THE EFFECT OF EXCESSIVE FUMIGATION ON WHEAT QUALITY

by

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A. B., Kansas State Teachers College of Pittsburg, 1941

A THESIS

submitted in partial fulfillment of the

requirements for the degree of

MASTER OF SCIENCE

Department of Milling Industry

KANSAS STATE COLLEGE
OF AGRICULTURE AND APPLIED SCIENCE

1946

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INTRODUCTION

The storage and transportation of damp wheat presents a very difficult problem, especially in years when there have been protracted rainfalls during the latter part of the harvest season. The farm and country elevators do not have the facilities for drying wheat and it must be shipped as soon as possible to the large terminal elevators where it can be dried to a suitable moisture content for storage. In years when there is much damp wheat, heavy losses result from heating in farm bins, in country elevators, and in transit. When wheat has been damaged by heating, or "bin burned", it is fit only to be used as feed. It cannot be blended into the milling wheat even in small amounts since the persistent moldy odor is carried over into the flour.

In normal seasons the small quantity of damp wheat which reaches the terminal elevator can be handled easily by drying or mixing with dry wheat. In the latter case the translocation of moisture takes place rapidly and, if properly handled, there is little danger of heating.

A new factor was introduced with the advent of the combine harvester, as the operators had a tendency to cut the grain too early. Combine harvested wheat tends to be higher in moisture than that resulting from the other less direct methods. (Larmour, Geddes and Cameron, 1933). The concept has developed that combined wheat tends to heat more readily than wheat thrashed by other methods. The theory has been advanced that combined wheat has not had a chance to "sweat" before harvesting and undergoes this in the bin or car thus providing conditions favorable for heating. In the other methods of harvesting the "sweating" occurs in the shock where the aeration and small bulk preclude any danger of heating. Larmour, Clayton, and Wrenshall (1935, p. 628) state:

> It seems probable that in the course of dessication of wheat during the latter part of the ripening process there is a stage at which there occurs a redistribution of bound and free water in the grain, accompanied by a syneretic effect which makes the grain feel damp even though there has been no absolute change in the moisture content.

The respiration and heating of cereal grains has attracted the attention of many workers due to the practical significance of the relations of moisture, heating and the keeping qualities of stored grain.

The role of the microflora in the heating of stored organic material was studied as early as 1907 by Miehe, who observed their presence in heating hay. While the heat produced in stored grain is undoubtedly due both to the respiration of the embryo and to the growth of microorganisms, several workers seem to favor the suggestion that the microorganisms are mainly responsible for the high temperatures associated with bin burning. To the present time there has not been reported any very successful segregation of these two factors, the difficulty being to sterilize the grain without injury to the embryo. Larmour, Clayton and Wrenshall (1935), however, did have partial success in sterilizing wheat by passing vapors of carbon tetrachloride through the material.

The general concensus has been that wheat which would not germinate would neither have the proper milling qualities nor would bake good bread. Generally these conclusions were based on experience with wheat that had suffered severe damage during storage.

A cooperative experiment between the Department of Milling Industry, Kansas State College, and the United States Department of Agriculture, Bureau of Entomology and Plant Quarantine, showed that wheat which had been killed by fumigation could be milled and made into good bread. This lead to the investigation of the effects of excessive fumigation on wheat quality. If wheat coming into storage with excessive moisture could be fumigated in such a manner as to reduce the microfloral count and lower the rate of respiration without damaging quality, it would be possible to save much grain which otherwise would be unfit for milling. The purpose of the research reported in this thesis was to investigate such possibilities.

REVIEW OF LITERATURE

Respiration is responsible for the heat energy released in a mass of damp grain (Bailey and Gurjar, 1919). Assuming that dextrose is the respiratory substrate, the gross chemical aspects of the process (Meyers and Anderson, 1939) may be represented as follows:

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

The water produced as a result of respiration, known as metabolic water, becomes a part of the general mass of water present in the respiring cells. The gaseous exchanges associated with respiration were studied long before any particular significance was attached to them. The observed results of respiration usually are the consumption of oxygen and the simultaneous release of carbon dioxide. More specifically, respiration is an oxidation process leading to the release of energy. Meyers and Anderson (1939, p. 510) state:

> However, gaseous exchanges of the usual type are not invariably accompaniments of the process of respiration. Furthermore, plants never "breathe" in any fundamentally acceptable sense of the word, frequent popular and semipopular statements to the contrary notwithstanding. Gases pass into or out of plant organs by diffusion through the stomates, lenticels, or directly through the peripheral walls of the epidermal cells. Within the plant body gases may be distributed by diffusion through the intracellular spaces or by cell to cell diffusion as solutes. For this reason plant physiologists use the term respiration primarily to refer to the oxidation of foods in the living cells resulting in the release of energy.

The Relationship of Moisture to Respiration Rate. The moisture content is recognized as the most critical of all factors influencing the respiration, heating and deterioration of stored grain. When dry grain is moistened there is a slight increase in the rate of respiration until the critical moisture level is reached; above this level a rapid acceleration in respiratory rate occurs. This has been demonstrated by Bailey and Gurjar (1919) with wheat, and by Bakke and Noecker (1933) who, in studying the relation of moisture and heating in stored oats, concluded that heating is neglible when the moisture content is below 15 percent. Swanson and Fenton (1933) reported that when enough moisture is present the temperature is raised and this in turn stimulates the rate of respiration. Larmour, Clayton and Wrenshall (1935) noted that this increase occurred in their studies of the heating of damp wheat, as did Bailey (1940) with wheat, rye, flaxseed, and other cereal grains. Leach (1944) observed that wheat kept at 25°C in an atmosphere of relative humidities between 92 and 100 percent showed a continuously accelerating rate of carbon dioxide output, and Ramstad and Geddes (1942), working with soy beans, found an increasing rate of respiration with increasing moisture content but did not get the sharp break above the 14 percent level reported by Bailey and Gurjar (1919). Milner and Geddes (1946), in their work on soy beans, confirmed the work of Bailey and Gurjar (1919) by observing a very marked increase in the respiratory rate in soy beans containing 14.6 percent moisture.

At the point where there is a marked increase in the rate of respiration there is a marked acceleration in the physiological and biochemical processes which make the grain more susceptible to storage injury. This point is referred to as the critical moisture level of the grain. In the literature there have been only a few studies devoted to this question. Kretovich (1945) divides all such works into two groups: (1) works based on determination of the energy of the grain respiration, and (2) works based on a quantitative study of enzyme activity in the grain. He states:

> The first method, the study of the energy of respiration, is more reliable, since a study of respiration informs us about the whole complex of biochemical processes occurring in the grains, and not one step or even one reaction of the whole complex of biochemical transformations which characterize the changes of substances in dormant grain. We must also note that the study of grain respiration gives us information not only on the gaseous exchanges of the tissues of the grain itself, but also of the microorganisms on the grain surface. Thus in the quantitative determination of the energy of respiration, we must consider this as a result of the whole sum of biochemical changes in the grain mass produced both from enzymatic properties of the grain and from those of its epiphitic flora, which at last, from the practical point of view, are particularly important.

