Effects of Methyl Bromide Fumigation on the Viability of Barley, Corn, Grain Sorghum, Oats, and Wheat Seeds[1]

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ABSTRACT

Methyl bromide is a highly efficient fumigant, and is widely used for the fumigation of stored grain, mills, and warehouses. One of its undesirable qualities, at least for seed fumigation, is its phytotoxicity. Viability of seeds may be seriously impaired by excessive treatments with methyl bromide.

Tests were conducted to determine the effects of methyl bromide on the viability of barley, corn, grain sorghum, oats, and wheat seeds when fumigated at 80° F. with different combinations of dosage, exposure, and seed moisture content. Gas analyses were made at the end of each fumigation to verify the methyl bromide concentrations. Germination tests were conducted 24 hours, 30 days, and with wheat only, 6 months after fumigation to observe immediate and delayed effects. There was a pronounced decrease in viability in some cases with the increased period of storage after fumigation. In some experiments standardized seedling evaluations were made so that sublethal, injurious effects could be observed. Many of the fumigated seeds sprouted but did not develop normally.

A margin of tolerance usually exists between the dosages required for insect control and those which are lethal to high quality, dry seeds. This margin of tolerance is dependent upon the complex interaction of several variable factors, including (1) the fumigant dosage applied, (2) the seed moisture content, (3) the length of exposure, (4) the kind of seed, (5) the period and conditions of storage after fumigation, (6) the fumigation temperature, (7) the history of the seed (age, previous fumigations, etc.), (8) the ratio of commodity to total space in the fumatorium (the sorption capacity), and (9) leakage factors in the fumatorium. Some of these factors were explored here, some have been reported by other workers.

In general, the results of the study show that little or no injury occurred when the following combination of conditions existed: (1) the seed moisture was less than 12%, (2) the dosage was less than 2 lb./1000 cu. ft., (3) the exposure period was less than 24 hours, and (4) the temperature was 80° F. High temperature, moisture, dosage, and long exposure all contribute to seed injury from fumigation. When combinations of fumigation conditions occur in which one (or more) of these variables is of a higher order than named above, moderate to extensive germination damage may be expected.

The over-all relative order of tolerance of the five species tested was oats > barley > grain sorghum > corn > wheat.

It is commonly recognized that fumigation of seeds with methyl bromide may cause injury to their germination and growing powers. In view of the fact that methyl bromide is a highly effective fumigant, it is desirable to know how much of this gas different seeds can tolerate under different conditions without damage to viability. Several varied and scattered bits of this type of information have accumulated; however, to the knowledge of the authors, no co-ordinated extensive tests have been previously conducted. At the present time tests similar to those described in this paper are underway by Strong and Lindgren (unpublished data) at the University of California, Citrus Experiment Station, Riverside, Calif., and by King and Garner, Texas A. & M. College, College Station, Tex. Further tests with methyl bromide and other fumigants are in progress at the Manhattan, Kans., Stored-Product Insects Laboratory, where the tests reported herein were conducted.

EXPERIMENTAL PROCEDURE AND MATERIALS.—*Seeds.*—These kinds of seeds were used: Barley (*Hordeum vulgare* L.), Beecher variety, six-row, smooth spring. Corn (*Zea mays* L.), DeKalb, yellow, hybrid. Grain sorghum (*Sorghum vulgare* Pers.), Midland variety, yellow, combine type. Oats (*Avena sativa* L.), Nemaha variety, white, spring. Wheat (*Triticum aestivum* L.), Pawnee variety, Hard Red Winter (Class IV).

Moisture Contents of Seeds.—Before fumigation of the seeds, the moisture content of various lots of each kind of seed was adjusted to 10, 11, 12, 13, and 14%. The moisture content was increased by introducing a calculated amount of distilled water by pipette to seeds held at room temperature in a 2-quart glass jar. The jar was then sealed and rolled a few minutes each day for 1 week to mix the seeds and water. Moisture was decreased,

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when necessary, by drying seeds in a forced-air oven at 90° F. Moisture measurements were made by use of a Steinlite[4] Moisture Tester. The manufacturer of this tester claims it has an accuracy of ±0.25%. Other investigators (Hlynka & Anderson 1949) found the error of estimate to be ±0.4%. Since no method was available to control the atmospheric humidity in the laboratory, the moisture contents of the seeds were not maintained at their original levels after fumigation.

Fumigation of Seeds.—Five-gallon glass bottles were used as the fumatoria (fig. 1). Small bags made of open-mesh rayon curtain scrim were used to contain the grain. Of the small grains, 100 grams of seeds were used in each bag; 50 grams of corn were used in each bag. These seed-filled bags were suspended in the center of the fumatoria. The number of bags in each bottle depended upon the number of moisture levels being tested. Seeds of each moisture level were placed in a separate bag, but one bag of each was placed in a bottle. The ratio of seeds to air space thus approximated that which would normally be found in a large warehouse partially filled with bags or small bins of seeds.

Methyl bromide was applied at the rates of 2, 4, 6, and 8 pounds per 1000 cu. ft. in all tests and at 0.6, 1, 3, and 5 pounds per 1000 cu. ft. in a few additional tests with wheat. The fumigant was measured at 0° F. in the liquid state by a cold pipette and was introduced into the fumigation bottle by releasing a partial (about 15 inches of mercury) vacuum created in the bottle just prior to

FIG. 1.—Seeds held in small bags (A) were fumigated in 5-gallon glass bottles as shown. The glass stopcock (B) and neoprene stopper (C) were used to admit the fumigant and to close the bottle.

fumigation. The air rushing through the stopcock and into the bottle greatly aided in vaporizing the liquid methyl bromide and dispersing the gas in the bottle. The fumigations were conducted at atmospheric pressure, 80° and for exposures of 4, 8, 12, and 24 hours.

Gas Analyses.—Methyl bromide concentrations were determined immediately before removal of the seeds from each fumatorium. This was done by use of a Gow-Mac thermal conductivity gas analyzer. This method of gas analyses is fully discussed by Phillips & Bulger (1953), Monro *et al.* (1953), and Phillips (1957).

Aeration of Seeds.—After removal from the fumatoria, the seeds were aerated for 24 hours by exposing them to the laboratory atmosphere in the open-mesh seed bags used in the fumigation. The portion of each sample not used in the first germination test was held in a 4-ounce glass jar for later tests. Each jar was covered with a screen lid to permit further aeration of the seeds and also to prevent entry of insects.

Germination Tests.—Germination tests were conducted at three different time intervals after fumigation so that immediate and delayed effects could be observed. These tests were started 1 day, 30 days, and with wheat only, 6 months after fumigation. Only whole, undamaged seeds were used for the germination tests.

Two Mangelsdorf germinators were used. Except for a few preliminary tests, the general germination technique and procedures followed those recommended by the U. S. Department of Agriculture (1952). Standardized seedling interpretations were made for one complete test series with wheat in which the seeds were evaluated as to "normal," "abnormal," or "dead." In all other tests, seeds which showed any sign of embryonic development, however slight, were counted as "germinated." According to the standardized seedling interpretations, only normal seedlings are considered germinated. Sprout counts were made after 5 and 10 days' incubation for the standardized seedling series. Normal seedlings found in the 5-day counts were removed from the blotters, while others were left in place for further incubation. A 7-day count was made for each of the samples where seedlings were not evaluated.

Mold Inhibitor.—Mold growth was a problem, especially in samples severely injured by the fumigant. After it was determined that no apparent difference was caused in seed viability by its use, a suspension of Captan (N-trichloromethyl thio tetrahydrophthalimide) in water was applied to the germination blotters before placement of the seeds.

Replicates.—Each fumigation was duplicated in nearly every test series. In some instances several replicate fumigation series were conducted. The numbers of seeds used in the germination tests ranged from 200 to 4100 for each dosage, moisture level, exposure, and kind of seed. These numbers are indicated in the tabular data.

Tabulation and Evaluation of Data.[5]—Results of in-

[4] The use of trade names in this paper is for identification purposes only and does not constitute an endorsement of the product by the United States Department of Agriculture.

[5] The authors are grateful to H. D. Nelson, Assistant Station Leader, Stored-Product Insects Laboratory, Manhattan, Kans., who performed the statistical analyses under the counsel of Dr. H. C. Fryer, in charge of the Statistics Laboratory, Kansas State College, Manhattan.

