AN ATTEMPT TO DE-VITALIZE CROP SEEDS IN A COMBINE WITH METHYL BROMIDE AND FORMALDEHYDE

Thesis for the Degree of M. S.

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Luther Aaron Fitch

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AN ATTEMPT TO DE-VITALIZE CROP SEEDS IN A COMBINE WITH METHYL BROMIDE AND FORMALDEHYDE

By

LUTHER AARON FITCH

AN ABSTRACT

Submitted to the College of Agriculture of
Michigan State University of Agriculture and
Applied Science in Partial Fulfillment of
the Requirements for the Degree of

MASTER OF SCIENCE

Department of Farm Crops

Year 1960

Approved ______________________

Some seed of every crop harvested during the summer was taken from an Experiment Station combine at the end of one harvest season. Forty-seven different genera and species of weeds were represented by seeds in this sample.

The effect of methyl bromide gas and 37% formaldehyde fumes, mist, and spray on the germination of some crop seeds was studied in an effort to de-vitalize seeds in a combine. Seeds used were those of crops commonly combine-harvested in Michigan, including wheat, rye, oats, barley, pea beans, and soybeans. Methyl bromide studies were conducted on wheat and oats in air-tight stoppered liter flasks. The effect of formaldehyde was studied on all six of the above crops in a ten cubic foot box and in a combine.

Temperature, moisture, concentration of gas, and length of duration of treatment were all involved in reduction of germination by methyl bromide. It was possible with a temperature of 130°F, and a seed moisture content of 20% to totally de-vitalize wheat with a twelve-hour exposure to 15 lbs./1000 cu. ft. of this gas. Oats under these same conditions still germinated 13%.

20% moisture wheat at 90°F germinated 12% after the above treatment, whereas on 13% moisture wheat the 90°F treatment reduced the germination only to 89% from a control of 97%. Oats were more resistant to injury by methyl bromide gas than was wheat.

LUTHER AARON FITCH ABSTRACT

Because of the extremes in moisture content, concentrations, and temperatures required for complete de-vitalization under atmospheric pressure, it was felt that methyl bromide offered little promise for this purpose without involving special pressure chamber facilities.

Mist or fumes of 37% formaldehyde, ranging in concentration from one-half to four gallons per 1000 cubic feet, killed seeds of wheat and rye at about 12% moisture nearly 100% in a combine in a twelve hour treatment duration at 60°F and above. This was accomplished by draping the combine with polyethylene to form a chamber and spraying the inside of the machine with formaldehyde.

As experimental work showed that effective penetration into layers of grain by formaldehyde gas or mist was very slight (about one inch in twelve hours) this method might be used on wheat or rye as a supplement to mechanical cleaning in a combine, but not as a replacement.

Germination of other moderately dry crop seeds studied was reduced considerably, and was even further reduced by a three-hour pre-soak treatment in steam to soften seed coats. Total de-vitalization however, in all areas of seed lodging in the machine was not obtained.

Dry seeds treated with methyl bromide showed very little loss in viability after a six-months storage period, but dry seeds treated with formaldehyde showed a substantial germination drop in three months storage time.

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INTRODUCTION

A major problem in the production of certified and foundation seed has been that of mechanical mixtures of different kinds or varieties through harvesting equipment. Mechanical cleaning of any of the commercially available models of combines is laborious, and 100% freedom from stray seed is generally impossible to attain. Although it is probable that a machine could be designed for ease and surety of cleaning - certainly it would be highly desirable to have such a combine for seed harvesting - as yet none is commercially available.

The extent of this problem is such that seed growers are customarily advised to grow only one variety of a given crop, to thoroughly clean the combine before moving to another kind of crop, and to discard the first 20 to 50 bushels harvested for seed. By following these recommendations, or even by simply relying on discarding the grain from the first round in the field, the certified seed grower generally comes within the tolerance. However, any crop mixtures seen by the buyer in the seed or in the resultant crop make for poor advertising.

Upon various occasions the inefficiency of the "first round discard" method has been very apparent. In preparing the combine for use in this study, an analysis was made of the material taken from the machine. Some seeds of every crop harvested on the experiment station during the season were found.

A sample of material containing 480 beans, 13 barley kernels, and one kernel of wheat was taken from loose dirt, chaff, and seeds under the feed-in auger of another combine, which had harvested 15 acres of wheat, 40 acres of barley, and 40 acres of beans in that order with no attempt at cleaning between fields.

