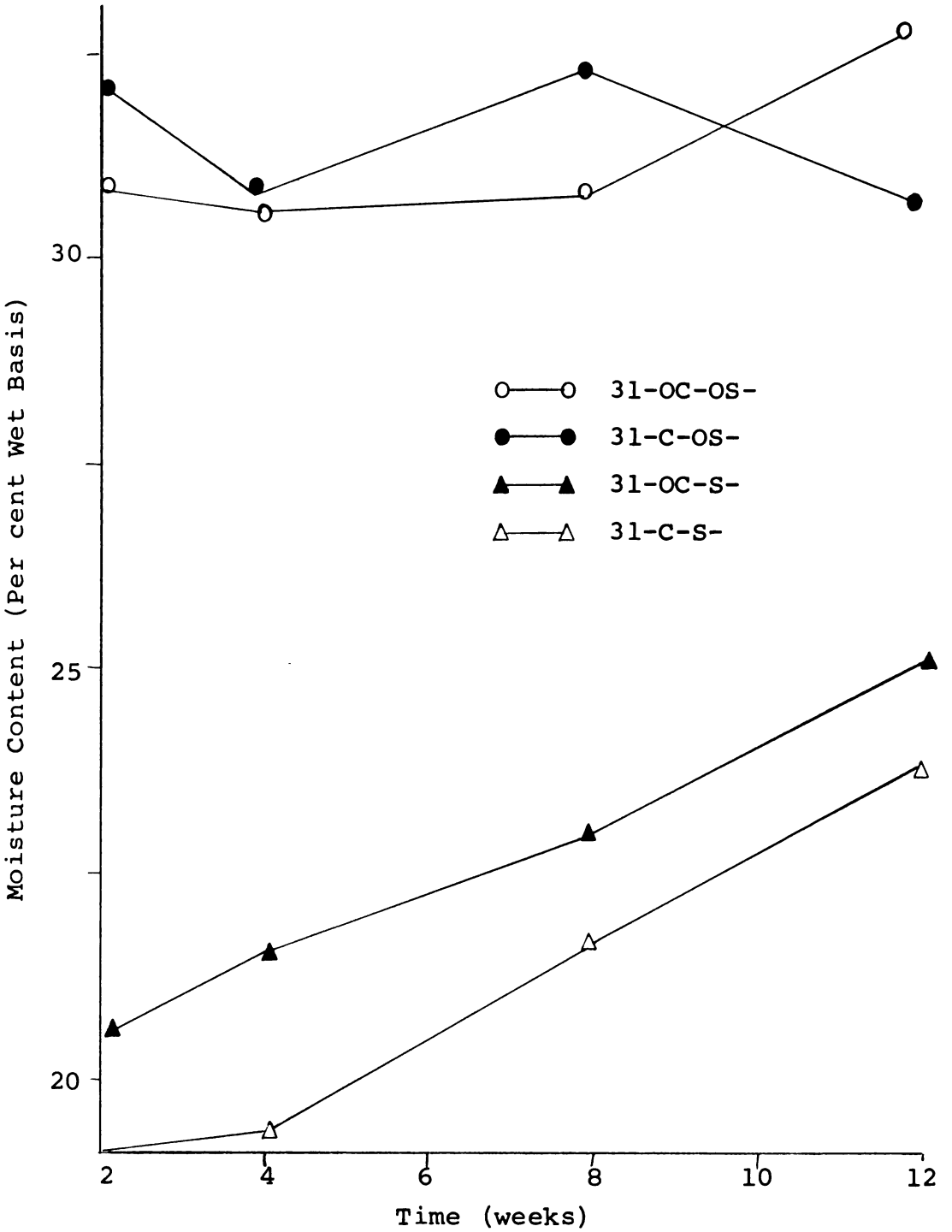

Fig. 4c.--Final moisture content of all treatments stored at 30.8% m.c.