Effect of Microflora. The role of microorganisms in heating of stored organic material was observed as early as 1907 by Miche, who noted their presence in heating hay. Isachenko et al. (1934) showed that the microorganisms had much to do with the rise in temperature of stored seeds. Other work done by James, Rettger, and Thom (1928), Wilson (1928), Bakke and Noecker (1933), Swanson and Fenton (1933), Larmour et al. (1935), Thomas (1937), Ramstad and Geddes (1942), Milner and Geddes (1945), Tervet (1945), Kretovich (1945), and Milner and Geddes (1946), has shown definitely that heating of damp grain is usually accompanied by the growth of microorganisms. While heat generated in stored damp grain is due to the respiration of both the embryo and the microflora, many of the aforementioned workers are of the opinion that the microflora are mainly responsible for the high temperatures which result in damage. This may also be an important factor in causing the sharp increase in the rate of respiration with samples of grain having a moisture content of 14 percent or higher.

The above discussion has dealt only with the respiration effects related to the grain itself and the contaminating microflora. It should be mentioned that other factors may influence the heating of stored grain. For example, researches such as that reported by Lindgren (1935) indicate that at least a part of the heat evolving in grain infested with insects is caused by the activity of these insects.

The Effect of Various Chemical Substances. Studies have been made of the specific influence of chemical compounds upon the rate of respiration. Irving (1911) noted that small doses of chloroform when brought in contact with the leaves of the cherry laurel (*Prunus laurocerasus*) caused an increase in the rate of respiration which persisted as long as the leaves remained in contact with the chloroform. Larger doses resulted in a temporary increase followed by a decrease to much below the initial rate. In general other compounds of this type have much the same effect.

Miege (1935) in studying the effects of insecticide treatments on the baking value of wheat flour divided the compounds into three classes: (1) those exerting no harmful effect, (2) those which were definitely harmful, and (3) those which had a beneficial effect. Smith and Staten (1942) studied the effects of carbon disulfide upon wheat, concluded that this compound seriously affected both the germination and baking quality. Cotton et al. (1946) in a study of the loss of viability of wheat due to fumigation, and its relation to baking quality states:

> ....that with excessive dosages or exposure period fumigants commonly used to treat wheat may cause injury to germination, particularly if the grain moisture is above 12.5 percent. At ordinary dosages and exposures these fumigants temporarily effect the baking quality of flour made from the wheat but the effects disappear if the grain is sufficiently aerated. Although loss of viability from many causes may impair baking quality, these studies indicate that partial or complete loss of viability due to fumigation does not necessarily impair baking quality even after storage for 12 months.

METHODS AND MATERIALS

The wheat samples selected for this study were obtained from commercial grain elevators and flour mills in order to have wheat which had been exposed to the contaminating atmospheres of the grain car and storage elevator. One sample was a No. 1 grade hard red winter wheat from Kansas, the other a soft red winter No. 1 grade from Indiana. Both hard and soft wheats were used to determine differences in their reaction to excessive fumigation.

Fumigation. Four fumigants or mixtures of fumigants were used in this study, as follows:

		Percent
1.	Ethylene dibromide ------	15
	Carbon tetrachloride ----	85
2.	Ethylene dichloride -----	75
	Carbon tetrachloride ----	25

		Percent
3.	Ethylene dibromide ------	05
	Ethylene dichloride -----	25
	Carbon disulfide --------	10
	Carbon tetrachloride ----	60

4. Methyl bromide

The special mixture No. 3 was compounded by Dr. R. T. Cotton[1] for use in farm bins. This mixture enables the fumigator to get a complete kill in the top, middle, and bottom of the bins. The ethylene dibromide mixture is now patented[2] and is being used as a spot fumigant for flour mills. According to Cotton et al. (1946), ethylene dichloride is a very good liquid fumigant which does not injure the germinating powers of the seed at twice the normal dosage.

Samples of 1,000 grams each were adjusted to three moisture levels, 12, 14 and 16 percent. It was thought unnecessary to carry the moisture levels any higher than 16 percent as wheat rarely is placed in the bins above this level. After the tempering water had been added and sufficient time allowed (overnight) to enable it to become distributed uniformly, the fumigant was then added by means of a measuring pipette, graduated in hundredths. Three fumigation levels were used, 10, 20, and 30 gallons per 1,000 bushels. Except in the case of methyl bromide, using a dosage of 10, 20, and 30 pounds per 1,000 cubic feet, all fumigations were carried out in a laboratory scale fumigator, at room temperature with an exposure period of 24 hours.

[1] Senior Entomologist, U. S. Department of Agriculture, Bureau of Entomology and Plant Quarantine, Manhattan, Kansas.

[2] F. W. Fletcher and Eugene Kenaga, U. S. Patent No. 2,391,474. January 1, 1946.

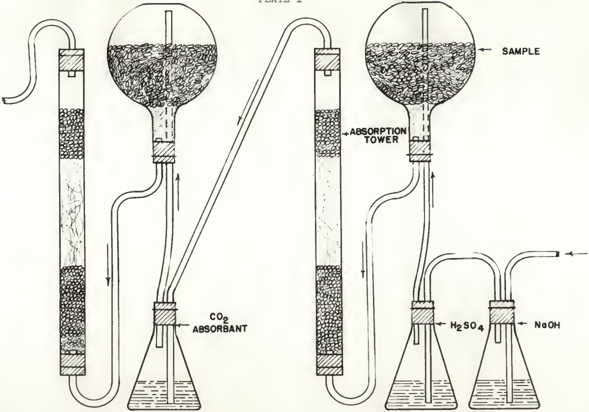
Respiration. One method has been used by many workers to determine the effect of various treatments and environmental conditions on wheat and other cereal grains is the rate of respiration of the grain, e.g. Bailey and Gurjar (1919), Larmour et al. (1935), Bailey (1940), Ramstad and Geddes (1942), and Milner and Geddes (1945)(1946). The general method as used by Bailey (1940) was followed in the present work, with modifications made necessary due to the lack of adequate control equipment.

A 250-gram sample of wheat was placed in an inverted 1,000 ml. florence flask. A stream of air was washed with a solution of concentrated sodium hydroxide to remove carbon dioxide and then passed through a solution of sulfuric acid. In order to avoid moisture changes in the wheat during aspiration, the sulfuric acid solution was of the proper specific gravity to bring the escaping air to a relative humidity in equilibrium with the hygroscopic moisture of the grain. The data for adjusting atmospheres to the proper relative humidity were taken from papers by Wilson (1921) and Coleman et al. (1925). The carbon dioxide-free, humidified air was passed into the top of the inverted flask, thus continuously washing the grain free of carbon dioxide. The air containing the respired carbon dioxide was bubbled through an absorption tower containing a saturated solution of barium hydroxide (32 grams barium hydroxide per liter). The air from the absorption tower was again washed with sodium hydroxide and then passed through a duplicate sample of tempered and respiring wheat. A series of three duplicate respirators was set up as depicted in Plate I. The air was pulled through the entire series by means of a water aspirator. The air bubbles entering the absorption tower were broken and dispersed by passing the air stream through a fine wire mesh and through glass beads and glass wool. Since no accurate equipment was available for regulating the volume of gas passing through the sample, an effort was made to have the air pass through slowly and yet have the entire chain aerated simultaneously. Each new series of samples was so handled as to reproduce the previous condition as closely as possible. The temperature in the room containing the respiration apparatus could not be controlled with any accuracy. With the exception of a few of the soft wheats, the average temperature during the determination of the respiration rates was 30°C, with a maximum of 33°C and a minimum of 26°C.