This problem is magnified in the production of foundation seed where tolerances are more strict, prices of seed are higher, buyers more critical, and acreages much less. The harvesting of increase plots of breeders', or other specially handled seed poses an even greater problem. Here, especially when the acreages range from one to 10 acres, one cannot afford to discard the first 20 bushels, but any method of harvesting other than combining is tedious and might well be costly in terms of timeliness.

In view of the difficulty of removing seeds from a combine this study has been organized to determine the possibility and feasibility of de-vitalization of seeds in a combine by chemical means. Characteristics desirable in a chemical for this purpose would be:

1. A high degree of toxicity to seeds with some residual effects if possible.
2. Non-corrosive to the metals and other materials used in machine construction.
3. Reasonable in cost.
4. Relatively low degree of toxicity to humans.

5. Volatile enough to assure movement and penetration of vapors into areas where seeds might lodge, and to insure evaporation of materials after treating.

REVIEW OF LITERATURE

No previous study on deliberate chemical de-vitalization of seeds has been reported except in relation to soil sterilization. All previous reports of seed injury by chemicals have been related to the purpose of maintaining rather than destroying viability.

Several chemical materials that are commercially used as fumigants and disinfectants have been reported as possessing a high degree of toxicity to seed. Methyl bromide and formaldehyde were chosen as the most logical of these for this study on the basis of reported injury to germination and relatively non-corrosive characteristics.

Toxicity to Seeds-methyl bromide

Toxic effects of methyl bromide on seed germination have been reported in many instances in recent literature. Lewis (11) explains the toxic action of this material ascribing to it the property of inhibition of -SH enzymes with the general formula $R\text{-}SH + CH_3Br \longrightarrow R\text{-}S\text{-}CH_3 + HBr$.

Lubatti and Harrison (13) have investigated the sorption of several fumigants, including methyl bromide, in wheat and report that sorption of fumigants by wheat takes place very slowly. Equilibrium is not reached even after several days. Methyl bromide sorption was the slowest of the fumigants studied, but toxicity to germination by this material was noted. In wheat at 16% moisture sorption of methyl bromide was 3 times higher than at 10% moisture. These workers report that by raising the temperature 30°F (from 55°-85°) the sorption was increased 1½ times. Age and origin of wheat did not affect sorption.

Lindgren et. al. (12) have studied the effect of several fumigants including methyl bromide on seed germination and show a decreased germination in several types of crop seeds with 12 hours exposures at 70°F to varying amounts of methyl bromide. Alfalfa seed at 6.2% moisture, and 92% germination still germinated 92% after treatment with 5 lbs/1000 cu. ft. of methyl bromide; but under the same treatment, germination of seed with a moisture content of 14.2% fell to 75%. Sweet sudan seed at 6.8% moisture and 95% germination germinated 94% after treatment, whereas at 14.6% moisture the germination fell to 14%.

Cobb (5) shows that the concentration of elemental bromine was from 4 to 30 times higher in the embryo of corn than in the endosperm, but that the relationship of germination to bromine in the endosperm appears to show the most direct correlation. In another experiment with fumigated seed rice Cobb reports that the reaction that is responsible for injury to viability, was independent of the total bromine present in the seed. This investigator includes limited findings relative to the factors of length of fumigations, time of storage, seed moisture content, temperatures, and re-fumigation. It was found that at 4 lbs of methyl bromide per 1000 cu. ft. fumigated at 65°F, Ramona seed wheat at 15% moisture showed a rapid drop in germination from 91% to 62% between 8 and 10 hours of treatment. This same wheat re-fumigated 30 days later at 15% moisture showed a germination of 97% after the two exposures of 2 hours, 78% after re-fumigation for 4 hours; 27% after 6 hours and after the 8 and 10 hour re-exposures germinations were 7 and 5%. These data indicate that an initial fumigation may, under some conditions, produce a change in the seeds which makes them more vulnerable to damage upon a second exposure. Work with temperatures and seed moisture contents indicated that below 95°F there were no significant decreases in seed corn germination when exposed to 12 hours at 4 pounds methyl bromide per 1000 cu. ft. There was a direct correlation however, between temperature increase and germination decrease; and between increase in seed moisture content and germination decrease. Only a slight decrease was found when germinated after one year as opposed to immediate germination.