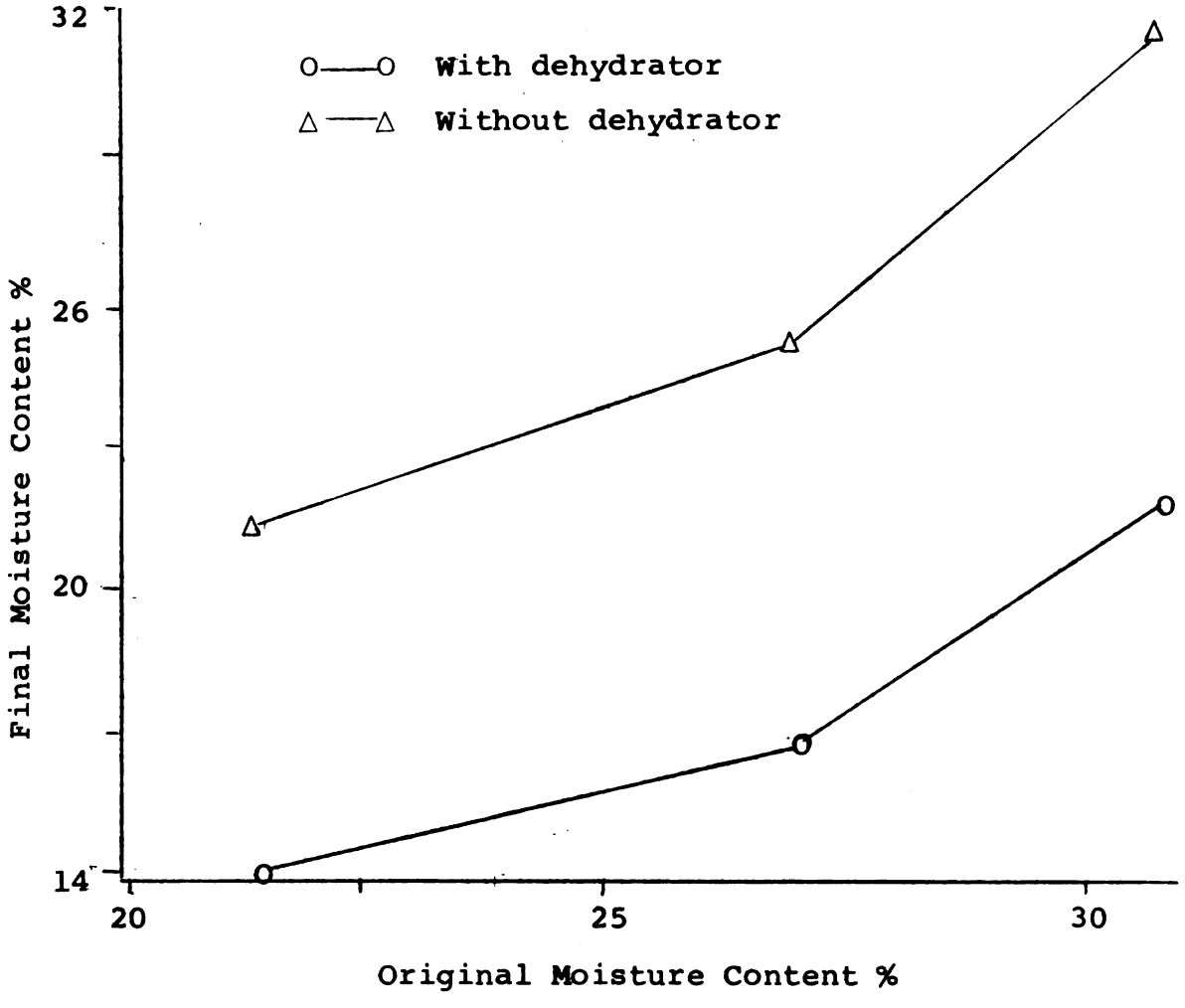

Fig. 4d.--Relationship between initial and final moisture content when corn is stored with and without the "dehydrator."

Table 1.--Analysis of variance: Measured variable: Moisture content at the end of each treatment.

Source of Variation	d.f.	Mean Square	Fratio	40.99
A	1	.03	22.48**	7.19
B	1	3.88	3133.43**	7.19
AB	1	.0003	.23	7.19
C	2	1.31	1059.18**	5.08
AC	2	.0002	.12	5.08
BC	2	.96	771.56**	5.08
ABC	2	.03	26.33**	5.08
D	3	.45	360.3 **	4.22
AD	3	.03	20.18**	4.22
BD	3	.03	204.88**	4.22
ABD	3	.25	2.05	4.22
CD	6	.003	43.76**	3.20
ACD	6	.05	10.71**	3.20
BCD	6	.01	85.14**	3.20
ABCD	6	.11	14.81*	3.20
Error	48	.02		
Total	95	.001		

** Highly significant
A = CO_2
B = Dehydrator
C = Moisture
D = Time

absorbed more water, but grains not in close proximity with them were extensively moldy.