The tempered samples were allowed to stand for a period of three days before aeration was started (Larmour et al., 1935) and were then aerated continuously for a period of seven days, a total of ten days from the time of tempering. At the end of the aeration period, the absorption towers were removed and washed with distilled water until the washings were neutral to phenolphthalein indicator. The excess barium hydroxide was titrated with a solution of 1.0 N hydrochloric acid. The results were calculated to give the number of milligrams of carbon dioxide per 100 grams of wheat per 24 hours.

Germination. Both before and after fumigation, all samples were tested for germination by the technique as described

PLATE I



in the Federal Seed Act of August 9, 1939, (1946)[1].

Microfloral Study. To study the total influence of fumigation upon wheat quality, it was necessary to make an investigation of its effect upon the microflora. Many authors are of the opinion that a large portion of the carbon dioxide evolved from damp stored cereal grains is due to the microfloral activity.

Following the technique described by Kent-Jones and Amos (1930), 10-gram samples of wheat were washed with 90 ml. of a 0.5 percent solution of sodium chloride containing about three grams of an inert, acid-washed sand. This solution was then plated in duplicate on nutrient agar, using the pour plate method at dilutions of 1:1,000 and 1:10,000. Preliminary studies showed that at higher concentrations the growth was too profuse to count. The bacterial count was made after an incubation period of 48 hours at 30°C. A second series of counts was made on the same samples to determine the extent of fungal contamination. These were plated on a rose bengal medium according to the procedure recommended by Smith and Dawson (1944) and incubated for five days at 30°C. The rose bengal concentration of one part per 1,500 is reported by these authors to be inhibitory to the bacteria but not to the fungi.

Milling and Baking. Samples of 1,000 grams each were milled on a Buehler experimental mill. The moisture was determined on each sample and tempered for milling to 16 percent for the hard wheat samples and 13.5 percent for the soft wheat

[1] The germination tests for this study were made in the Kansas State Seed Laboratory, Manhattan, Kansas.

samples. The flour obtained was of 68 to 72 percent extraction. Each sample required approximately 20 minutes to mill, allowing seven minutes of that time to clean out the mill between samples. The resulting flours were baked in an experimental baking laboratory, using 100 grams doughs and a malt-phosphate-bromate formula as follows:

	Percent
Flour	100
Sugar	5
Salt	1
Malt	0.3
$NH_4H_2PO_4$	0.1
Yeast	3
Water	As required

EXPERIMENTAL RESULTS

Hard Red Winter Wheats

Fumigation. The fumigants applied to the damp wheats had a varying effect upon the odor of the wheat at the time of milling as will be noted from Table 1. These odors occurring in the wheat prior to milling did not always carry over into the flour and into the baked bread. When an odor did persist in the flour, it was one of two types, a musty odor which was not present in the baked product, or a typical methyl bromide odor which was quite noticeable in the bread to one accustomed to the odor, but disappeared after a few hours' aeration.

Respiration. The effect of excessive fumigation upon the rate of respiration is quite marked, especially at the higher moisture levels and at higher concentrations of the fumigant. In untreated wheat that had been tempered to 12, 14, and 16

Table 1. The effect of various fumigants on the odor of wheat prior to milling.

Treatment		12 percent	14 percent	16 percent
*Ethylene dibromide,	10 gal.	No odor	Slight odor	Slightly musty
*Ethylene dichloride,	10 gal.	Slightly musty	Slight odor	Fruity odor
*Special mixture,	10 gal.	Odor of CS_2	No odor	Slightly sour
**Methyl bromide,	10 lbs.	Slight CH_3Br odor	Odor of CH_3Br	Odor of CH_3Br
Ethylene dibromide,	20 gal.	No odor	Fruity odor	Fruity odor
Ethylene dichloride,	20 gal.	No odor	No odor	No odor
Special mixture,	20 gal.	Slight odor of CCl_4	Slight odor of CCl_4	Slight odor of CCl_4
Methyl bromide,	20 lbs.	Slight odor of CH_3Br	Medium odor of CH_3Br	Heavy odor of CH_3Br
Ethylene dibromide,	30 gal.	Slight fruity odor	Odor of ethylene dibromide	Odor of ethylene dibromide
Ethylene dichloride,	30 gal.	Strong odor of CCl_4	Strong odor of CCl_4	Strong odor of CCl_4
Special mixture,	30 gal.	Odor of CCl_4	Strong odor of CCl_4	Very strong odor of CCl_4
Methyl bromide,	30 lbs.	Strong odor of CH_3Br	Very strong odor of CH_3Br	Very, very strong odor of CH_3Br

* Gallons per 1,000 bushels.
** Pounds per 1,000 cubic feet.

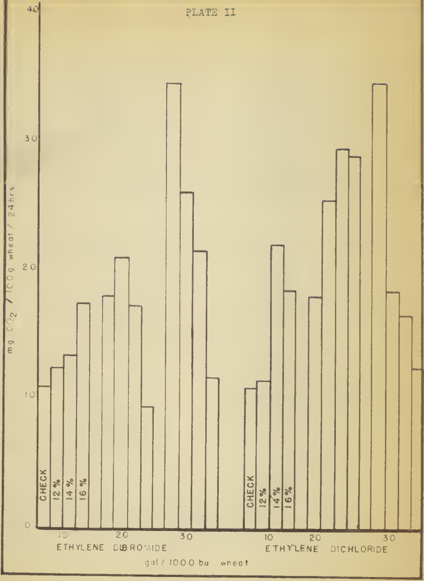
percent moisture levels, the rate of respiration increased gradually until the 14 percent moisture level was reached. However, when the moisture of the sample was above 14 percent there was a very sharp increase in the amount of carbon dioxide produced as will be noted in Plate II.

The effect of ethylene dibromide-carbon tetrachloride mixture will be observed in Plate II. It is apparent that with the smaller dosage, 10 gallons per 1,000 bushels, the effect was not as great as at the higher concentration. In fact, the general shape of the graph is not unlike the normal, untreated sample, except that at the 16 percent moisture level there was not the great increase in carbon dioxide production. The fumigant apparently exerted some retarding action upon the respiration rate. However, it should be noted that when the dosage of the fumigant was increased to 20 and 30 gallons per 1,000 bushels, the effect was quite marked. The initial rate of respiration was much higher at the 12 percent level than with the check sample. This confirms observations of Irving (1911), who showed that the leaves of the cherry laurel (<u>Prunus laurocerasus</u>), when brought in contact with small doses of chloroform increased in their rate of respiration as long as they remained in contact with the chloroform. Following the initial increase in the amount of carbon dioxide produced, a marked decrease to much below that of the normal rate was observed. This marked decrease was characteristic of all samples fumigated with ethylene dibromide at the 20 and 30 gallons per 1,000 bushel rates. The samples treated at the 20 gallon rate made a slight

EXPLANATION OF PLATE II

The effect of ethylene dibromide and ethylene dichloride on the amount of carbon dioxide produced.