Toxicity to Seeds-formaldehyde

From about 1900 to 1930 formaldehyde was widely used to control covered smut of small grains. Due to concern during this period that this treatment was causing loss of viability in seed grain, rather extensive research was done on the subject. In very few cases, however, were treatments made which completely destroyed germination and in all cases exposure to the chemical was by the dip method commonly used in seed treating. These studies involved dipping the grain in formalin solutions of various strengths for varying lengths of time with the intent to minimize injury to the seed, but maximize the fungicidal action.

Several early workers including Stewart and Stephens (18) Droste, (8) Skinner, (16) Cranefield, (6) Stevens (17), and Bakke and Plagge (3) report varying degrees of injury to seed by formalin treatments.

Horn and Osol (9) working with formaldehyde gas as a fumigant for pest control report that little success is expected at less than 60% relative humidity. Braun (4) showed that injury to seed from formalin treatment could be reduced by pre-soaking the grain for 10 minutes. Treatments were with very dilute solutions, however, so Braun's explanation of the diluting effect of the previously imbibed water on the dilute formaldehyde seems plausible.

Hurd (10) in a comprehensive study on effects of formaldehyde on seed viability noted that wheat was least injured by fumes of 36.2% formaldehyde in an absolutely dry chamber but was entirely killed when the relative humidity was 30% and above. She states that "post treatment injury is usually cumulative, increasing in degree the longer the seed is stored. This seed injury upon drying apparently is due to a deposit of paraformaldehyde on the seed, which forms as the formaldehyde solution evaporates. The solid paraformaldehyde, being volatile, is constantly breaking down into formaldehyde gas. This gas, being thus concentrated and held so close to the seed penetrates it slowly, probably going into solution in the testa."

Barley was found to be less susceptible to post-treatment injury probably because of protection afforded by the glumes, according to Hurd.

Atwood (2) reports a slow entrance of formaldehyde through the seed coat, which he feels substantiates Hurd's theory of paraformaldehyde breakdown. Chemical activity, including respiration rate, diastatic activity, and catalase activity were very drasticly reduced by high concentration of formaldehyde.

Corrosive Effect on Materials

Rabald in Corrosion Guide (14) lists formaldehyde and paraformaldehyde as being non-corrosive (Amts. less than $2.4 g/m^2/$ day) to Al, Al-Si alloys, Cu, brass, bronzes, sn, sn-cu alloys, Al-bronze alloys, Cu-silicon alloys, Fe, steel, cast iron and other metals; rubber, lacquers, and fabrics. This index of "non corrosive" is the lowest corrosion rating given.

Formic acid, a decomposition product of standard 37% formaldehyde which comprises from .03% to .2% of aqueous solution of formaldehyde (19) is listed as being corrosive to iron and steel but not to other metals listed above. Will and Landtblom (20) substantiate this relatively non-corrosive action of formaldehyde on metals. With exposures by these workers of Cu, Sn, Zn, Al, Fe, steel, and brass in a 5% solution of formalin for 10 days no corrosive effect was noted. Dorset (7) lists several advantages of formaldehyde as a fumigant including: 1. Its action is not greatly hindered by albuminous or organic substances. 2. It does not injure fabrics, paint or metal. 3. It can be used safely in households. Disadvantages listed by Dorset are its penetrating odor, and the long length of exposure required.

Little has been reported on corrosive effects of methyl bromide. Although corrosive properties of this material are not listed in corrosion guide, methyl chloride, which should react very similarly is given a "non corrosive" rating to all metals listed, but is highly corrosive to rubber.

Toxicity to Humans

Concerning human toxicity, any fumigant type material capable of destroying seed viability should be handled with a great deal of caution. Of the two materials under observation formaldehyde fumes are more easily identified due to their sharp and irritating odor. Formaldehyde probably is safer to handle since it is in solution and does not have to be kept under pressure at room temperature as does methyl bromide.

PART I STUDIES WITH METHYL BROMIDE

EXPERIMENTAL MATERIALS AND METHODS

Dowfume, contining about 98% methyl bromide and 2% chloropicrin, was used as a source of methyl bromide. A quantity of the gas was released from the pressure can, and stored at atmospheric pressure, by replacement of water, in a five-gallon metal can. Displaced from this container into an air burette, it was metered out as needed.