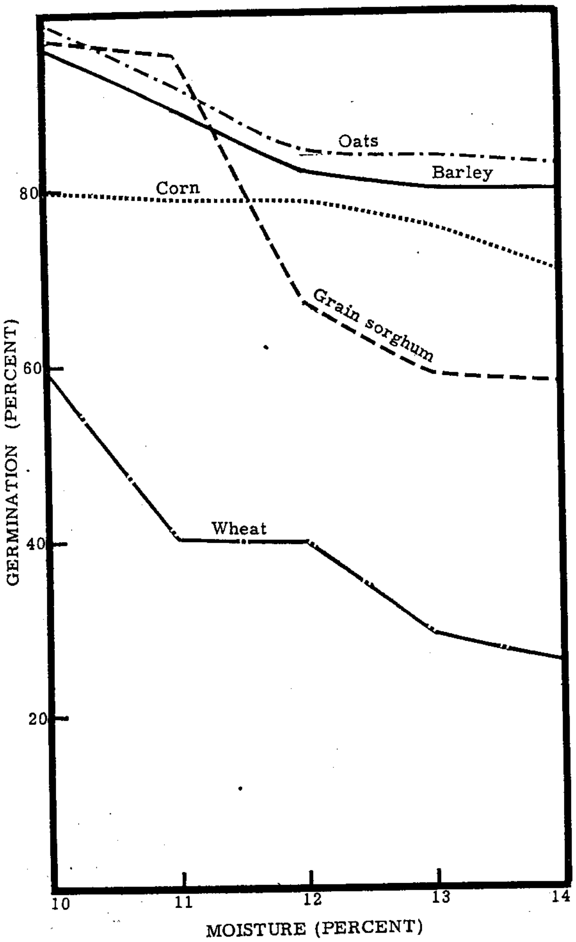

FIG. 2.—An example of the effects of seed moisture content during fumigation on germination (30-day test, 24-hour exposure, methyl bromide 2 lb./1000 cu. ft.).

dividual samples were calculated as percentages and analyzed by the analysis of variance. Least significant differences were computed at the 5% level (L.S.D.*). In some instances, as when a sample was lost or insufficient seeds were available for a complete series, results for the missing individual samples were estimated by an unbiased method. This was done so that each series would have equal numbers of samples, thus simplifying the work involved in the analysis of variance. Wheat data, except the selected data, are expressed in terms of corrected differences between the treated samples and the checks. This correction was made by use of Abbott's formula to account for abnormal and dead seeds in the check samples.

RESULTS.—*General.*—The data presented in this report reinforce, delineate, and extend the existing rather empirical knowledge of the facts relating to the effects of methyl bromide fumigation on seed viability. Injury to seeds was found to be directly related to increases in (1) fumigant concentration, (2) length of exposure, (3) seed moisture content, and (4) length of storage period after fumigation. From other work by Cotton and Frankenfeld (unpublished data), it was demonstrated that the degree of injury increased in close association with increasing fumigation temperatures.

The rates of increasing injury are not uniformly correlated with increases in the variable factors named above, but certain critical thresholds exist. These thresholds are so complexly interdependent that a minor change in one variable may cause the whole system to shift considerably.

Although seedling evaluations were not made for all of the kinds of seeds tested, it was apparent that many of the sprouts from fumigated seeds were slow in developing and were often malformed. This was especially notable in the higher dosages and longer exposures. These observations were confirmed by the seedling evaluations with wheat.

Barley.—Figures 2, 3, and 4 illustrate the fact that barley was one of the more resistant kinds of seed. A summary and least significant differences are presented in table 1 and shows that there was a significant interaction among all the variables involved; namely, dosage ×exposure×moisture×the time interval between fumigation and germination test (germination test number).

A few examples are cited from table 1 in the following

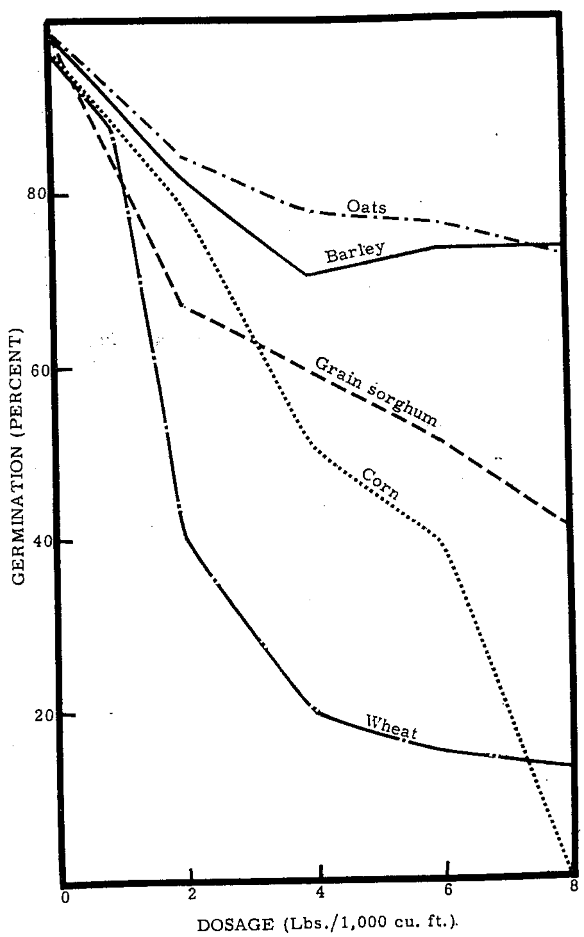

FIG. 3.—An example of the effects of fumigant dosage on seed germination (30-day test, 12% moisture, 24 hours exposure to methyl bromide).

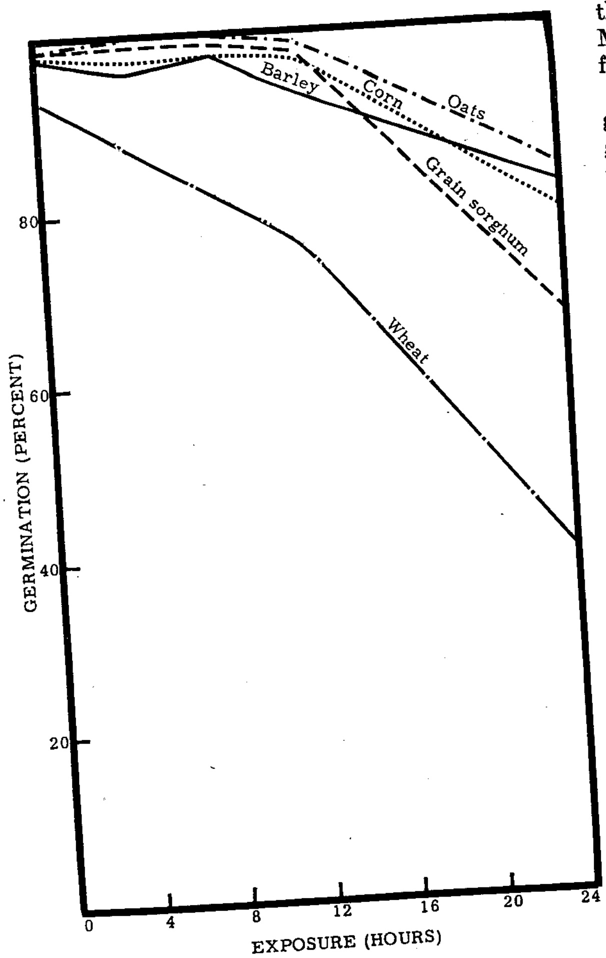

FIG. 4.—An example of the effects of length of fumigant exposure on seed germination (30-day test, 12% moisture, methyl bromide 2 lb./1000 cu. ft.).

tabulation. This tabulation shows the combinations of variables in which significant germination reductions first occurred. The comparisons are between treated samples and their respective checks (L.S.D.* = 6.3). Except for a few instances, all combinations of conditions in which any one of the variables was greater than listed below also resulted in significant injury.

Dosage (Lb./1000 cu. ft.)	*Exposure (Hours)*	*Moisture (Per Cent)*	*Germination Test No.*	
			First	*Second*
2	12	13	x	x
2	24	11	x	x
4	8	11		x
4	12	10	x	x
6	4	13		x
6	4	14	x	x
6	8	10		x
8	4	11	x	x
8	8	10	x	x

As indicated in column 2 of table 1, gas analyses at the end of each fumigation showed that the fumigant concentrations were quite close to the amounts applied.

The results of the barley tests compare favorably with those found by other workers (Fisk & Shepherd 1938, Mackie 1938, Mayer & Gammon unpublished data, California State Senate 1955, Lindgren *et al.* 1955).

Corn.—Figures 2, 3, and 4 show some representative germination values for corn after methyl bromide fumigation, and also the relative susceptibility of corn as compared with the other species tested. As shown in figure 2, increasing the moisture content above 12% caused a slight increase in susceptibility when the seeds were exposed to a dosage of 2 pounds per 1000 cu. ft., for 24 hours. The 12% moisture level is apparently the second moisture threshold with corn, since the treated seeds having only 10% moisture germinated 80% while the checks germinated about 99%. Figure 3 indicates that corn was very sensitive to increasing dosages. The dosage threshold for 12% moisture and 24 hours' exposure lies some place below 2 pounds per 1000 cu. ft. The exact point is not known because no dosages less than 2 pounds were tested in corn. Figure 4 expresses the relationship between increasing lengths of exposure and damage to viability when the dosage was 2 pounds per 1000 cu. ft. and the moisture was 12%. Under these conditions the treatments were tolerated until exposures greater than 12 hours were used. The reaction of corn to increasing exposures was similar to that of oats, barley, and grain sorghum.

A summary and statistical statement are given in table 2. The interaction among all the variables involved was not significant. Interactions of moisture×dosage×exposure and of germination test number×dosage×exposure were significant. The data were arranged according to the combinations just listed, and L.S.D.* values were computed. The following tabulation cites examples of combinations of variables in which significant reductions in germination first occurred. The comparisons are between treated samples and their respective checks.

Dosage (Lb./1000 cu. ft.)	*Exposure (Hours)*	*Moisture (Per Cent)*	*Germination Test No.*		*L.S.D.**
			First	*Second*	
		(Moisture×Dosage×Exposure)			
2	24	10	(Combined)		8.1
4	8	10	(Combined)		5.9
6	4	13	(Combined)		8.1
6	8	10	(Combined)		8.1
8	4	10	(Combined)		8.1
		(Germination test number×Dosage×Exposure)			
2	24	(All combined)	x	x	5.3
4	8	(All combined)	x	x	3.7
6	4	(All combined)		x	5.3
6	8	(All combined)	x	x	5.3
8	4	(All combined)	x	x	5.3

Combinations of variables in which any one factor was increased above those listed resulted in increased injury. Some of the more severe treatments reduced germinations nearly to zero.