PLATE II



40
30
20
10
0
mg CO2 / 100g wheat / 24 hrs
CHECK
12 %
14 %
16 %
CHECK
12%
14%
16%
10
20
30
ETHYLENE DIBROMIDE
10
20
30
ETHYLENE DICHLORIDE
gal / 1000 bu wheat

increase between the 12 and 14 percent moisture level, followed by the characteristic decrease at the 16 percent moisture level. This increase was not observed at the 30 gallon rate but a sharp decrease at all moisture levels.

Ethylene dichloride did not effect as great a change in the rate of respiration as did ethylene dibromide. At both the 10 and 20 gallon rates increased carbon dioxide production was noted between the 12 and 14 percent moisture levels followed by a slight decrease in the rate but not to a value below the initial rate of respiration at the 12 percent level (Plate II). However, at the highest concentration of the fumigant there was a steadily decreasing rate of respiration with increasing moisture.

Fumigation with the "special mixture", the composition of which has been given previously, resulted in an initial stimulation especially at the 14 percent moisture level, followed by a very marked decrease in the rate of respiration (Plate III).

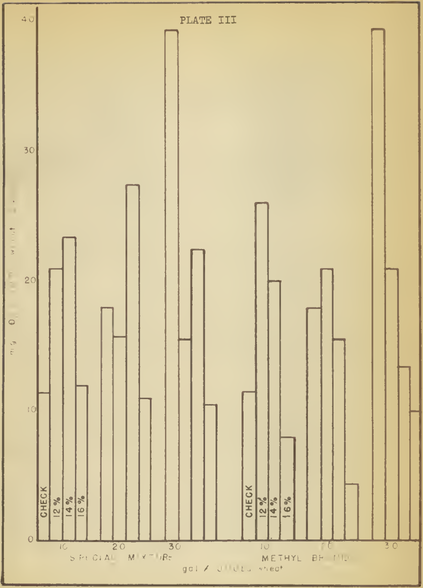
The three fumigants described above all had a similar effect upon the respiration rate. The effect of methyl bromide, however, was quite different; in no case was the rate higher than the initial rate at the 12 percent moisture level. There was an initial stimulation caused by the fumigant, but in all cases this was followed by a very marked decrease in the rate of respiration, as will be noted in Plate III.

Germination. The effects on wheat germination of the four fumigants used in this study are shown in Plates IV and V. It will be noted that the effects of ethylene dibromide and ethylene

EXPLANATION OF PLATE III

The effect of the Special Mixture and methyl bromide on the amount of carbon dioxide produced.

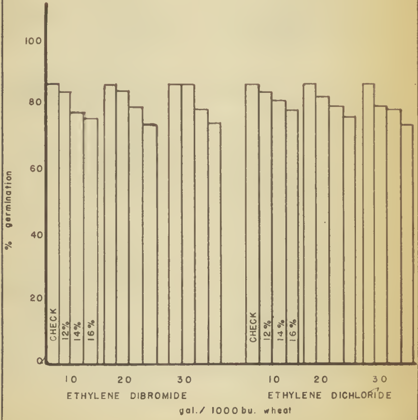
PLATE III



EXPLANATION OF PLATE IV

The effect of ethylene dibromide and ethylene dichloride on the viability of hard red winter wheat.

PLATE IV

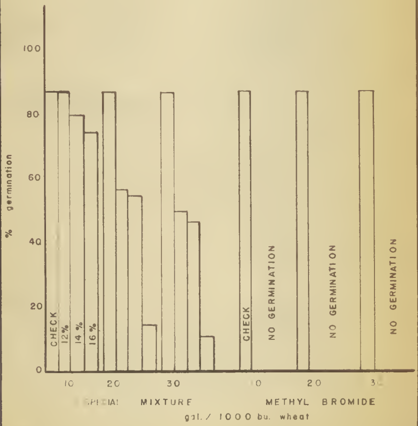


100
80
60
40
20
0
% germination
CHECK
12%
14%
16%
CHECK
12%
14%
16%
10
20
30
10
20
30
ETHYLENE DIBROMIDE
ETHYLENE DICHLORIDE
gal./ 1000 bu. wheat

EXPLANATION OF PLATE V

The effect of Special Mixture and methyl bromide on the viability of hard red winter wheats.

PLATE V


100
80
60
40
20
0
% germination
CHECK
12%
14 %
16 %
CHECK
NO GERMINATION
NO GERMINATION
NO GERMINATION
10
20
30
20
MIXTURE
METHYL BROMIDE
gal. / 1000 bu. wheat

dichloride, Plate IV, upon germination were relatively small, considering the excessive dosages used. As the moisture of the wheat was increased, the number of wheat kernels which germinated decreased. This probably was due to the fact that the outer bran coat of the wheat was made more permeable by the addition of water.

The special mixture, Plate V, very seriously impaired the germination, in this instance probably due to the presence of carbon disulfide in the mixture. The percentage of viability was inversely proportional to the percentage of the moisture. The samples at the 16 percent moisture level and at the highest concentration of fumigant germinated only to the extent of 11 percent, as compared to the 87 percent germination for the control. Cotton et al. (1946) observed that carbon disulfide lowered the viability of wheat fumigated at 10.5, 12.5 and 14.0 percent moisture when stored over a period of 12 months.

Of all the fumigants used, methyl bromide had the greatest effect upon the viability of the wheat (Plate V). At all moisture levels and all concentrations of the fumigant, samples treated with methyl bromide failed to germinate. According to Shepard, Lindgren, and Thomas (1937), methyl bromide is somewhat more toxic to the granary weevil (Sitophilus granarius L.) than hydrocyanic acid. Ratcliffe, Gay, and Fitzgerald (1940) assess the relative toxicity to some of the common fumigants to the granary weevil, as follows: carbon disulfide rated as unity, hydrogen cyanide, 6, ethylene oxide, 3-5.5, ethylene dichloride, one-fourth to one-third, methyl bromide, 7. Carbon tetrachloride, used in mixtures with certain of these fumigants to reduce fire hazards, has a toxicity of about one twelfth that of carbon disulfide. According to the above rating of toxicity given methyl bromide, it is not surprising to observe the drastic effect of this compound upon the germ plasm of the wheat kernel.