Seed wheat and oats were brought to calculated moisture contents of 13.2%, 20%, and 30%, and stored at 40°F in glass jars. Samples of about 400 of these seeds were placed in one-liter flasks into which methyl bromide was metered to give:

4	ml. of CH_3Br per liter	or approximately	1	lb. per 1000	cu.ft.
8	"	"	2	"	
20	"	"	5	"	
40	"	"	10	"	
60	"	"	15	"	

Various treatment temperatures were achieved by placing the stoppered liter flasks into a large can over which was suspended a heat lamp. By raising or lowering the lamp, temperatures of 90°F, 110°F, 120°F, 130°F, and 150°F were maintained. Temperatures were measured with a mercury bulb thermometer inserted through the stopper with the bulb resting on the inside base of the flask.

Exposure was from 12 - 36 hours in a ventilated hood. Preliminary tests to determine the extent of injury by fumes which might be given off by treated seed during germination were made by staggering rows of control and heavily treated seeds in the same towel. Since no effect was observed on control plants, all subsequent tests were made using the rolled towel method. Three replications of 100 seeds of each treatment were germinated both immediately after treatment and again six months later. Treated seeds held for storage study were mixed with soybeans and stored in small cloth bags at room temperature (60° - 70°F) to simulate conditions of air movement and ventilation that might be encountered in normal seed storage. Soybeans were used as storage material as it was felt this would involve only a simple screening operation to separate out the treated seeds, but the isolation of individual treated seeds necessary for maximum volatilization of residual materials would still be accomplished.

Classification of seedlings as "normal" was more lenient than is the officially accepted method given in "_Rules for Seed Testing_" (1). Since the purpose was complete de-vitalization it was felt that any seed which might ultimately make a seed-bearing plant should be considered normal if the roots and shoot were 2 cm. in length and still growing after 15 days from the date germinations were started.

EXPERIMENTAL RESULTS

Figure 1 gives a comparison of the kind and variety of seed, seed moisture contents, concentration of methyl bromide, and treatment temperatures for a twelve hour treatment duration.

Control germinations run on the stored seed showed percentages of 97 for the 13.2% wheat, 96 for the 20% wheat; 98 for the 13% oats, and 96 for the 20% oats. Values given in Figure 1 represent an average of three germinations of 100 seeds on each of two replications of each treatment. Statistical analyses were not run, but all replications fell within the officially accepted deviations given in "Rules for Seed Testing" (1).

Reaction of oats and wheat were similar with respect to temperatures, moisture, and concentration of methyl bromide. Oats had an over-all higher degree of resistance.

Treatment Duration

Table 1 shows the effect of treatment duration on wheat at 13% and 20% moisture levels. It may be noted that beyond 12 hours the killing effect tended to increase at a greatly decreasing rate.