5.4 The Dehydrating Effects of Sodium and Calcium Chlorides

In another experiment, "dehydrators" of sodium chloride and calcium chloride were prepared. The grains were the same as described earlier. They were remoistened to 29.12% m.c. The treatment consisted of mixing the sodium chloride dehydrator--"dehydrator a"--with corn in the proportion of 1:3 by weight. After a definite period, this was screened off and remixed with calcium chloride "dehydrator"--"dehydrator b"--in the same proportion as above. If the original dehydrator was "dehydrator a" it was changed to "dehydrator b" and vice versa. Of those stored with either "dehydrator," one set of samples was "sealed" and another was "opened." Flasks that were "opened" had a two size-six hole rubber stoppers. These holes were big enough to allow insects to invade the storage medium. The "dehydrators" were changed two and five days later. A code was used to identify the treatments. Example:

Na - O- 2- 1 means
Stored with "dehydrator a".....Na
"Opened".....O
Storage time in days before change.....2
Total storage period in months.....1

Ca- C- 5- 2 means
Stored with "dehydrator b".....Ca
"Closed".....C
Storage time in days before change.....5
Total storage period in months.....2

Table 2 shows the result of this treatment. At the end of the first month, treatment Ca - C - 5 - 1 had 10.85% m.c., the lowest, and treatment Ca - C - 2 - 2 had 14.08% m.c., being the highest. The grains were free flowing, rattled on the screen and there was no single case of moldiness. This result compared to Fig. 4c shows that this method was more effective in reducing the moisture content of the grains.

Table 2.--Effects of alternating "dehydrators" on the final moisture content and germination of corn stored at 29.02% at the end of 1 and 2 months.

Treatment	Final moisture content %	Germination %
Na - O - 2 - 1	12.37	98.33
Na - O - 2 - 2	12.94	93.33
Ca - O - 2 - 1	11.58	95
Ca - O - 2 - 2	12.62	95
Na - C - 2 - 1	11.92	100
Na - C - 2 - 2	13.08	95
Ca - C - 2 - 1	11.74	95
Ca - C - 2 - 2	14.08	95
Na - C - 5 - 1	12.94	80
Na - C - 5 - 2	13.78	83.33
Ca - C - 5 - 1	10.85	88.33
Ca - C - 5 - 2	12.58	91.65

5.5 Respiratory Activity

Bailey and Gurjar (5) stated that moisture was a determining factor in respiration. They believed that it increased with increasing moisture in a gradual and fairly uniform fashion until 14.5% m.c. after which the rate is rapid. Accumulation of CO_2 in a sealed respiratory chamber is generally believed to retard respiration. This fact was first recognized by the Egyptians and the Romans. Controlled atmoshperic storage is based on this principle.

Figs. 5a, 5b, and 5c show the volume of CO_2 produced per 100 gms. dry matter. Samples stored without the "dehydrator" at 21% m.c. produced 94.9 cc. of CO_2 in 270 hours. Those stored at the same moisture but with the "dehydrator," and also not flushed with CO_2, produced 4.2 cc. of CO_2 in 270 hours. The difference of (94.9/4.2) 22.6--fold must have been due to retardation of respiration resulting from reduced moisture content. Samples stored with the "dehydrator" at 31% m.c. (31-OC-OS-) produced 211.9 cc. of CO_2 while those stored without the "hydrator" (31-OC-S) produced 101.6 cc. of CO_2 in 270 hours. This represents (211.9/101.6) 2-fold increase in production. The "dehydrator" was not as effective in reducing the moisture at 31% as in 21%, hence more CO_2 was

produced. Flushing out the interseed atmosphere also retarded respiration. The "dehydrator" was more effective than the flushing out of the interseed atmosphere. The "dehydrator" was more effective at lower than at higher moisture levels.

Results from dry matter loss indicated that more dry matter was lost when stored without a "dehydrator" than stored with it. A comparison between 21-OC-OS- and 21-OC-S- showed that the latter lost only 57% of its expected value. No doubt the "dehydrator" reduced the moisture of the grain, therefore retarding respiration. The importance of this can not be overemphasized when these grains are being stored either for seed or food.

5.6 Expected Dry Matter Loss

From Dexter's "pound-molecular volume" the amount of dry matter lost in storage can be computed from the amount of gas produced.

$$C_6H_{12}O_6 + 6O_2 + CO_2 = 6CO_2 + 6H_2O + 673 \text{ kg. Cal}$$

180 lb. + 3x360 cu. ft. = 6x360 cu. ft. + 6x18 lbs.