Milling and Baking. All samples for this study were milled on a Buehler mill to approximately 70 percent extraction. Due to the small size of the sample and the type of the machine used for milling, it was impossible to observe any differences in the milling quality of the wheat that might have been attributed to the fumigants. All samples milled well, and the only abnormalities observed during the process were the odors of some of the fumigated wheats. The flour samples were baked, using 100-gram experimental loaves and the formula given on page 14. Absorption was determined with the Brabender Farinograph. The absorption as determined for the control sample was used for all flours, since the only difference in these flours was the fumigation treatment. The flour weighed up for baking on a 14 percent moisture basis. Mixing time was determined on the National Swanson-Working Recording Micro-mixer. The mixing time as determined for the control sample was used for all samples. From the handling characteristics of the dough, it was observed that increased dosage of any one fumigant slightly lowered the absorption and decreased the optimum mixing time. Samples which had been fumigated at a high rate, had more mellow handling characteristics, and felt slack during the punching and moulding process. Table 2 gives the complete data relative to loaf volume, crumb color, and texture. A general tendency for the loaf volume and the crumb characteristics to decrease in value with increased moisture and with increased concentration of the fumigant will be observed in Table 2. The crumb color had a tendency to become slightly darker with the high rate of fumigation; in the case of those samples fumigated with methyl bromide at the rate of 30 pounds per 1,000 cubic feet the crumb color became quite gray. The doughs of these samples were sticky and slack, and quite hard to handle.

Microflora: Bacteria. The four fumigants used in this work in most cases had a very definite bacteriacidal action at each rate of fumigation and at all moisture levels. The data presented in Tables 3, 4, 5, and 6 give the actual number of colonies found on each of the nutrient agar plates after 48 hours incubation at 30°C. In Table 4, showing the effect of ethylene dichloride on the bacterial growth, it will be noted that at the 20 gallon per 1,000 bushels fumigation rate and at the 16 percent moisture level, the colony count exceeds that of the control sample. At the 20 pounds per 1,000 cubic feet dosage and 16 percent moisture level, methyl bromide treated samples also showed a greater number of colonies than did the control (Table 6).

Microflora: Fungi. No fungal growth was found with the control (nonfumigated) sample at either the 12 or 14 percent moisture levels, thus rendering comparison at these levels impossible. Accordingly, only the data for the 16 percent

Table 2. Baking data for hard red winter wheat floure.

Treatment		Flour protein percent	Mixing time min.	Absorption on 14% M.B. percent	Loaf volume cc.	Crumb* color percent	Crumb' grain & texture	Odor
Control sample		11.5	2.25 1/	63.5 1/	750	85cy	85-o	---
**12% Ethylene dibromide,	10 gal.	11.5			730	84y	80-o	---
14% Ethylene dibromide,	10 gal.	11.4			730	82y	80-o	---
16% Ethylene dibromide,	10 gal.	11.3			625	80y	78-o	---
**12% Ethylene dichloride,	10 gal.	11.5			738	84cy	80-o	None
14% Ethylene dichloride,	10 gal.	11.5			730	84cy	80-o	---
16% Ethylene dichloride,	10 gal.	11.6			683	84cy	80-o	---
**12% Special mixture,	10 gal.	11.5			684	84cy	80-o	Slight
14% Special mixture,	10 gal.	11.7			705	84cy	80-o	Slight
16% Special mixture,	10 gal.	11.4			710	85cy	80-o	Slight
***12% Methyl bromide,	10 lbs.	11.0			693	84cy	82-o	Slight
14% Methyl bromide,	10 lbs.	11.3			703	84cy	82-o	Slight
16% Methyl bromide,	10 lbs.	11.4			615	84cy	82-o	Slight
12% Ethylene dibromide,	20 gal.	11.0			745	85cy	82-o	None
14% Ethylene dibromide,	20 gal.	11.0			600	84cy	80-o	None
16% Ethylene dibromide,	20 gal.	11.2			550	84y	80-o	None
12% Ethylene dichloride,	20 gal.	11.6			798	85cy	82-o	None
14% Ethylene dichloride,	20 gal.	11.6			785	85cy	82-o	None
16% Ethylene dichloride,	20 gal.	11.2			703	84y	82-o	None
12% Special mixture,	20 gal.	11.3			775	85cy	84-o	Slight
14% Special mixture,	20 gal.	11.3			755	85cy	84-o	Slight
16% Special mixture,	20 gal.	11.3			755	85cy	84-o	Slight

Table 2 (concl.)

Treatment		Flour protein percent	Mixing time min.	Absorption on 14% M.B. percent	Loaf volume cc.	Crumb* color percent	Crumb ' grain & texture	Odor
12% Methyl bromide,	20 lbs.	11.4	2.25 1/	63.5 1/	753	86cy	84-o	Heavy
14% Methyl bromide,	20 lbs.	11.3			780	86cy	84-o	Heavy
16% Methyl bromide,	20 lbs.	11.5			758	86cy	84-o	Heavy
12% Ethylene dibromide,	30 gal.	11.3			700	78y	80-o	Slight
14% Ethylene dibromide,	30 gal.	11.4			650	76y	80-o	Slight
16% Ethylene dibromide,	30 gal.	11.6			550	70y	76-o	Slight
12% Ethylene dichloride,	30 gal.	11.6			700	78cy	80-o	None
14% Ethylene dichloride,	30 gal.	11.2			675	76y	78-o	None
16% Ethylene dichloride,	30 gal.	11.5			600	74y	76-o	None
12% Special mixture,	30 gal.	11.4			650	78cy	80-o	Slight
14% Special mixture,	30 gal.	11.5			705	80cy	80-o	Slight
16% Special mixture,	30 gal.	11.3			710	80cy	80-o	Slight
12% Methyl bromide,	30 lbs.	11.2			695	76cg	80-o	Heavy
14% Methyl bromide,	30 lbs.	11.5			635	72cg	76-o	Heavy
16% Methyl bromide,	30 lbs.	11.0			585	70-g	70-o	Heavy

* c = creamy
y = yellow
g = gray
' o = open
** Gallons per 1,000 bushels
*** Pounds per 1,000 cubic feet
1/ Mixing time and absorption were constant for all samples.

Table 3. The effect of ethylene dibromide on bacterial growth, 1:10,000 dilution.

Dosage	Moisture levels		
	12%	14%	16%
Control	32	100	30
10 gallons	7-10	0	3-10
20 gallons	16-18	7-29	1-5
30 gallons	2-3	1-1	1-8

Table 4. The effect of ethylene dichloride on bacterial growth, 1:10,000 dilution.

Dosage	Moisture levels		
	12%	14%	16%
Control	32	100	30
10 gallons	0-0	0-1	25-261
20 gallons	1-9	57-60	20-24
30 gallons	0-2	7-7	283-301

Table 5. The effect of the special mixture on bacterial growth, 1:10,000 dilution.

Dosage	Moisture levels		
	12%	14%	16%
Control	32	100	30
10 gallons	0-2	0-1	4-31
20 gallons	23-25	2-3	0-1
30 gallons	4-8	0-2	3-4

Table 6. The effect of methyl bromide on bacterial growth, 1:10,000 dilution.

Dosage	Moisture levels		
	12%	14%	16%
Control	32	100	30
10 pounds	0-0	0-1	0-8
20 pounds	1-5	0-3	25-70
30 pounds	1-3	0-19	1-3

Table 7. The effect of fumigation on fungal growth at the 16 percent moisture level, 1:1,000 dilution.