Thermal conductivity gas readings at the end of each fumigation indicated that even though the samples were not large the seeds had sorbed a small amount of the methyl bromide during the 24-hour exposure periods.

Table 1.—Beecher barley: effects of methyl bromide fumigation[a] on seed germination.

Dosage and Length of Exposure (Hours)	CH_3Br Analyses (Oz./1000 Cu. Ft.)	Average Per Cent Germination[b] at Indicated Moisture Content of Seeds and Germination Test[c]									
		10 Per Cent		11 Per Cent		12 Per Cent		13 Per Cent		14 Per Cent	
		Test 1	Test 2	Test 1	Test 2	Test 1	Test 2	Test 1	Test 2	Test 1	Test 2
2 lb./1000 cu. ft.											
Untreated check	—	99.7	98.0	99.7	99.3	99.3	97.7	99.0	98.7	99.0	99.0
4	27	99.4	97.8	99.4	99.2	99.2	96.4	99.0	99.2	98.4	97.0
8	28	98.6	99.2	97.4	97.6	97.2	98.2	97.2	95.2	98.2	97.8
12	29	97.8	98.2	98.2	97.8	93.4	93.4	90.0	91.2	93.0	87.2
24	30	94.8	95.6	91.4	88.8	82.4	81.6	81.2	79.8	79.2	79.8
4 lb./1000 cu. ft.											
Untreated check	—	98.3	97.0	100	99.3	97.3	96.7	93.3	87.7	90.7	88.3
4	65	99.0	97.4	98.4	96.2	97.2	91.4	96.0	87.4	97.0	90.4
8	62	98.8	93.4	95.8	84.2	93.4	77.4	87.8	74.4	92.0	74.4
12	62	87.0	84.8	85.4	77.8	81.8	76.2	77.6	75.8	75.0	73.2
24	62	72.8	72.8	74.6	74.6	75.0	69.8	75.2	77.8	71.0	72.6
6 lb./1000 cu. ft.											
Untreated check	—	97.7	98.3	99.7	97.0	99.3	98.3	98.3	95.6	99.3	96.7
4	99	98.4	95.4	98.0	93.2	95.8	94.6	92.6	85.0	91.8	84.2
8	102	94.4	89.4	81.8	79.4	79.8	80.0	78.2	73.4	78.0	75.2
12	99	86.8	86.4	79.8	81.2	76.6	79.6	80.0	73.0	71.2	72.0
24	101	79.6	78.8	74.8	77.2	70.8	72.8	70.0	70.2	71.2	70.8
8 lb./1000 cu. ft.											
Untreated check	—	99.3	97.0	98.2	97.0	98.7	95.3	96.0	95.0	98.3	94.7
4	132	94.6	92.4	84.0	84.8	81.2	84.4	87.2	76.2	75.4	72.0
8	128	85.6	80.6	72.8	71.6	74.8	79.8	76.0	77.4	65.8	57.2
12	128	74.8	75.2	73.4	76.8	76.6	72.6	70.6	76.8	52.6	67.6
24	127	74.2	74.2	71.2	73.2	71.6	73.4	68.8	73.2	45.6	49.2

[a] The fumigation temperature was 80° F. Five hundred seeds tested under each set of conditions except that 300 seeds were tested in the untreated checks.
[b] All seeds showing embryonic development were counted as germinated.
[c] First germination test was started after fumigated seeds were aerated 24 hours. Second test was started after 30 days' aeration.
Least significant mean differences at the 5% level (L.S.D.*).
Comparing samples having 300 seeds, L.S.D.* = 7.0.
Comparing samples having 300 seeds and samples having 500 seeds, L.S.D.* = 6.3.
Comparing samples having 500 seeds, L.S.D.* = 5.1.

Readings at other times were well within the usual limits of variation (table 2, column 2).

A series of tests was conducted by Frankenfeld in 1939 (unpublished data), the results of which indicate that corn had a higher degree of tolerance than shown in this paper. The technique used by Frankenfeld differed from that used in these tests in that the ratio of corn to free air space was much higher in the former, simulating that which is found in bulk storage. As mentioned earlier, in the tests reported here the ratio of seeds to free air space approximated that which would be found in a large warehouse partially filled with bags or small bins of seeds. It is believed that this higher corn-to-air space ratio in Frankenfeld's tests caused a greater amount of the gas to be held by sorption, thus accounting for the higher tolerance indicated. Other factors, such as seed variety, may also effect these differences.

Grain Sorghum.—In general, grain sorghum appeared to be more tolerant than wheat and less tolerant than barley and oats. Figure 2 shows that sensitivity to 2 pounds/1000 cu. ft. for 24 hours was greatly increased when seed moisture was increased above 11%. When compared with corn under these conditions, the tolerance of grain sorghum was considerably above corn at moistures of 10% and 11% but dropped below corn at moisture levels of 12, 13, and 14%. This indicates that at a 2-pound dosage and 24 hours' exposure grain sorghum was more sensitive than corn to moisture changes.

Figure 3 shows that grain sorghum (12% moisture and 24 hours' exposure) was more sensitive than barley or oats to increasing fumigant dosage, but less sensitive than wheat or corn.

Increasing exposures (fig. 4) were tolerated by grain sorghum (12% moisture and 2-pound dosage) through 12 hours' exposure, but serious damage resulted from 24 hours' exposure.

The breaking points (thresholds) varied in relation to the combinations of all factors involved and may be found for each combination by studying table 3. Analysis of variance revealed a significant interaction among the four factors involved; namely, moisture × dosage × exposure × germination test (delayed effect). The L.S.D.* was found to be 5.5 when comparing treated with untreated samples (table 3). On the basis of this criterion, the following tabulation cites examples of combinations of variables listed in table 3 in which significant germination reductions first occurred:

Dosage (Lb./1000 cu. ft.)	*Exposure (Hours)*	*Moisture (Per Cent)*	*Germination Test No.*	
			First	*Second*
2	24	12	x	x
4	8	12		x
4	12	10	x	x
6	8	12	x	x
6	12	10	x	
8	4	11	x	x
8	8	10	x	x

Table 2.—Yellow hybrid corn: effects of methyl bromide fumigation[a] on seed germination.

Dosage and Length of Exposure (Hours)	CH_3Br Analyses (Oz./1000 Cu. Ft.)	Number of Seeds Tested in Each Set of Conditions	Average Per Cent Germination[b] at Indicated Moisture Contents (1st and 2nd Tests Combined)					Number of Seeds Tested in Each Set of Conditions	Average Per Cent Germination[b] in Indicated Germination Test[c] (All Moisture Levels Combined)	
			10 Per Cent	11 Per Cent	12 Per Cent	13 Per Cent	14 Per Cent		Test 1	Test 2
2 lb./1000 cu. ft.										
Untreated check	—	400	99.3	100	99.0	99.3	99.5	1000	99.7	99.1
4	32	400	100	98.8	97.8	98.0	99.3	1000	99.1	98.4
8	32	400	99.0	97.8	98.0	96.8	97.3	1000	99.1	96.4
12	33	400	98.8	98.0	95.3	96.8	91.8	1000	97.2	95.0
24	27	400	79.5	81.3	78.5	74.8	72.3	1000	77.5	77.0
4 lb./1000 cu. ft.										
Untreated check	—	800	98.1	94.0	98.1	98.9	99.6	2000	98.0	97.5
4	64	800	98.6	97.5	96.6	96.4	96.8	2000	97.6	96.8
8	64	800	89.4	84.0	78.8	81.0	70.6	2000	78.7	82.9
12	61	800	76.5	74.4	70.9	69.5	56.9	2000	69.3	70.0
24	55	800	73.3	64.8	47.5	28.6	11.5	2000	45.2	45.0
6 lb./1000 cu. ft.										
Untreated check	—	400	99.3	100	99.5	99.5	100	1000	99.8	99.5
4	97	400	97.5	96.3	92.3	89.8	89.5	1000	94.9	91.2
8	97	400	83.0	76.0	79.5	67.3	62.8	1000	77.5	69.9
12	94	400	75.8	76.3	75.3	37.8	49.0	1000	69.3	56.3
24	92	400	71.0	68.3	71.8	2.3	7.5	1000	46.2	34.1
8 lb./1000 cu. ft.										
Untreated check	—	400	99.3	99.5	99.5	99.3	100	1000	99.8	99.2
4	126	400	90.8	86.8	81.3	80.8	84.3	1000	87.6	81.9
8	125	400	80.3	81.8	72.8	66.8	67.3	1000	77.4	73.1
12	126	400	76.5	73.3	44.3	41.8	60.3	1000	72.5	45.9
24	124	400	60.5	50.5	0.8	2.0	16.3	1000	31.9	20.1

[a] The fumigation temperature was 80° F.

[b] All seeds showing embryonic development were counted as germinated.

[c] First germination test was started after fumigated seeds were aerated 24 hours. Second test was started after 30 days' aeration.

Least significant mean differences at the 5% level (L.S.D.*) when considering these interactions:

Moisture×Dosage×Exposure:
- Comparing samples having 400 seeds, L.S.D.* = 8.1.
- Comparing samples having 400 seeds with samples having 800 seeds, L.S.D.* = 7.2.
- Comparing samples having 800 seeds, L.S.D.* = 5.9.