Since the sample stored in this experiment was about half a pound, the formula:

$$C_6H_{12}O_6 + 6O_2 = 6CO_2 + 6H_2O + 673 \text{ kg. Cal}$$

180 gm. + 6x22.4 liters = 6x22.4 liters + 6+18 gm.

shall be used for this calculation. The volume of air in the flask stored at 31% for 2 weeks was 117.9. If oxygen is 20% of air, then 23.6 cc. was oxygen. Under aerobic condition, this will produce 23.6 cc. of CO_2. Substrate needed for this convertion is:

$$\frac{X_1}{180} = \frac{23.6 \text{ cc.}}{6x22.4 \text{ x } 1000 \text{ cc.}} = .03 \text{ gm.}$$

Average CO_2 production of 2 samples for 2 weeks at 31% m.c. was 315. That is 315-23.6 was produced anaerobically. Substrate needed for this convertion is:

$$\frac{X_2}{180} = \frac{291.4}{2x22.4 \text{ x } 1000 \text{ cc.}} = 1.17 \text{ gm.}$$

$\therefore$ Total substrate consumed = $X_1 + X_2$

.03 + 1.17 = 1.20 gm.

This is close to the observed dry matter loss of 1.25 gm. in the 2 samples.

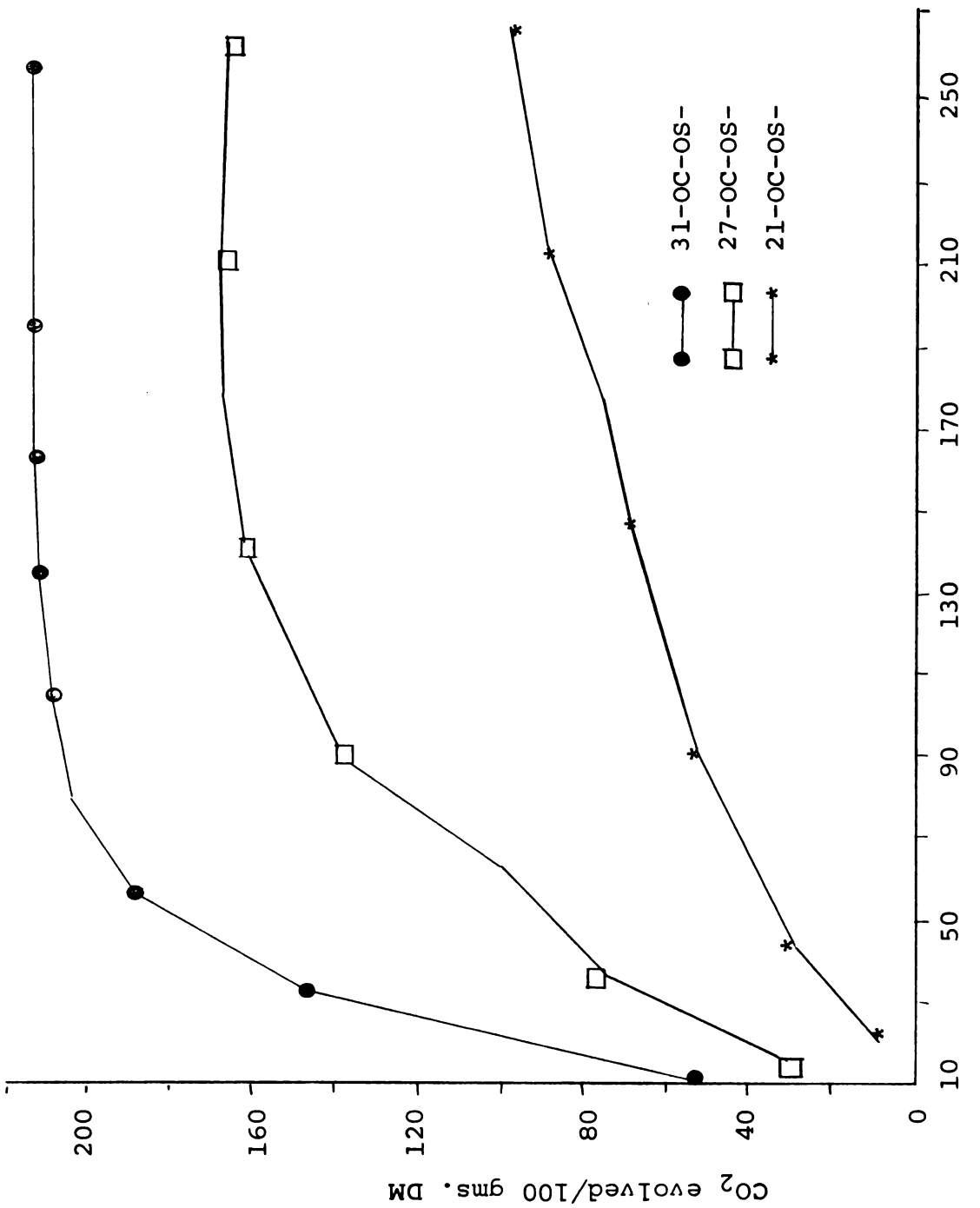

Fig. 5a.--CO_2 production 100 gm DM at all moisture levels stored without "dehydrator" and interseed atmosphere not flushed out.