Fig. I GERMINATION AFTER 12-HR. EXPOSURE TO CH_3Br

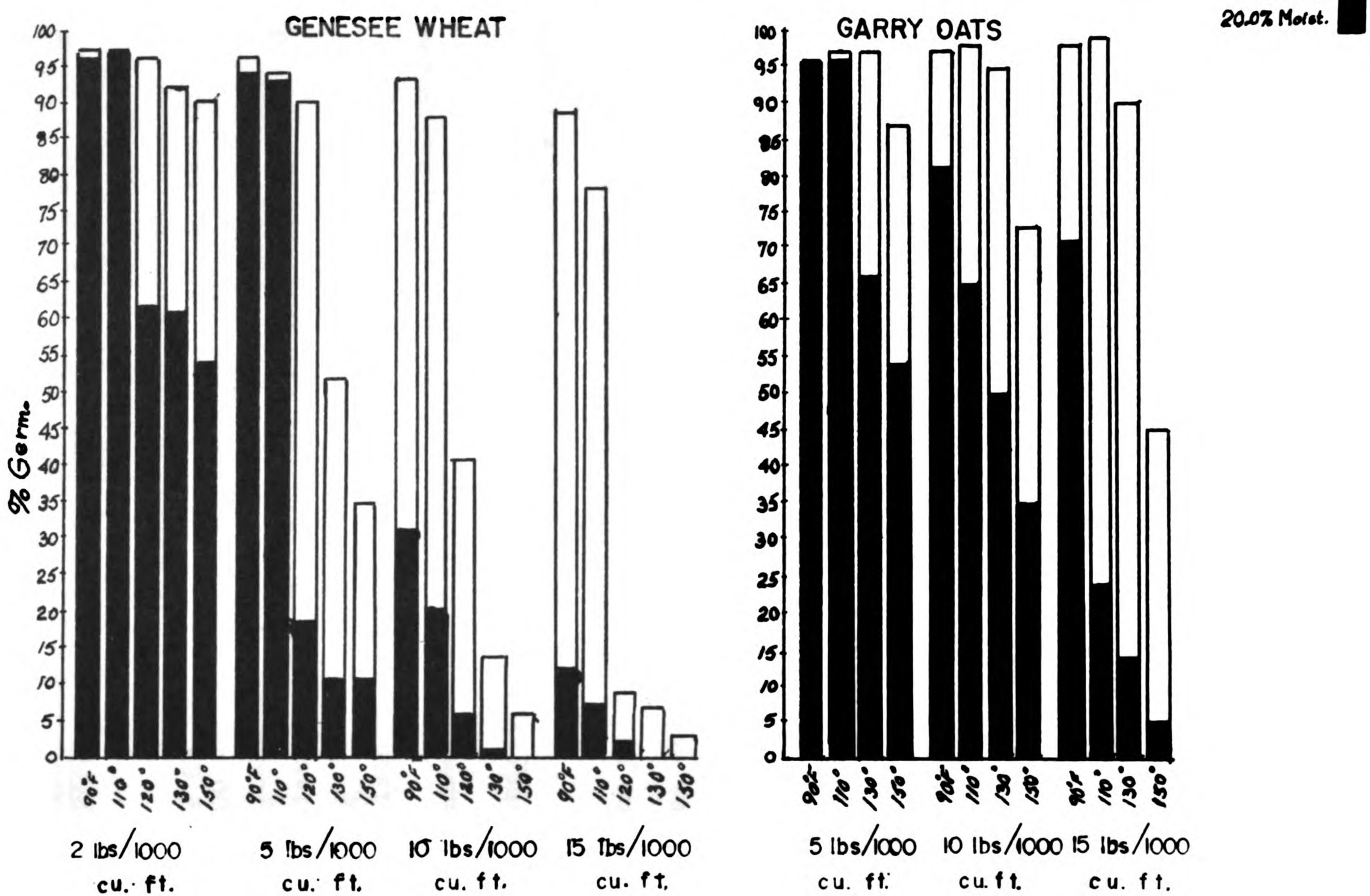


TABLE 1

Effect of Length of Treatment Time on Wheat Germination (treated at 130°F)

lbs CH_3Br/ 1000 cu ft	12 hours				24 hours				36 hours			
	2	5	10	15	2	5	10	15	2	5	10	15
13% moisture	92	50	15	13	95	17	8	5	92	12	6	5
20% moisture	61	11	1	0	50	12	0	0	24	1	0	0

Concentration

It is evident from data in Table 1 and Figure 1 that the major drop in germination due to concentration as such was between the 5 lb. per 1000 cu. ft. and the 10 lb. per 1000 cu. ft. level on the 20% moisture wheat. The germination of oats however was still dropping at a more or less even rate at the highest concentration used. At 13% moisture concentrations at the highest level had little effect without increased temperatures.

Storage Time

Seeds of wheat and oats treated at 13% moisture with methyl bromide and stored for six months in cloth bags at room temperature showed no decrease in germination (Table 2) 30% moisture wheat under the same conditions showed a decrease in germination, even when the treatment was light. Germination of untreated grain did not decrease during 6 months of storage.

TABLE 2

Effect of Length of Storage Time on Germination of Wheat and Oats Given a Previous 10 Hour Exposure to Methyl Bromide at 80°F

	Wheat						Oats					
lbs CH_3Br/ 1000 cu ft	2 lbs.		5 lbs.		10 lbs.		2 lbs.		5 lbs.		10 lbs.	
% moisture	13	30	13	30	13	30	13	30	13	30	13	30
% germ. at time of treatment	98	90	99	90	94	1	98	97	96	90	98	7
% germ. 6 mo. after treatment	97	74	97	67	98	0	95	91	97	87	94	0

SUMMARY AND DISCUSSION OF METHYL BROMIDE TREATMENTS

A one to two day delay in germination and some apparent reduction in vigor was noted even in treated lots of wheat and oats showing 100% of control germination. In some badly injured lots of seed the coleoptile and radicle did not appear to initiate any growth. In other lots the primary leaves and the seminal roots did initiate growth but did not continue to develop.

The greater resistance of oats to injury could probably be attributed to the adhering lemma and palea, since those hulled oats present appeared to be even more susceptible than wheat to treatment injury.

An interaction of all factors, including moisture, temperature, duration of treatment, concentrations, and storage, was involved in reduction of germination in seeds of wheat and oats.