Test Number×Dosage×Exposure:
- Comparing samples having 1000 seeds, L.S.D.* = 5.3.
- Comparing samples having 1000 seeds with samples having 2000 seeds, L.S.D.* = 4.6.
- Comparing samples having 2000 seeds, L.S.D.* = 3.7.

Combinations of variables in which any one factor was increased above those listed resulted in increased injury. Results of gas analyses at the end of each fumigation indicated that the methyl bromide concentrations were approximately equal to the amount applied.

Experiments reported by Lindgren *et al.* (1955) indicate similar results as reported above.

Oats.—Oats were more tolerant to methyl bromide than any other seed tested, as shown in figures 2, 3, and 4. Increasing the moisture, dosage, and exposure caused moderate increases in damage, but not to the extent observed with the other seeds tested.

Statistical analysis of the data disclosed that significant interactions occurred only at the 2-way level; namely, dosage×exposure, moisture×exposure, and dosage×germination test number. A summary of the individual tests, arranged according to the combinations having significant interactions, is given in table 4. By using the appropriate least significant difference values which are listed at the bottom of table 4, comparisons can be made in the table to show which treatments caused injury and also whether differences existed between first and second germination tests.

The following tabulation cites those combinations of variables in which significant injury first occurred:

Dosage (Lb./1000 cu. ft.)	*Exposure (Hours)*	*Moisture (Per Cent)*	*L.S.D.**
	(Dosage×Exposure)		
2	24	All combined	5.4
4	8	All combined	5.4
6	4	All combined	5.4
	(Exposure×Moisture)		
All combined	12	10	6.0
All combined	8	11	6.0
All combined	4	13	6.0

Each combination in which either factor was increased resulted in increased injury; those with smaller factors were not damaged. Relationships of germinations in the first tests (24 hours' aeration) and the second tests (30 days' aeration) were somewhat erratic; however, two definite trends are shown. The 4-pound dosage (all moisture levels and exposures combined) caused a significantly greater reduction in the first test than was found in the second test. The reverse situation was true for samples treated with the 8-pound dosage.

Table 5 lists average germinations for each combination of moisture×exposure×dosage×germination test number. Although the interaction at this level was not significant, it is obvious that the following are the approximate upper limits of tolerance:

Dosage (Lb./1000 cu. ft.)	*Exposure (Hours)*	*Moisture (Per Cent)*
2	24	11
4	8	11
4	12	10
6	4	12
8	4	11

In fact, some of the examples just cited sustained moderate amounts of injury. In each case, an increase in one of the variables resulted in damage.

Gas analyses at the end of each of the exposures corroborate the theoretical dosages, since they lie within the usual limits of experimental error.

The results of the tests with oats agree with the preliminary findings of Fisk & Shepherd (1938) and those in the khapra beetle research (California State Senate 1955).

Wheat.—General.—Wheat was definitely the most susceptible species tested. Several hundred seeds were used in each of the test series with wheat, partly because of its erratic response and also because wheat viability is of great economic importance. Flour millers do not want dead wheat. There is experimental evidence that shows germ damage from fumigation is directly associated with inferior bread quality when made from flour of damaged wheat (Cotton *et al.* 1946).

Figure 2 illustrates the fact that the sensitivity does not always change in direct and uniform proportions to changes in moisture content. It appears that certain critical moisture thresholds exist here also which are, obviously, dependent upon the combination of the other variables involved. Similar thresholds undoubtedly occur for the other variables, too, but in the combinations used for the data in figures 3 and 4, they are not readily apparent. Figures 3 and 4 indicate that injury to wheat is more uniformly related to dosage and exposure than to moisture, under the conditions of these tests.

Interactions of Variable Factors.—Graphic representations of the germination results from seeds treated with various combinations of dosage, moisture, and exposure aid in giving a clearer understanding of these interactions. Figures 5, 6, and 7 present a few such combinations of summarized data.

The relationship of dosage×moisture×germination is illustrated in figure 5. During a 4-hour exposure, significant, but not drastic, reductions in viability resulted from treatments of 6-pound dosage×14% moisture and of 8 pounds×10% and 11% moisture levels. Severe injury was associated with the 8-pound treatment of seeds having 12, 13, and 14% moisture levels.

Data showing the association of dosage×exposure ×germination are portrayed in figure 6. Germination of

Table 3.—Midland grain sorghum: effects of methyl bromide fumigation[a] on seed germination.

Dosage and Length of Exposure (Hours)	CH_3Br Analyses (Oz./1000 Cu. Ft.)	Average Per Cent Germination[b] at Indicated Moisture Content of Seeds and Germination Test[c]									
		10 Per Cent		11 Per Cent		12 Per Cent		13 Per Cent		14 Per Cent	
		Test 1	Test 2	Test 1	Test 2	Test 1	Test 2	Test 1	Test 2	Test 1	Test 2
2 lb./1000 cu. ft.											
Untreated check	—	97.3	99.7	97.7	98.7	99.3	99.0	98.7	99.0	98.0	98.7
4	27	98.2	98.6	96.8	98.2	98.2	98.6	98.2	98.8	98.0	97.8
8	28	98.4	99.4	98.6	98.6	97.6	99.6	99.0	99.0	95.4	97.8
12	29	99.4	98.4	97.6	98.8	97.4	97.6	96.2	98.0	97.8	95.6
24	30	97.4	96.6	94.6	96.2	70.0	66.8	63.2	58.8	57.6	58.2
4 lb./1000 cu. ft.											
Untreated check	—	98.7	98.7	98.0	97.7	97.7	97.7	98.3	98.3	97.7	99.3
4	65	99.0	99.0	99.0	99.0	98.4	97.8	99.2	96.8	98.6	98.8
8	62	98.8	97.4	94.6	98.0	92.4	91.2	62.4	75.6	71.6	75.4
12	62	90.0	91.8	65.8	85.6	57.8	64.4	40.4	54.0	57.6	50.4
24	62	56.2	58.0	55.6	55.6	57.4	59.0	31.0	22.0	2.2	2.6
6 lb./1000 cu. ft.											
Untreated check	—	99.0	99.7	99.3	98.3	99.7	99.3	99.0	98.7	98.0	99.7
4	99	97.4	98.4	98.8	98.6	97.2	98.4	97.2	95.8	97.4	97.6
8	102	98.2	97.6	96.4	96.4	70.2	80.6	54.4	60.6	45.8	48.8
12	99	93.0	97.4	61.8	80.0	56.8	61.4	50.2	47.0	16.4	16.4
24	101	56.0	56.2	52.4	56.0	51.6	51.4	0	1.6	0	0
8 lb./1000 cu. ft.											
Untreated check	—	99.0	99.0	98.7	100	98.3	100	98.0	99.7	99.7	98.7
4	132	95.2	97.4	90.0	94.8	90.4	94.0	87.2	85.8	67.0	79.2
8	128	79.8	68.4	58.4	60.2	59.4	54.8	50.0	57.6	1.2	0.2
12	128	57.8	62.4	58.4	61.6	50.8	57.2	20.2	18.6	0	0
24	127	54.2	58.2	47.2	58.6	44.8	40.8	0.2	0	0	0

[a] The fumigation temperature was 80° F. Five hundred seeds tested under each set of conditions except that 300 seeds were tested in the untreated checks.
[b] All seeds showing embryonic development were counted as germinated.
[c] First germination test was started after fumigated seeds were aerated 24 hours. Second test was started after 30 days' aeration.
Least significant mean differences at the 5% level (L.S.D.*):
Comparing samples having 300 seeds, L.S.D.* = 6.1.
Comparing samples having 300 seeds with samples having 500 seeds, L.S.D.* = 5.5.
Comparing samples having 500 seeds, L.S.D.* = 4.7.

Table 4.—Nemaha oats: effects of methyl bromide fumigation[a] on seed germination. Summary of individual tests.

Length of Exposure (Hours) and Germination Test[b]	Number of Seeds Tested in Each Set of Conditions	Average Per Cent Germination After Indicated Dosage (Lb./1000 Cu. Ft.)				Number of Seeds Tested in Each Set of Conditions	Average Per Cent Germination at Indicated Moisture Contents				
		2 Lbs.	4 Lbs.	6 Lbs.	8 Lbs.		10 Per Cent	11 Per Cent	12 Per Cent	13 Per Cent	14 Per Cent
Untreated checks	3000	99.7[c]	99.0	99.2	99.1	2400	99.1[d]	99.4	99.6	99.1	99.0
4	5000	99.6	98.7	89.4	88.1	4000	99.2	95.6	93.9	92.1	91.2
8	5000	99.4	89.4	82.1	78.7	4000	95.2	87.3	85.5	85.5	83.4
12	5000	98.4	83.9	78.2	75.8	4000	89.5	83.7	84.5	83.1	79.5
24	5000	86.7	72.5	71.7	64.6	4000	77.9	77.2	77.2	74.8	60.9
First test	11500	95.9[e]	86.4	83.6	81.1						
Second test	11500	97.1	89.2	82.8	78.3						

[a] The fumigation temperature was 80° F.
[b] First germination test was started after fumigated seeds were aerated 24 hours. Second test was started after 30 days' aeration.
[c] All moistures and first and second germination tests combined for each value given in this section.
[d] All dosages and first and second germination tests combined for each value given in this section.
[e] All moistures and exposures combined for values given in this section.

Least significant mean differences at the 5% level (L.S.D.*) when considering these interactions:

Dosage×Exposure:
Comparing samples having 3000 seeds, L.S.D.*=6.0.
Comparing samples having 3000 seeds with samples having 5000 seeds, L.S.D.*=5.4.
Comparing samples having 5000 seeds, L.S.D.*=4.7.