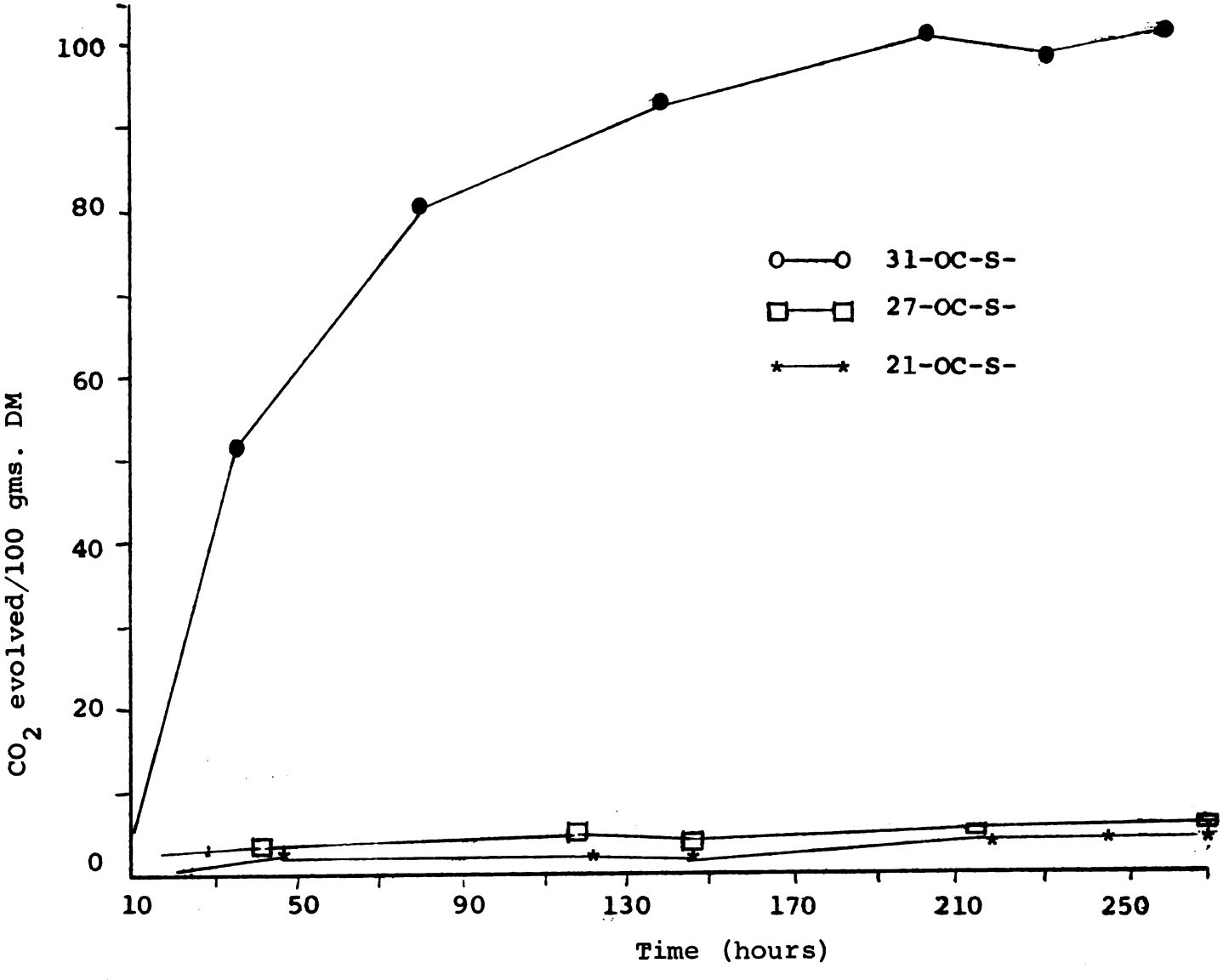

Fig. 5b.--CO_2 production/ 100 gm. DM at all moisture levels stored with "dehydrator" and interseed atmosphere not flushed out.