Dosage	Treatment			
	Ethylene dibromide	Ethylene dichloride	Special mixture	Methyl bromide*
Control	37	37	37	37
10 gal./1,000 bu.	0-1	25-261	4-31	0-8
20 gal./1,000 bu.	0-1	350-400	0-3	0-1
30 gal./1,000 bu.	2-6	15-20	0-0	19-32

* The dosage for methyl bromide was pounds per 1,000 cubic feet.

moisture level can be presented. These data are found in Table 7 and indicate the definite fungicidal action of all fumigants except ethylene dichloride; this latter had no action upon the fungal contamination present in the wheat.

Soft Red Winter Wheat

Fumigation. The effect of fumigation on the soft wheat was quite similar to that found for the hard red winter wheats in that the odor of the fumigant was noticeable, especially at the high concentrations. Methyl bromide applied to the soft wheat at the rate of 30 pounds per 1,000 cubic feet produced a stronger odor than with the hard red winter wheats.

Respiration. The effect of the fumigants upon the rate of respiration of the soft wheat samples, more or less paralleled the results obtained for the hard wheat. The ethylene dibromide-carbon tetrachloride mixture caused an initial stimulation considerably greater than was observed for the hard wheats; however, the general nature of the change was the same for both the hard and soft wheats (Plate VI). Ethylene dichloride did not affect as great a change in the amount of carbon dioxide produced by the wheat as did ethylene dibromide. The highest level of fumigation resulted in a steadily-decreasing rate of respiration as the moisture was increased (Plate VI). The special mixture effected the same type of change in the respiration (Plate VII) of the soft wheat as was observed with the hard red winter wheat. The hard wheat treated with this special

EXPLANATION OF PLATE VI

The effect of ethylene dibromide and ethylene dichloride on the amount of carbon dioxide produced by soft red winter wheat.

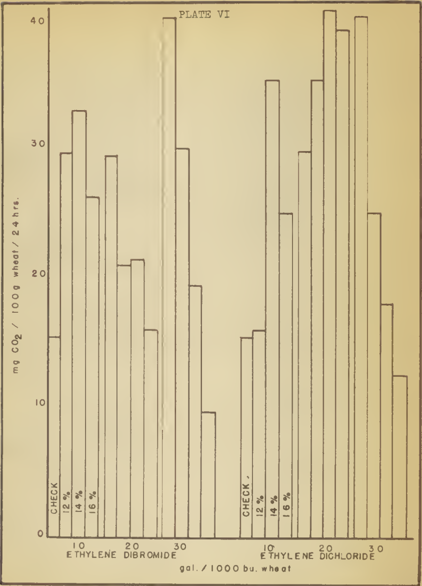
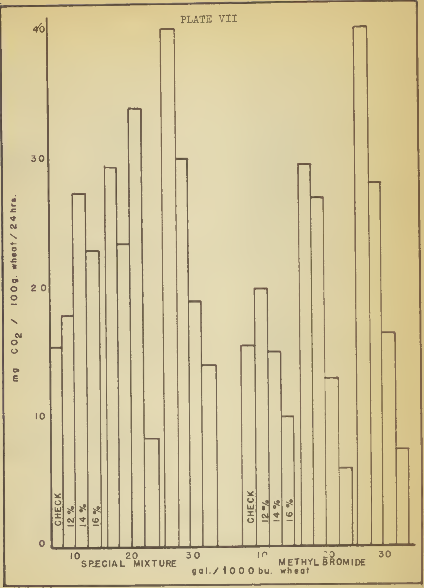


PLATE VI
mg CO_2 / 100 g wheat / 24 hrs.
0
10
20
30
40
CHECK
12 %
14 %
16 %
CHECK .
12 %
14 %
16 %
10
20
30
ETHYLENE DIBROMIDE
10
20
30
ETHYLENE DICHLORIDE
gal. / 1000 bu. wheat

EXPLANATION OF PLATE VII

The effect of the Special Mixture and methyl bromide on the amount of carbon dioxide produced by soft red winter wheat.

PLATE VII


40
30
20
10
0
mg CO_2 / 100g. wheat / 24 hrs.
CHECK
12 %
14 %
16 %
CHECK
12 %
14 %
16 %
10
20
30
10
20
30
SPECIAL MIXTURE
METHYL BROMIDE
gal./1000 bu. wheat

mixture gave an increase in the rate of carbon dioxide production to the 14 percent moisture level followed by a decrease. This was true of all rates of fumigation and at all moisture levels. Generally, this too was the observation regarding the soft wheat samples, except that at the 30 gallons per 1,000 bushel rate a decreasing rate of respiration resulted.

Methyl bromide (Plate VII) had the same tendency to decrease the rate of respiration of the soft wheat at all moisture levels and all concentrations. This also was true for the hard wheat samples.

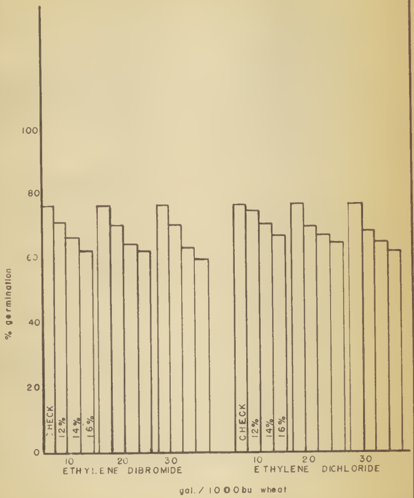
Germination. With the exception of the special mixture, only slight differences could be observed in the effect of fumigation on the viability of the soft wheat (Plate VIII) as compared to hard wheat. The special mixture caused a more pronounced effect upon the germination of the soft wheat (Plate IX). All concentrations of methyl bromide at all moisture levels caused a complete loss in the viability of the soft wheat.

Milling and Baking. Soft wheat samples of 1,000 grams each were milled on a Buehler type experimental mill to approximately 65 percent extraction. As was the case with the hard wheat samples, it was difficult to observe any differences in the milling quality attributable to the fumigation. A longer period was required to clean out the mill between samples than was necessary for the hard wheat, due to the soft finely divided nature of soft wheat flours. The absorption was determined with the aid of the Brabender Farinograph and mixing

EXPLANATION OF PLATE VIII

The effect of ethylene dibromide and ethylene dichloride on the viability of soft red winter wheat.

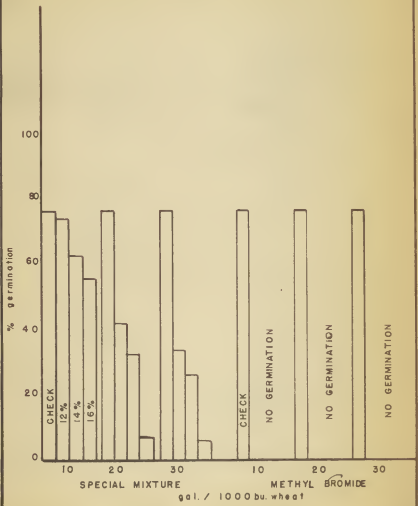
PLATE VIII


100
80
60
40
20
0
% germination
CHECK
12%
14%
16%
CHECK
12%
14%
16%
10
20
30
ETHYLENE DIBROMIDE
10
20
30
ETHYLENE DICHLORIDE
gal. / 1000 bu wheat

EXPLANATION OF PLATE IX

The effect of Special Mixture and methyl bromide on the viability of soft red winter wheat.