Moisture×Exposure:
Comparing samples having 2400 seeds, L.S.D.*=6.6.
Comparing samples having 2400 seeds with samples having 4000 seeds, L.S.D.*=6.0.
Comparing samples having 4000 seeds, L.S.D.*=5.2.

Dosage×Test:
L.S.D.*=2.2.

Table 5.—Nemaha oats: effects of methyl bromide fumigation[a] on seed germination.

Dosage and Length of Exposure (Hours)	CH_3Br Analyses (Oz./1000 Cu. Ft.)	Average Per Cent Germination[b] at Indicated Moisture Content of Seeds and Germination Test[c]									
		10 Per Cent		11 Per Cent		12 Per Cent		13 Per Cent		14 Per Cent	
		Test 1	Test 2	Test 1	Test 2	Test 1	Test 2	Test 1	Test 2	Test 1	Test 2
2 lb./1000 cu. ft.											
Untreated check	—	100	100	99.7	100	99.7	99.3	99.7	100	99.3	99.7
4	35	99.2	100	99.8	100	99.6	100	99.8	99.2	98.6	99.8
8	34	100	99.8	99.6	99.4	99.0	100	99.4	98.0	99.4	99.4
12	34	100	100	98.4	99.8	98.4	99.2	96.2	98.0	95.4	98.6
24	34	99.8	99.6	85.6	91.6	81.4	84.4	79.4	84.4	78.8	83.4
4 lb./1000 cu. ft.											
Untreated check	—	97.7	99.0	99.3	98.3	98.7	100	99.3	99.7	99.3	99.0
4	67	99.2	99.4	98.8	99.4	98.4	98.8	97.6	98.6	97.8	98.6
8	68	98.8	99.2	89.8	97.0	83.6	88.8	83.8	85.2	84.8	82.8
12	66	97.8	98.2	75.2	83.8	83.8	83.2	76.8	82.6	77.2	80.4
24	67	72.6	73.6	74.0	74.4	75.0	77.4	65.4	80.0	60.8	71.4
6 lbs./1000 cu. ft.											
Untreated check	—	99.0	99.3	99.7	99.7	99.3	100	96.0	100	99.3	99.3
4	98	99.0	99.0	93.2	89.8	94.6	88.2	89.2	86.0	85.0	88.0
8	98	86.4	91.4	84.2	84.4	82.6	76.2	80.6	82.2	76.6	76.2
12	96	76.2	79.0	78.6	82.2	80.2	78.0	85.0	76.8	73.2	72.8
24	96	72.4	75.4	75.2	74.0	78.8	75.8	76.8	72.8	59.4	56.4
8 lb./1000 cu. ft.											
Untreated check	—	99.0	99.0	99.7	99.0	100	100	99.0	99.0	99.3	96.7
4	134	99.4	98.8	95.2	88.2	89.8	81.8	87.6	78.8	80.4	81.4
8	132	96.4	89.8	69.6	74.6	78.8	75.0	78.0	76.6	74.4	73.4
12	132	85.2	80.0	75.6	76.2	79.2	73.8	79.6	70.0	73.4	65.2
24	125	73.8	69.2	70.0	72.6	72.0	72.4	70.4	68.8	39.6	37.6

[a] The fumigation temperature was 80° F. Five hundred seeds tested under each set of conditions except that 300 seeds were tested in the untreated checks.
[b] All seeds showing embryonic development were counted as germinated.
[c] First germination test was started after fumigated seeds were aerated 24 hours. Second test was started after 30 days' aeration.
There are no significant interactions among the above data. Least significant differences are computed for those interactions which are significant and are presented in table 4.

wheat having 12% moisture was significantly, but not drastically, reduced by treatments of 2 pounds×8 hours and of 2 pounds×12 hours. The only samples surviving without injury were those in treatments of 2 pounds, 4 pounds, and 6 pounds×4 hours. All treatments other than those listed above caused great amounts of injury.

Interactions of moisture×exposure×germination are shown in figure 7. When fumigated with 2 pounds of methyl bromide per 1000 cu. ft., slight to moderate damage was done to samples in treatments of 4 hours×14% moisture, 8 hours×12, 13, and 14% moisture, and 12 hours×all moisture levels. Extensive reductions in germination resulted at all moisture levels×24 hours' exposure.

Summary of All Tests with Wheat.—The results of all tests conducted with wheat are summarized in table 6. All these data were not included in the statistical analyses because of the irregular and widely varying numbers of seeds used in each different set of conditions. Representative data were selected for statistical evaluation. The results of these selected data are summarized in table 7 and those of the seedling evaluations in table 8, both of which will be discussed subsequently.

A study of the corrected differences in germination between check and treated samples (table 6) shows that a

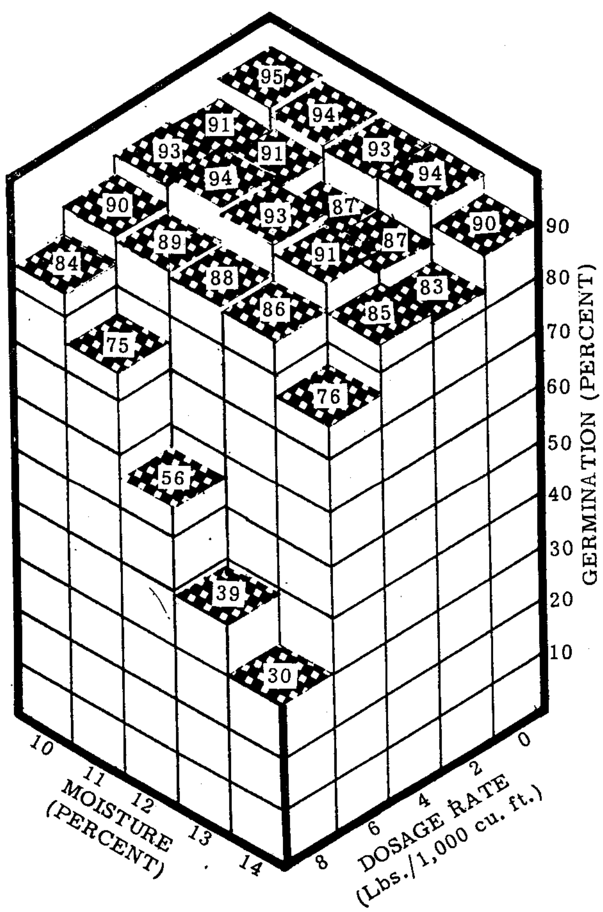

FIG. 5.—Data illustrating the interactions of seed moisture content and fumigant dosage on germination of Pawnee wheat 30 days after fumigation with methyl bromide for 4 hours at 80° F.

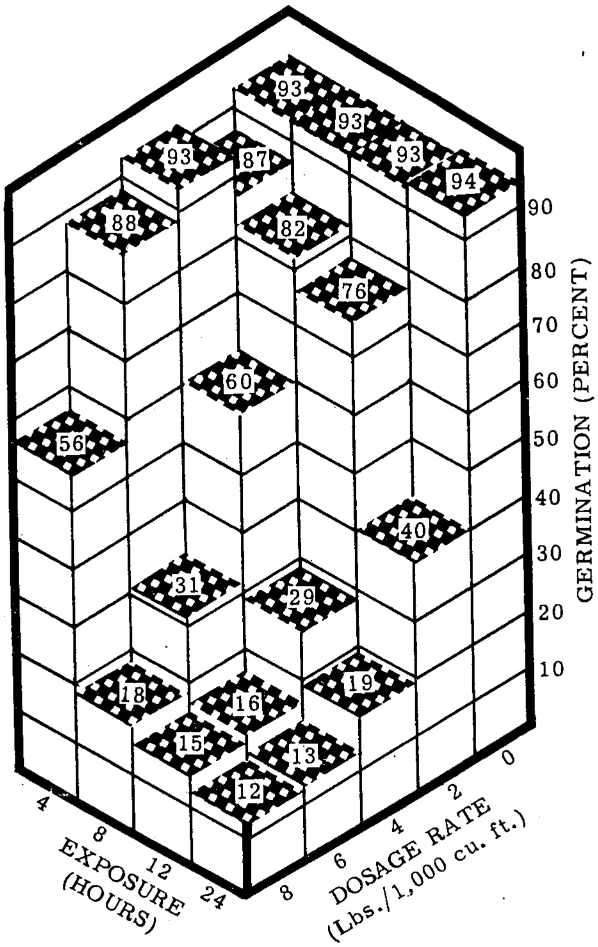

FIG. 6.—Data illustrating the interactions of length of fumigant exposure and dosage on germination of Pawnee wheat 30 days after fumigation with methyl bromide at 80° F. (moisture content of wheat 12%).

dosage of 0.6 pound per 1000 cu. ft. was tolerated by all samples, including those having 14% moisture which were exposed for 24 hours.

The 1-pound dosage caused slight injury in a few samples with the maximum reduction being 11%.

Two-pound dosages caused injury in proportion to the moisture content and the length of exposure. For example, seeds having 14% moisture were injured as follows: (third germination test, 6 months after treatment) 4 hours' exposure, 9% dead; 8 hours' exposure, 15% dead; 12 hours' exposure, 34% dead; and 24 hours' exposure, 70% dead. Lesser amounts of damage resulted when the seed moisture content was lower; however, slight to extensive injury occurred in nearly every sample.

The treatment with a 3-pound dosage×24-hour exposure was tolerated by the samples having 10% moisture, but considerable damage resulted in samples having 12% and 14% moisture.