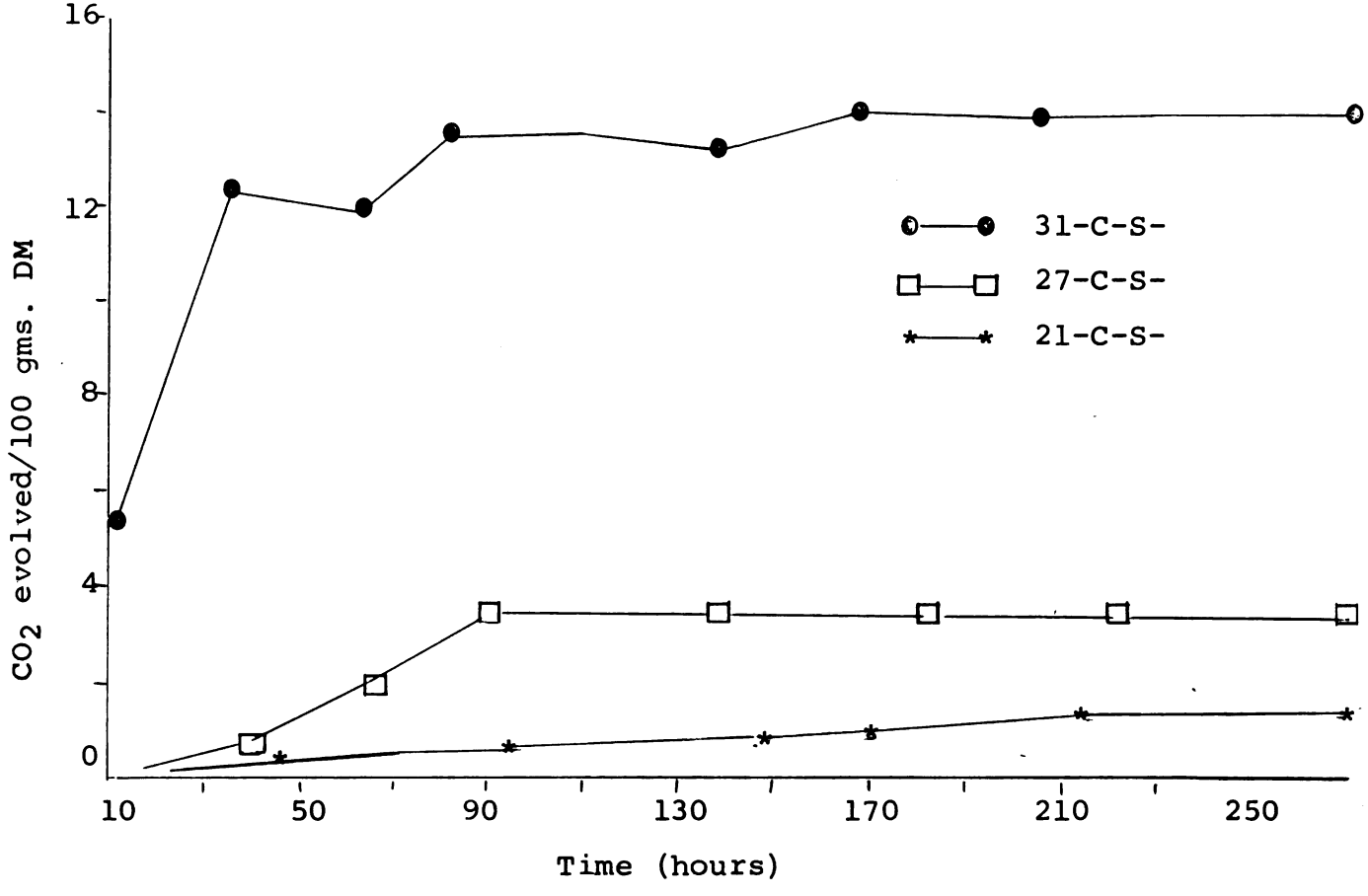

Fig. 5c.--CO_2 production/100 gm. DM at all moisture levels stored with "dehydrator" and interseed atmosphere flushed out.

5.7 Viability Test

Germination is a sensitive index of the soundness of grains. The embryo was regarded by Bailey and Gurjar (5) as the seat of respiration because it is richer in enzymes than the endosperm. Cereals that are viable are fit for consumption if they are clean. This is the consensus.

The results of the germination test are plotted in Figure 6. No samples stored without the "dehydrator" germinated excepting sample 21- C-OS-2- which had 43%. Samples stored with the "dehydrator" at 21% m.c. (21- OC-S-) had 96.67% and 68.33% at the end of 2 and 12 weeks respectively and sample 21- C-S- had 96.67% and 56.67% during the same period. Figure 6 also shows that the decline of the former treatments was more gradual than the latter. Flushing out the interseed gas seemed to have increased viability slightly. In all samples, germination declined with increasing moisture level and time. The effect of moisture was more than that of time (Table 3).