PLATE IX



% germination
100
80
60
40
20
0
CHECK
12%
14%
16%
CHECK
NO GERMINATION
NO GERMINATION
NO GERMINATION
10
20
30
10
20
30
SPECIAL MIXTURE
METHYL BROMIDE
gal. / 1000 bu. wheat

time was determined on the National Swanson-Working Recording Micro-mixer. The absorption for the soft wheat flours was much lower than for hard wheat due to the lower level of gluten present. As was observed with the hard wheat flours, the general tendency for decreased loaf volume and inferior crumb and grain characters was inversely proportional to the concentration of fumigant applied. Table 8 gives the complete baking data recorded for the soft wheat flours.

Microflora: Bacteria. The four fumigants in most cases had the same bacteriocidal action with the soft wheats as was noted for the hard wheat samples. Tables 9, 10, 11, and 12 give the actual number of colonies found on the nutrient agar plates. As noted for the hard wheat, ethylene dichloride did not appreciably decrease the growth, even at the higher concentrations.

Microflora: Fungi. Growth was observed at all moisture levels for the control (nonfumigated) sample, enabling comparison of the data at all moisture levels (Tables 13, 14, 15, and 16). These data indicate a definite fungicidal action of all fumigants except ethylene dichloride; this latter fumigant had no fungicidal effects upon the organisms present in the wheat.

DISCUSSION OF EXPERIMENTAL RESULTS

The studies carried out with excessive fumigation of hard and soft wheats at various moisture levels and with various

Table 8. Baking data for soft red winter wheat.

Treatment		Flour protein percent	Mixing time min.	Absorption on 14% M.B. percent	Loaf volume cc.	Crumb* color percent	Crumb' grain & texture	Odor
Control		9.6	1.75 1/	58.4 1/	680	80cy	85-o	None
**12% Ethylene dibromide,	10 gal.	9.6			590	78cy	82-o	None
14% Ethylene dibromide,	10 gal.	9.6			660	80cy	85-o	None
16% Ethylene dibromide,	10 gal.	9.5			670	80cy	85-o	None
**12% Ethylene dichloride,	10 gal.	9.6			660	80cy	85-o	None
14% Ethylene dichloride,	10 gal.	9.6			680	80cy	85-o	None
16% Ethylene dichloride,	10 gal.	9.7			685	80cy	85-o	None
**12% Special mixture,	10 gal.	9.6			655	80cy	85-o	Very slight
14% Special mixture,	10 gal.	9.5			645	80cy	85-o	Very slight
16% Special mixture,	10 gal.	9.6			670	80cy	85-o	Very slight
***12% Methyl bromide,	10 lbs.	9.4			660	80cy	85-o	Slight
14% Methyl bromide,	10 lbs.	9.5			680	80cy	85-o	Slight
16% Methyl bromide,	10 lbs.	9.5			675	80cy	85-o	Slight
12% Ethylene dibromide,	20 gal.	9.5			655	80cy	85-o	None
14% Ethylene dibromide,	20 gal.	9.5			575	78cy	80-o	None
16% Ethylene dibromide,	20 gal.	9.6			680	80cy	84-o	None
12% Ethylene dichloride,	20 gal.	9.6			680	80cy	85-o	None
14% Ethylene dichloride,	20 gal.	9.6			670	80cy	84-o	None
16% Ethylene dichloride,	20 gal.	9.6			660	80cy	85-o	None

Table 8. (concl.)

Treatment		Flour protein percent	Mixing time min.	Absorption on 14% M.B. percent	Loaf volume cc.	Crumb* color percent	Crumb ' grain & texture	Odor
12% Special mixture,	20 gal.	9.7	1.75 1/	58.4 1/	645	80cy	85-o	Slight
14% Special mixture,	20 gal.	9.6			665	80cy	85-o	Slight
16% Special mixture,	20 gal.	9.6			675	80cy	85-o	Slight
12% Methyl bromide,	20 lbs.	9.5			655	78cy	80-o	Heavy
14% Methyl bromide,	20 lbs.	9.6			670	78cy	80-o	Heavy
16% Methyl bromide,	20 lbs.	9.5			665	30cy	80-o	Heavy
12% Ethylene dibromide,	30 gal.	9.5			660	80cy	85-o	None
14% Ethylene dibromide,	30 gal.	9.3			700	84cy	86-o	None
16% Ethylene dibromide,	30 gal.	9.6			660	80cy	85-o	None
12% Ethylene dichloride,	30 gal.	9.6			655	80cy	85-o	None
14% Ethylene dichloride,	30 gal.	9.6			630	80cy	85-o	None
16% Ethylene dichloride,	30 gal.	9.6			600	80cy	85-o	None
12% Special mixture,	30 gal.	9.6			680	85cy	80-o	Slight
14% Special mixture,	30 gal.	9.4			630	80cy	78-o	Slight
16% Special mixture,	30 gal.	9.7			595	76cy	76-o	Slight
12% Methyl bromide,	30 lbs.	9.2			590	76cy	76-o	Heavy
14% Methyl bromide,	30 lbs.	9.8			635	78cy	78-o	Heavy
16% Methyl bromide,	30 lbs.	9.5			620	78cy	76-o	Heavy

* c = creamy
y = yellow
' o = open
** Gallons per 1,000 bushels
*** Pounds per 1,000 cubic feet
1/ Mixing time and absorption were constant for all samples.

Table 9. The effect of ethylene dibromide on bacterial growth, 1:10,000 dilution.

Dosage	Moisture levels		
	12%	14%	16%
Control	73	204	232
10 gallons	20-30	32-36	162-190
20 gallons	7-16	2-3	15-25
30 gallons	2-6	0-5	0-9

Table 10. The effect of ethylene dichloride on bacterial growth, 1:10,000 dilution.

Dosage	Moisture levels		
	12%	14%	16%
Control	73	204	232
10 gallons	2-3	4-10	66-90
20 gallons	59-61	243-272	175-200
30 gallons	7-8	25-36	222-367

Table 11. The effect of the special mixture on bacterial growth, 1:10,000 dilution.

Dosage	Moisture levels		
	12%	14%	16%
Control	73	204	232
10 gallons	56-65	185-193	83-102
20 gallons	25-32	9-15	25-31
30 gallons	10-13	0-4	3-15

Table 12. The effect of methyl bromide on bacterial growth, 1:10,000 dilution.

Dosage	Moisture levels		
	12%	14%	16%
Control	73	204	232
10 pounds	1-2	1-5	0-5
20 pounds	1-3	21-29	0-16
30 pounds	1-2	16-22	2-7

Table 13. The effect of ethylene dibromide on fungal growth, 1:1,000 dilution.