Four-pound dosages were tolerated for 4 hours' exposure by all except 12% (or more) moisture samples, which sustained slight to moderate injury. Eight-hour exposures resulted in viability reductions ranging from

7% kill in the samples with 10% moisture to 55% kill in those samples with 14% moisture. The ranges of damage resulting from 12- and 24-hour exposures to dosages of 4 pounds per 1000 cu. ft. were 25% to 83% and 64% to 87%, respectively. The degree of injury again was dependent upon the moisture content of the seeds.

Five-pound dosages caused from 12 to 92% mortality of seed embryos.

Six- and 8-pound applications are not safe for wheat unless the moisture content is less than 10% and/or the exposure is shorter than 4 hours.

Selected Data.—Table 7, showing representative germination averages and L.S.D.* values for wheat, was constructed to aid in a more detailed study of the data. The data were selected in units of entire series so that each set of conditions would be represented by the same number of replicates as every other series selected. It should be noted that in the 1-pound dosage series the germination was consistently lower in the first germination test than in the second. This difference was caused by inadvertent drying of the sprouting seeds.

The following tabulation cites examples in table 7 in which significant germination reductions first occurred. The comparisons are between treated samples and their respective checks (L.S.D.* = 7.7):

Dosage (Lb./1000 cu. ft.)	*Exposure (Hours)*	*Moisture (Per Cent)*	*Germination Test No.* First	*Germination Test No.* Second
1	8	12	x[6]	
1	12	14		x
1	24	10	x[6]	
1	24	11	x	x
2	8	12		x
2	12	11		x
2	12	13	x	x
2	24	10	x	x
4	8	11	x	x
4	12	10	x	x
6	4	11		x
6	4	14	x	x
6	8	10	x	x
8	4	10		x
8	8	10	x	x

[6] These are questionable values because of inadvertent drying of seeds as explained in the text.

As a rule, combinations of variables having one factor of a greater value resulted in increased damage, and those with a smaller factor survived the treatment without injury. Other comparisons may be made in the table by using the appropriate L.S.D.* value. Except in the 1-pound treatment series, the delayed effect is indicated by comparing each pair (first and second) of germination tests.

Gas analyses showed that the fumigant concentrations were quite close to the calculated dosages.

Comparison with Results of Other Research with Wheat.—The results of the wheat tests differ considerably from those reported by Cotton and Frankenfeld in 1955 (unpublished data) in that their data showed that the drier seeds were more susceptible than those with 14 and 16% moisture. A contrast of the two methods of fumigation may indicate the key to the differences in results: Frankenfeld fumigated seeds in 1-quart containers fairly well loaded with wheat. The tests currently reported involved fumigations with relatively few seeds in 20-quart containers. Thus the ratio of commodity to air space was greatly different between the two experiments. It is commonly understood that the degree of fumigant sorption by the seeds is proportional to their moisture contents, *i.e.*, the higher moisture is associated with greater sorption. It is possible, therefore, that the high-moisture seeds in Frankenfeld's tests sorbed and held methyl bromide in portions of the seeds other than the embryo, and thereby reduced the effective fumigant concentration below levels toxic to the wheat embryos. If this were the case, the relatively few seeds in the tests reported here would not have had the sorptive capacity to demonstrate this theoretical phenomenon. Further research should be conducted to prove or disprove the theory because of the significant practical implications.

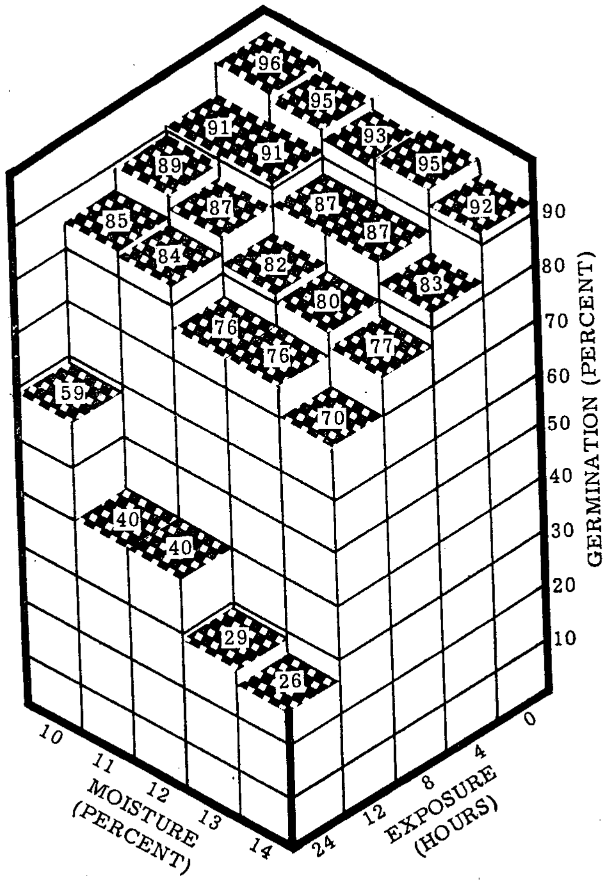

Fig. 7.—Data illustrating the interactions of seed moisture content and length of fumigant exposure on germination of Pawnee wheat 30 days after fumigation with methyl bromide, 2 lb./1000 cu. ft. at 80° F.

Fisk & Shepherd (1938) reported that a dosage of 10 pounds per 1000 cu. ft. for 5 hours' exposure was tolerated by "dry" wheat seeds. Piper & Davidson (1938) reported that fumigations which gave 100 per cent kill to 5 species of test insects (approximately 1 pound per 1000 cu. ft.

Table 6.—Pawnee wheat: effects of methyl bromide fumigation[a] on seed germination. Summary of all tests conducted.

Dosage and Length of Exposure (Hours)	CH_3Br Analyses (Oz./1000 Cu. Ft.)	Mean Per Cent Difference[b] Between Germination[c] of Fumigated and Check Samples at Indicated Germination Test and Moisture Content												
		One Day After Fumigation					30 Days After Fumigation					6 Months After Fumigation		
		10 Per Cent	11 Per Cent	12 Per Cent	13 Per Cent	14 Per Cent	10 Per Cent	11 Per Cent	12 Per Cent	13 Per Cent	14 Per Cent	10 Per Cent	12 Per Cent	14 Per Cent
0.6 lb./1000 cu. ft.														
24	11	− 1[d] (500)[f]	—[e]	1 (500)	—	0 (500)	1 (500)	—	2 (500)	—	1 (500)	—	—	—
1 lb./1000 cu. ft.														
4	16	− 4 (400)	− 6 (500)	− 6 (500)	− 4 (500)	3 (500)	2 (500)	− 1 (500)	− 5 (500)	− 3 (500)	− 8 (500)	—	—	—
8	15	− 2 (500)	− 6 (500)	−10 (500)	− 1 (400)	0 (500)	1 (500)	− 7 (500)	− 6 (500)	− 7 (500)	− 6 (500)	—	—	—
12	16	− 6 (300)	− 7 (500)	− 4 (500)	2 (500)	4 (500)	− 5 (500)	− 5 (500)	− 9 (500)	− 5 (500)	−11 (500)	—	—	—
24	16	− 3 (900)	7 (500)	− 6 (1000)	6 (500)	− 3 (1000)	− 1 (1000)	− 9 (500)	− 7 (1000)	−11 (500)	− 9 (1000)	—	—	—
2 lbs./1000 cu. ft.														
4	31	0 (2600)	− 2 (2000)	− 2 (2600)	− 1 (2000)	1 (2600)	− 4 (2600)	− 3 (2000)	− 5 (2600)	− 7 (2000)	− 9 (2600)	− 2 (600)	− 6 (600)	− 9 (600)
8	31	1 (2600)	− 3 (2000)	− 5 (2600)	− 6 (2000)	0 (2600)	− 6 (2600)	− 7 (2000)	−11 (2300)	−15 (2000)	−15 (2600)	− 3 (600)	−12 (600)	−15 (600)
12	31	− 3 (3100)	−23 (2500)	−18 (3100)	−22 (2500)	−19 (3100)	−11 (3100)	−12 (2500)	−18 (3100)	−20 (2500)	−24 (3100)	− 6 (600)	−31 (600)	−34 (600)
24	31	−32 (4100)	−57 (2500)	−56 (4100)	−70 (2500)	−62 (4100)	−39 (4100)	−58 (2500)	−57 (4100)	−60 (2500)	−72 (4100)	−41 (600)	−62 (600)	−70 (600)
3 lbs./1000 cu. ft.														
24	46	− 5 (1000)	—	−47 (1000)	—	−74 (1000)	1 (1000)	—	−41 (1000)	—	−71 (1000)	—	—	—
4 lbs./1000 cu. ft.														
4	64	0 (2100)	− 2 (1500)	− 4 (2100)	− 3 (1500)	− 6 (2100)	− 1 (2100)	0 (1500)	− 1 (2100)	− 2 (1500)	− 8 (2100)	− 3 (600)	− 8 (600)	−21 (600)
8	64	− 7 (2100)	−22 (1500)	−42 (2100)	−45 (1500)	−55 (2100)	− 8 (2100)	−19 (1500)	−36 (2100)	−44 (1500)	−53 (2100)	−10 (600)	−32 (600)	−40 (600)
12	64	−31 (2600)	−61 (2000)	−70 (2600)	−81 (2000)	−83 (2600)	−28 (2600)	−57 (2000)	−69 (2600)	−78 (2000)	−82 (2600)	−25 (600)	−59 (600)	−76 (600)
24	63	−65 (3600)	−83 (2000)	−81 (3600)	−87 (2000)	−86 (3600)	−64 (3600)	−81 (2000)	−80 (3600)	−83 (2000)	−87 (3600)	−70 (600)	−73 (600)	−73 (600)
5 lb./1000 cu. ft.														
24	82	−22 (1000)	—	−88 (1000)	—	−92 (1000)	−12 (1000)	—	−85 (1000)	—	−92 (1000)	—	—	—
6 lb./1000 cu. ft.														
4	95	−12 (1600)	− 4 (1000)	−14 (1600)	−11 (1000)	−22 (1600)	− 6 (1600)	− 6 (1000)	− 6 (1600)	− 9 (1000)	−17 (1600)	−12 (600)	−13 (600)	−46 (600)
8	99	−39 (2100)	−44 (1500)	−67 (2100)	−82 (1500)	−84 (2100)	−41 (2100)	−45 (1500)	−67 (2100)	−74 (1500)	−81 (2100)	−85 (600)	−81 (600)	−82 (600)
12	97	−66 (2100)	−79 (1500)	−84 (2100)	−87 (1500)	−89 (2100)	−66 (2100)	−80 (1500)	−83 (2100)	−86 (1500)	−88 (2100)	−76 (600)	−82 (600)	−89 (600)
24	96	−75 (3100)	−87 (1500)	−87 (3100)	−88 (1500)	−94 (3100)	−85 (3100)	−86 (1500)	−86 (3100)	−88 (1500)	−95 (3100)	−87 (600)	−86 (600)	−94 (600)
8 lb./1000 cu. ft.														
4	130	−22 (2100)	−14 (1500)	−38 (2100)	−54 (1500)	−82 (2100)	−12 (2100)	−19 (1500)	−39 (2100)	−59 (1500)	−64 (2100)	−30 (600)	−45 (600)	−68 (600)
8	128	−56 (2100)	−74 (1500)	−82 (2100)	−88 (1500)	−86 (2100)	−64 (2100)	−74 (1500)	−80 (2100)	−84 (1500)	−87 (2100)	−73 (600)	−80 (600)	−77 (600)
12	128	−77 (2100)	−84 (1500)	−85 (2100)	−87 (1500)	−92 (2100)	−78 (2100)	−84 (1500)	−84 (2100)	−88 (1500)	−93 (2100)	−77 (600)	−78 (600)	−87 (600)
24	123	−81 (3100)	−87 (1500)	−88 (3100)	−89 (1500)	−96 (3100)	−78 (3100)	−87 (1500)	−87 (3100)	−88 (1500)	−95 (3100)	−78 (600)	−79 (600)	−94 (600)