Although germination was low in samples stored with the "dehydrator" at 27% and 31%, yet the grains felt hard and firm. There were no signs of decay. Secondary dormancy was suspected. Results of a trial showed that they were not

dormant. The loss of viability could have been due to injury resulting from NaCl solution, since the moisture content began to increase after 2 weeks (Figs. 5a, 5b, 5c). This could be avoided by changing the "dehydrator" after a few days.

Table 2 shows the result of grains stored with "dehydrators" "a" and "b." The least germination was 80%, highest, 100%. This practice of changing the "dehydrator" was very effective in preserving the grains for germination.

In another experiment in which corn at 29.02% m.c. was mixed with the "dehydrator" in the ratio of 1 part of "dehydrator" to 2 parts of corn by weight, no grain germinated at the end of 3 months. The final moisture content was 18.64%. They were clean and free flowing. Increasing the quantity of the "dehydrator" did not improve germination. By changing the damp "dehydrator" after 2 or 3 days, most of the water, and salt solution may be removed and germination injury from this source eliminated. The results in Table 4 show that the "dehydrator" should be changed after 2 or 3 days.

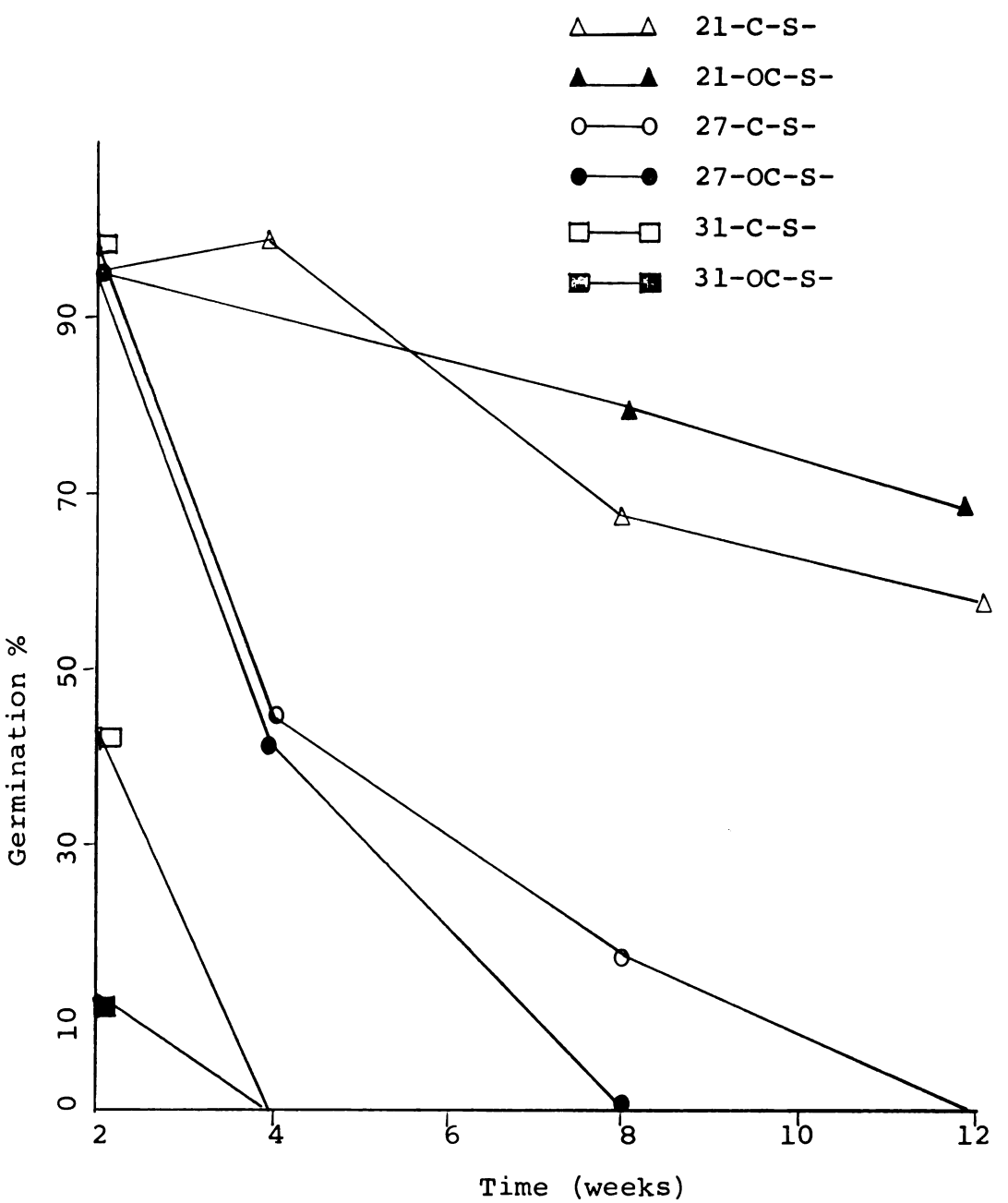

Fig. 6.--Germination results for all treatments stored with the "dehydrator" with and without CO_2.