Dosage	Moisture levels		
	12%	14%	16%
Control	42	141	196
10 gallons	0-5	32-63	0-1
20 gallons	29-30	120-137	10-12
30 gallons	3-4	31-52	2-6

Table 14. The effect of ethylene dichloride on fungal growth, 1:1,000 dilution.

Dosage	Moisture levels		
	12%	14%	16%
Control	42	141	196
10 gallons	56-120	0-15	261-300
20 gallons	20-27	201-262	350-402
30 gallons	22-33	180-183	150-172

Table 15. The effect of special mixture on fungal growth, 1:1,000 dilution.

Dosage	Moisture levels 12%	14%	16%
Control	42	141	196
10 gallons	11-17	4-29	4-31
20 gallons	11-21	2-10	100-114
30 gallons	0-0	0-23	0-0

Table 16. The effect of methyl bromide on fungal growth, 1:1,000 dilution.

Dosage	Moisture levels 12%	14%	16%
Control	42	141	196
10 pounds	0-0	0-2	71-92
20 pounds	9-12	7-12	62-96
30 pounds	10-11	15-17	90-95

fumigants illustrated that very definite changes in such factors as respiration, germination, and baking quality occurred.

Respiration. The effect of the four fumigants used in this study on the overall respiration of wheat was quite marked. With both the hard red winter and soft red winter wheats there was a general tendency for the wheat of low moisture content (12 percent) to have a higher rate of respiration when fumigated than when nonfumigated. When the moisture of the wheat was above 14 percent there occurred a rapid decrease in the rate of respiration to a point much below that of the nonfumigated control sample. In those samples fumigated with methyl bromide the decrease in respiration rate was marked, even at the 14 percent moisture level.

Methyl bromide had a greater effect on the respiratory rate than did any of the other compounds used in this research. In every instance the rate of carbon dioxide production was greater at the 12 percent moisture levels than at either the 14 or 16 percent levels. Since the methyl bromide treatment caused the loss of all viability of the grain, the carbon dioxide evolved from these samples must have been due primarily to the microflora present on the wheat. Methyl bromide did have some bacteriocidal and fungicidal action but not all growth was killed. This indicates the very high rate at which the microflora respire, since relatively high values were obtained when a large portion of the microflora were killed by the fumigants.

The respiration of both the hard and soft wheats were affected similarly by each fumigant in all dosages, the main differences observed being the slightly higher rate of respiration of the soft wheat samples. This was to be expected since the ratio of endosperm to germ is much greater in the soft wheat than in the hard wheat and the entire kernel less vitreous due to the lower protein content. This makes the diffusion of solutes from cell to cell during the oxidation of food materials more rapid in soft than in hard wheats. The found differences substantiate the observations of Bailey and Gurjar (1919).

Germination. All four fumigants caused some loss in viability regardless of the moisture content or dosage of the fumigant. Ethylene dibromide and ethylene dichloride did not affect the germination as greatly as did the special mixture and methyl bromide. The special mixture very seriously impaired the germination, especially at the high moisture levels and high concentrations of the fumigant, and methyl bromide, at any of the three levels used, totally destroyed all germinating power. When partial and impairment of germination occurred, it was more pronounced with the soft wheat samples than with the hard, probably due to the more permeable character of the bran coat.

It is important that wheat or other cereals, which are to be planted or in which high viability is an important factor, should not be treated with the special mixture or methyl bromide at more than normal dosage. Wheats for milling purposes, however, may be treated with those fumigants at rates as high as

10 gallons per 1,000 bushels without danger of injury to the baking quality even though germination has been destroyed.

Milling and Baking. Fumigation had no measurable effect upon the milling quality of the samples used in this work.

The effect of excessive fumigation on the odor of the milled products was quite noticeable in the case of both the special mixture and methyl bromide, especially when the hot bread was removed from the oven. This odor largely disappeared after a short period of aeration, to give a loaf with an acceptable odor, except with those samples fumigated with methyl bromide at high concentrations. The overall effect of fumigation on the baking quality was not serious. At the high moisture levels and at high concentrations of the fumigant, there was a slight reduction in loaf volume and a detrimental effect on grain characteristics. Methyl bromide affected the baking quality to a greater extent than did the other fumigants used. Its effect was not great at the 10 pound per 1,000 cubic feet rate but at higher concentrations was definitely injurious. Wheat may be fumigated at normal or even higher dosages without injuring the baking quality. All fumigants may be used as high as 10 gallons per 1,000 bushels without seriously affecting the baking quality.

The work presented in this manuscript is in general agreement with the data reported by others in the field. The effect of fumigation on the production of carbon dioxide confirms studies of Irving (1911), Bailey and Gurjar (1919), and Larmour et al. (1935).

Further study should be made of the effect of various fumigants on the quality of stored wheat. This should be done on a commercial scale, involving sufficient quantities of wheat to give valid storage conditions. Work should be done on the germicidal effect of these fumigants with particular reference to moisture content and dosage.

These results may be applied to practical conditions inasmuch as some of the fumigants used did reduce the amount of carbon dioxide produced, had a definite germicidal effect, and did not impair the baking quality. Great care must be used in applying methyl bromide at high concentrations with high moisture wheats as it seriously affects germination, has a disagreeable odor which may be carried over into the milled products, and may have a harmful effect on the baking quality.

SUMMARY AND CONCLUSIONS

Samples of hard red winter and soft red winter wheats were tempered to three moisture levels and fumigated with four fumigants at three concentrations. The effect of these fumigants upon wheat respiration was determined. Germination tests were made on all samples. To estimate the effect of these compounds on the microflora, samples were plated on nutrient agar and rose bengal media. One thousand-gram samples were milled on an experimental mill and the resulting flours baked to determine the effect of the fumigation on baking quality. The conclusions based on this study are as follows:

1. All fumigants used had a definite effect on the respiration. This was characterized by a stimulation at low moisture content followed by a decrease in the amount of carbon dioxide produced at high moisture levels.

2. The moisture content at the time of fumigation is quite significant. The higher the moisture content, the more effective was the fumigant.

3. All fumigants used lowered the percentage of germination. The viability of the wheat was inversely proportional to the moisture content when fumigated. Methyl bromide-treated wheat failed to germinate no matter what dosages and moisture levels had been used.

4. With the exception of ethylene dichloride, all fumigants had a definite bacteriocidal and fungicidal action.

5. Fumigation did not have any measurable effect on the

milling quality of either hard or soft wheat, except to impair the odor of the flour.

6. The baking quality was only slightly impaired by all fumigants except methyl bromide. This latter had a deleterious effect at the 20, and 30 pound per 1,000 cubic feet dosage.

ACKNOWLEDGMENTS

The author wishes to express his appreciation to Dr. J. A. Shellenberger for his advice and aid throughout the research work, to Dr. Eric Kneen for assistance in the preparation of the final manuscript, to Dr. R. T. Cotton for his help in outlining the research problem, to Nicholas Holowaychuk for the translation of the Russian literature used herein, and to the Department of Bacteriology for the use of their facilities in making the study of the micro-flora.

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