[a] The fumigation temperature was 80° F.
[b] Differences were corrected by Abbott's formula to account for dead seeds in checks.
[c] All seeds showing embryonic development were counted as germinated.
[d] Negative numbers indicate a lower value in the treated sample than in the check sample.
[e] No test made.
[f] Number of seeds tested in treated samples given in parentheses.

for 12 hours in atmospheric vault at 58° to 70° F.) had no effect on wheat viability.

In the khapra beetle research program (California State Senate 1955), preliminary test results indicated that only slight injury resulted from fumigations of wheat under these conditions: 5 lb./1000 cu. ft., 12 hours' exposure, 10% moisture, and 50° to 53° F.

The effects of repeated fumigations and of the time interval between fumigation and germination test were pointed out by Walkden & Schwitzgebel (1951). Wheat viability decreased more than 50% in association with 2 fumigations and 2 years of storage. Viability of untreated check wheat decreased only about 5%. The fumigant used was a 3 to 1 mixture (by volume) of ethylene dichloride and carbon tetrachloride containing 10% methyl bromide. Tests with the 3 to 1 mixture alone did not cause injury; in fact, it stimulated germination in some instances.

New Wheat.—A few tests were conducted with wheat about 1 month after harvest. The response of these seeds was much more erratic than for aged seeds. The tendency was for the new seeds to be more susceptible. Samples from this same lot of wheat about 3 months later responded similarly to those samples from other aged lots of wheat.

Seedling Evaluations.—Table 8 presents a summary of

Table 7.—Pawnee wheat: effects of methyl bromide fumigation[a] on seed germination. Summary and statistical statement of selected data.

Dosage and Length of Exposure (Hours)	CH_3Br Analyses (Oz./1000 Cu. Ft.)	Average Per Cent Germination[b] at Indicated Moisture Content of Seeds and Germination Test[c]									
		10 Per Cent		11 Per Cent		12 Per Cent		13 Per Cent		14 Per Cent	
		Test 1	Test 2	Test 1	Test 2	Test 1	Test 2	Test 1	Test 2	Test 1	Test 2
1 lb./1000 cu. ft.											
Untreated check	—	85.0[d]	92.3	88.3	96.3	89.7	95.0	84.3	94.3	82.7	97.7
4	16	81.2	94.0	81.6	95.2	83.8	90.4	78.6	90.8	80.8	90.2
8	15	80.8	92.6	81.8	89.4	80.8	89.2	81.4	87.0	79.0	92.2
12	16	80.6	87.0	80.8	91.4	84.8	88.0	84.2	88.8	82.6	87.4
24	16	77.2	89.2	80.6	86.6	77.2	83.8	77.0	84.2	79.8	84.0
2 lb./1000 cu. ft.											
Untreated check	—	100	98.6	98.7	96.0	98.3	97.3	99.0	99.3	99.7	93.7
4	27	99.2	96.4	99.0	97.2	99.0	93.6	99.6	97.8	99.2	94.0
8	28	98.8	95.6	98.2	96.0	97.8	89.4	97.6	93.6	98.2	95.2
12	29	96.8	98.0	94.8	87.0	94.8	89.0	84.0	87.2	87.4	86.0
24	30	63.4	66.0	26.0	27.2	29.8	27.2	23.2	24.4	29.6	18.6
4 lb./1000 cu. ft.											
Untreated check	—	97.7	97.3	99.3	98.7	94.3	95.3	98.3	95.3	94.0	96.7
4	65	97.2	96.7	96.8	96.4	96.7	96.8	98.2	96.0	91.0	90.0
8	62	93.0	95.0	81.4	75.0	62.4	47.2	66.0	37.6	24.6	21.2
12	62	85.2	89.4	32.8	27.8	15.2	10.0	7.4	8.0	5.6	4.6
24	62	10.8	15.0	6.8	5.6	5.4	5.0	6.0	4.0	2.8	0.6
6 lb./1000 cu. ft.											
Untreated check	—	99.3	96.0	98.0	95.7	98.7	96.3	99.7	93.7	97.7	96.7
4	99	95.8	92.4	94.6	83.4	94.8	88.8	93.8	83.0	82.2	70.2
8	102	91.6	83.8	66.4	61.0	32.0	21.4	18.4	11.8	7.4	5.6
12	99	55.6	38.0	20.2	10.0	9.2	8.2	8.4	8.0	3.8	2.8
24	101	4.8	7.8	5.8	5.0	4.6	4.6	4.6	4.8	1.2	2.2
8 lb./1000 cu. ft.											
Untreated check	—	98.3	93.7	98.7	86.7	98.0	89.3	97.3	96.0	97.0	87.7
4	132	94.0	78.4	85.0	44.8	70.4	32.2	56.8	11.4	17.8	5.4
8	128	42.0	12.4	12.6	8.6	5.2	4.4	5.0	5.8	5.0	2.4
12	128	9.2	5.5	6.2	6.6	5.0	5.0	3.8	3.2	1.8	0.6
24	127	6.2	4.2	7.6	5.4	5.8	3.0	3.4	3.4	0.2	0.6

[a] The fumigation temperature was 80° F. Five hundred seeds tested under each set of conditions except that 300 seeds were tested in the untreated checks.
[b] All seeds showing embryonic development were counted as germinated.
[c] First germination test was started after fumigated seeds were aerated 24 hours. Second test was started after 30 days' aeration.
[d] Note that all samples in first test of the 1-lb. dosage showed lower germination than in the other tests. This was caused by inadvertent drying during incubation.
Least significant mean differences at the 5% level (L.S.D.*):
Comparing samples having 300 seeds, L.S.D.* = 8.7.
Comparing samples having 300 seeds with samples having 500 seeds, L.S.D.* = 7.7.
Comparing samples having 500 seeds, L.S.D.* = 6.7.

the tests conducted for the purpose of determining sublethal and delayed effects. The mean per cent differences in the table express corrected differences between fumigated samples and their respective checks. Only seedlings which are normal are counted as germinated by official standards (U.S. Dept. of Agriculture 1952).

Injured seeds developed more slowly than untreated seeds. This is illustrated by the fact that corrected germination differences (normal seedlings) were smaller in the 10-day counts than in the 5-day counts. This means that many of the treated seeds had abnormal sprouts after 5 days' incubation which became normal by the time of the 10-day examination. The check samples were almost completely normal after 5 days. Differences between 5-day and 10-day germination results were greatest in fumigated samples when final counts showed injuries were in the range of about 20 to 50% reduction in 10-day normal seedlings.

Pronounced increases in the percentages of abnormal seedlings are shown in table 8. These are especially notable when germinations (10-day normal seedlings) were reduced by 20 to 75% below the check samples. The abnormal seedlings appeared stunted and/or twisted with many of them lacking essential parts. Frequently the tip of the plumule failed to emerge from the coleoptile, but continued to grow in the form of a loop. As a general rule, a small number of seeds survived the most severe treatments and developed normally, indicating that a few select individuals have a high degree of tolerance to methyl bromide.