Table 3.--Analysis of Variance; Measured Variable: Germination of all samples (48) stored with the "dehydrator."

Source of Variation	d.f.	Mean Square	Fratio	FO.99
A	1	0.011	4.59	7.82
B	2	2.251	917.48**	5.61
AB	2	0.016	6.48*	5.61
C	3	0.683	278.48**	4.72
AC	3	0.011	4.34*	4.72
BC	6	0.143	58.20*	3.67
ABC	6	0.015	6.01*	3.67
Error	24	0.002		
Total	47			

** Highly significant

A = CO_2

B = Moisture

C = Time

Table 4.--Effects of changing the "dehydrator" on final moisture content and germination

"Dehydrator" changed at time indicated*			At the end of 250 hrs.	
Time (Hrs)	Moisture Content %	Germination %	Moisture Content %	Germination %
0	26.60	96.67	-	-
6	18.64	100.00	15.18	98.33
12	17.80	100.00	14.90	98.33
24	16.99	96.67	14.47	95.00
48	14.95	100.00	13.56	96.67
80	17.88	95.00	15.02	91.67
120	19.01	90.00	15.56	86.67
210	19.46	83.33	16.06	83.33
240	19.74	80.00	17.18	75.00
250	-	-	19.93	78.33

*Each treatment had 2 samples. The "dehydrator" was screened off from both at the time indicated (Column 1). Moisture content and viability were determined on one sample. The other sample was stored with fresh "dehydrator" until the end of 250 hours when moisture content and viability were determined.

5.8 Edibility

All samples stored without the "dehydrator" (21-OC-OS-) at 21% m.c. rated very good. Samples stored with the "dehydrator" at 21% m.c. rated just fair (3.25). A "t" test showed that the difference was not significant. The edible quality rated higher when the interseed gas was flushed out or when the volume of CO_2 produced was high and the rate, fast. All samples stored without the "dehydrator" had characteristic silage smell. This smell lessened with soaking. In parts of Nigeria, eko (Yoruba), with a bit of fermented taste is preferred to that which is bland. Quite often the water that remains after the starch has settled down may be saved. This water is then used in preparing the eko because it improves the flavor.

6. SUMMARY AND CONCLUSIONS

6.1 Summary

A new, cheap, simple and easy method for preserving high moisture corn on a small scale has been found. The method involves the mixture of sawdust impregnated with sodium chloride with the grain.

The objective of this experiment was to investigate the feasibility of storing shelled high moisture corn in closed containers for use as food and seed. Three moisture levels 21.4%, 27%, and 30.8% m.c. were used. The experiment was conducted in the laboratory for 12 weeks in Spring when the temperatures approximated those predominant in the tropical and subtropical countries.

6.2 Conclusions

Sawdust-salt mixtures prepared as described above were effective in reducing the moisture content of shelled high moisture corn.

Thorough mixing of the grain with the "dehydrator" is highly important. The rate of drying depends upon the rate of moisture diffusion from the grain to the "dehydrator."

Mixing high moisture corn up to 21% in the ratio of 3:1 of corn to "dehydrator" by weight is recommended. Removing the damp "dehydrator" and replacing it with a dry one after two or three days is recommended for longer storage or damper grains.

Although the 1-inch cube wood absorbs more moisture than the sawdust, the latter is recommended for use because it might be cheaper and easier to obtain and is certainly simpler to mix and redry.

Once the "dehydrator" is prepared, it is "everlasting." All that is needed when it is too wet is to redry it. If NaCl is used, drying at atmospheric relative humidity below 75% is possible, but not with $CaCl_2$.

High moisture shelled corn can be stored with the "dehydrator" for food. The container should be airtight and completely filled in order to reduce the amount of trapped oxygen available for mold growth.

With high moisture corn, flushing out the interseed atmosphere might not be necessary since the volume of CO_2 produced is enough.

The evolution of gases from high moisture corn practically ceases after 2 weeks. Containers can be sealed and stored away without fear of pressure building up later to break the seal.

Flushing out the interseed atmosphere reduced dry matter loss. The advantage arising from this practice did not justify the expenses. Proper mixing and packing could compensate for the effect of flushing.

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