As in the other series, gas analyses at the end of each fumigation showed concentrations closely approximating the calculated amount applied.

Discussion of Results and Relation of Seed Tolerance to Dosages Required for Insect Control.—It should not be a surprise to learn that methyl bromide gas will damage or kill the seeds of our domestic plants in view of the fact that the fumigant is used as an effective means of controlling such undesirable weeds as bindweed and crabgrass. It will kill the seeds of these plants as well as the plants proper (Adamson 1956). Of course, dosages used for such purposes are considerably higher than needed for stored-grain fumigation, and a margin of tolerance exists between treatments required for insect control and those which are lethal to seeds.

It is obvious that an absolute line of demarcation can

Table 8.—Pawnee wheat: seedling evaluations to determine the effects of methyl bromide fumigation[a] on seed viability.

Dosage, Length of Exposure, and Moisture	CH_3Br Analyses (Oz./1000 Cu. Ft.)	Mean Per Cent Difference[b] between Fumigated[c] and Check[d] Samples											
		1 Day After Fumigation				30 Days After Fumigation				6 Months After Fumigation			
		5-Day Count	10-Day Count			5-Day Count	10-Day Count			5-Day Count	10-Day Count		
		Normal	Normal	Abnormal	Dead	Normal	Normal	Abnormal	Dead	Normal	Normal	Abnormal	Dead
2 lb./1000 cu. ft.[a]													
4 hours	31.8												
10%		2	2	−1[e]	−1	−5	−2	0	2	1	1	−3	1
12%		−4	0	−2	2	7	19[f]	−8	−8	−15	−10	3	6
14%		−6	−2	1	2	−2	12[f]	−5	−5	−23	−25	13	9
8 hours	31.0												
10%		0	0	0	0	−2	−1	0	1	−1	−1	−3	3
12%		−17	−2	1	1	−11	−8	−1	−6	−44	−43	29	11
14%		−14	−3	1	3	−20	−4	−3	7	−56	−53	32	7
12 hours	31.0												
10%		−24	−3	2	1	−43	−14	3	10	−50	−48	39	6
12%		−46	−18	10	7	−50	−23	6	14	−55	−55	20	32
14%		−43	−19	13	6	−42	−42	12	25	−74	−73	31	34
24 hours	29.0												
10%		−61	−29	15	13	−67	−64	7	52	−71	−72	18	46
12%		−67	−56	16	38	−68	−57	−3	54	−78	−78	11	62
14%		−71	−63	21	39	−75	−74	2	69	−77	−78	2	70
4 lb./1000 cu. ft.													
4 hours	63.0												
10%		−20	−2	3	−1	−9	−7	0	7	−7	3	6	1
12%		−15	−8	2	5	−7	−8	0	7	−57	−42	31	8
14%		−25	−13	4	8	−40	−23	6	14	−66	−49	19	20
8 hours	64.0												
10%		−14	−4	0	4	−37	−8	−2	10	−62	−29	15	6
12%		−63	−37	10	25	−68	−56	21	33	−69	−62	25	32
14%		−57	−43	18	22	−71	−64	23	34	−72	−71	19	41
12 hours	62.3												
10%		−59	−28	12	15	−68	−45	19	23	−68	−67	34	25
12%		−65	−59	12	45	−76	−70	11	56	−77	−77	12	58
14%		−85	−76	1	70	−84	−82	1	76	−75	−76	−10	76
24 hours	60.0												
10%		−71	−69	1	64	−77	−76	−2	71	−74	−71	−12	69
12%		−78	−74	3	70	−81	−81	0	78	−74	−74	−4	73
14%		−88	−77	−4	77	−88	−85	−3	83	−80	−79	−6	72
6 lb./1000 cu. ft.													
4 hours	95.5												
10%		−49	−30	−1	29	−63	−38	27	10	−81	−69	55	12
12%		−53	−33	−1	31	−59	−42	30	9	−77	−71	53	13
14%		−67	−49	+1	46	−79	−72	36	27	−73	−80	25	46
8 hours	103.0												
10%		−76	−72	2	63	−75	−74	14	56	−83	−82	4	74
12%		−79	−69	−1	62	−80	−80	10	64	−77	−80	6	70
14%		−82	−79	−8	80	−89	−85	−1	78	−86	−88	0	81
12 hours	100.5												
10%		−79	−78	−4	76	−85	−84	4	77	−83	−83	3	76
12%		−78	−77	−8	77	−80	−80	−5	79	−84	−84	−1	82
14%		−86	−81	−8	81	−91	−90	−3	85	−96	−96	−1	89

[a] The fumigation temperature was 80° F.
[b] Differences were corrected by Abbott's formula to account for abnormal and dead seeds in checks.
[c] Six 100-seed samples were used for each set of conditions for the fumigated seeds.
[d] Except in a few cases, three 100-seed samples were used for each set of conditions for the check samples.
[e] Negative numbers indicate a lower value in the treated sample than in the check sample.
[f] For unknown reasons the check samples for these samples became very moldy and resulted in lower germination than the other checks or the treated samples.

Table 8.—(*Continued*)

Dosage, Length of Exposure, and Moisture	CH_3Br Analyses (Oz./1,000 Cu. Ft.)	Mean Per Cent Difference[b] between Fumigated[c] and Check[d] Samples											
		1 Day After Fumigation				30 Days After Fumigation				6 Months After Fumigation			
		5-Day Count	10-Day Count			5-Day Count	10-Day Count			5-Day Count	10-Day Count		
		Normal	Normal	Abnormal	Dead	Normal	Normal	Abnormal	Dead	Normal	Normal	Abnormal	Dead
24 hours	97.0												
10%		−83	−82	−9	83	−84	−85	−3	85	−86	−86	−4	87
12%		−80	−77	−9	78	−87	−85	−5	84	−90	−90	0	86
14%		−96	−90	−5	87	−97	−96	−4	92	−96	−97	−4	94
8 lb./1000 cu. ft.													
4 hours	129.0												
10%		−64	−59	−33	64	−68	−65	36	24	−72	−67	27	29
12%		−75	−73	6	65	−70	−68	26	37	−78	−78	29	45
14%		−76	−76	−3	73	−76	−74	4	61	−88	−80	1	69
8 hours	132.0												
10%		−64	−61	−43	74	−77	−75	−2	72	−74	−74	−14	73
12%		−79	−79	−2	78	−77	−76	−7	76	−81	−82	−1	80
14%		−89	−83	0	75	−90	−84	0	77	−93	−85	−2	79
12 hours	132.0												
10%		−68	−68	−43	78	−78	−78	−3	77	−70	−74	18	77
12%		−83	−82	−2	82	−78	−80	−8	81	−81	−80	−1	78
14%		−93	−85	−4	83	−94	−89	0	82	−98	−96	−2	87
24 hours	125.0												
10%		−69	−71	−19	75	−78	−78	−6	80	−77	−78	−3	78
12%		−81	−82	−2	82	−84	−82	−7	82	−84	−82	0	80
14%		−98	−93	−4	90	−98	−96	−2	92	−100	−98	−5	94

not be drawn to show the "safe" level of methyl bromide fumigation for insects in seeds. Rather, one should consider these factors as reported here and by others: (1) dosage, (2) moisture content of the seed, (3) length of fumigant exposure, (4) kind of seed, (5) the condition and period of storage after fumigations, (6) fumigation temperature, (7) history (previous fumigations, age, storage conditions, etc.) of the seed, (8) ratio of commodity to total space in the fumatorium (*i.e.*, sorption capacity), and (9) leakage factors in the fumatorium.

In general, for the first fumigation under carefully controlled conditions, it is possible to achieve satisfactory insect control without damage to seed viability. For example, Lindgren *et al.* (1955) reported methyl bromide LD_{95} values of 8.0 and 17.0 milligrams per liter at 70° F. for 24-hour and 8-hour exposures, respectively, for larvae of the khapra beetle (*Trogoderma granarium* (Everts)), which is considered one of the more resistant species. Dennis & Whitney (1955) found 2.7 and 5.1 milligrams per liter adequate to kill 100% of adult rice weevils (*Sitophilus oryza* (L.)) and confused flour beetles (*Tribolium confusum* Duv.), respectively, when fumigated at 70 to 80° F. for 20 hours' exposure in cylinders without the presence of a commodity. For all practical purposes milligrams per liter is equivalent to ounces per 1000 cubic foot.

Other laboratory tests by Whitney in 1956 (unpublished data) in grain-filled recirculators at 73° to 78° F. indicate that the following dosage rates are adequate to kill all stages of the rice weevil and the confused flour beetle adults, when exposed 24 hours: (1) In wheat, 10% moisture: 5 mg./l.; 12% moisture: 7.5 mg./l.; 14% moisture: 7.5 mg./l.; (2) in corn, 11.3% moisture: 7.5 mg./l.; (3) in grain sorghum, 14% moisture: 12.5 mg./l.; (4) in barley, 11% moisture: 7.5 mg./l.; and (5) in oats, 12% moisture: 7.5 mg./l. Naturally, the interstitial gas concentrations were initially higher than the calculated dosages because of displacement and soon became lower because of the sorption factor.

Thus it is shown that the minimum treatments necessary for insect control may be used without expectation of injury to seeds. The margin of tolerance, however, may be quite narrow, and such factors as repeated fumigations and low initial viability may result in unexpected damage. Further testing is in progress.

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