For this method to be useful, essentially complete devitalization is necessary. Under field conditions moisture content of grain will rarely be much above 13% at harvest time, the temperature will not exceed 90°F, and a 12-hour period of treatment seems a general maximum. More than 15 lbs. of methyl bromide per 1000 cu. ft. seems too expensive.

Since germination, even of wheat, under these "field conditions" was hardly affected, studies with methyl bromide were discontinued and formaldehyde was tried.

It is felt that methyl bromide might be used effectively for seed de-vitalization in a combine if special equipment could be used to achieve higher concentrations, increased seed moisture contents, or increased temperatures.

PART II PRELIMINARY STUDIES WITH FORMALDEHYDE

EXPERIMENTAL METHODS AND MATERIALS

Wire baskets containing wheat were suspended over small amounts of 37% formaldehyde in a stoppered liter flask, and a rapid drop in germination was observed. Further preliminary work, using seeds of a known moisture content of wheat, rye, oats, barley, pea beans, and soybeans was conducted in a 10-cubic foot wooden frame enclosed in polyethylene. The frame was one foot wide by two feet high, and five feet long. Cross-members were placed at three different levels to support three wire mesh slides. 1 3/4 inch diameter by 2 inches high screen wire baskets containing the seeds to be treated were placed on the slides, thus enabling the seeds to be removed immediately after treating. Treatments were made in June, July, and August of 1959.

A fine mist spray was used in this investigation as it was felt that some killing action beyond that of the fumes might be accomplished by direct contact of the liquid particles, especially if, as Hurd (10) believes, there is a residual killing action from deposits of formaldehyde polymers on the seed. The mist was delivered by air dispersion from a specially designed glass atomizing flask.

Several experiments were made to test the effects on germination of concentration of formaldehyde, duration of exposure, thickness of seed layers, pre-soaking, and storage time.

No attempt was made to control temperatures as all treatments were made out of doors, but temperature readings made periodically throughout the duration of each treatment ranged from a minimum of 62°F to maximum of 84°F. Treatments of up to 12-hour durations were made late in the evening and vented at night or early in the morning to achieve approximately "harvesting weather" temperature conditions, and to minimize, as much as possible, temperature fluctuations. For the few treatments of longer durations the box was simply kept shaded during the day.

Germinations of all treated and control seed were run in sterile sand with four replicates of one-hundred seeds in small grains and four replicates of fifty seeds in pea beans and soybeans.

The treatment duration series was set up using durations of 6, 12, 24, and 36 hours at a concentration of 1 gal./1000 cu. ft. or 40 ml. of spray in the 10 cu. ft. box. Results are shown in Table 3.

A combined trial to show effects of different concentrations was run using the six different kinds of seed at four concentrations of formaldehyde.

For the investigation on penetration two different size ranges of wheat, oats, and barley were obtained by screening over several size ranges of hand screens. Seeds from the same lot of wheat which stayed over a 7/64 by 3/4 slotted screen and those whole plump seeds which passed through a 1/13 by 1/2 slotted screen were used. A wire tube 6 inches long and 3 inches in diameter with one end closed was used as a container. A one-inch layer of large seeds was placed at the bottom, then a one-inch diameter paper tube was entered on top of this layer and the remaining five inches of the wire tube was filled with the large kernels. The small seed was placed to a depth of four inches in the paper cylinder, and the paper withdrawn carefully leaving a 1" by 4" core of wheat surrounded on all sides by a one-inch thickness of similar kind but larger seed. Because of the size variation it was a simple matter of re-screening after the treatment to separate out this core and make a germination comparison of the two fractions. Two replicates of each of wheat, oats, and barley were run in this fashion with the position of the large and small seed being reversed in the second replication.

To test the hypothesis that the tolerance of barley, pea beans, and soybeans might be overcome if the seeds or seed coats were higher in moisture, an investigation was made using a 3 hour pre-soak treatment. Seed was placed in the wire baskets, and positioned at four levels in the box. One basket of seed was placed in each of the four positions and sprayed with a fine mist of water using a volume of one quart. These seeds were allowed to soak for three hours before the trays were removed, a series of the fresh seeds positioned on the trays, and a treatment made with 80 ml. of formaldehyde (2 gal./1000 cu. ft.).

EXPERIMENTAL RESULTS

Controls showed 90% or better germinations on the six kinds of seed used in the treatment duration experiment. Although some drop in germination was noted beyond a 12 hour treatment period it was so slight that the use of longer periods was not warranted. All further treatments were made on the basis of a 12 hour treatment period.

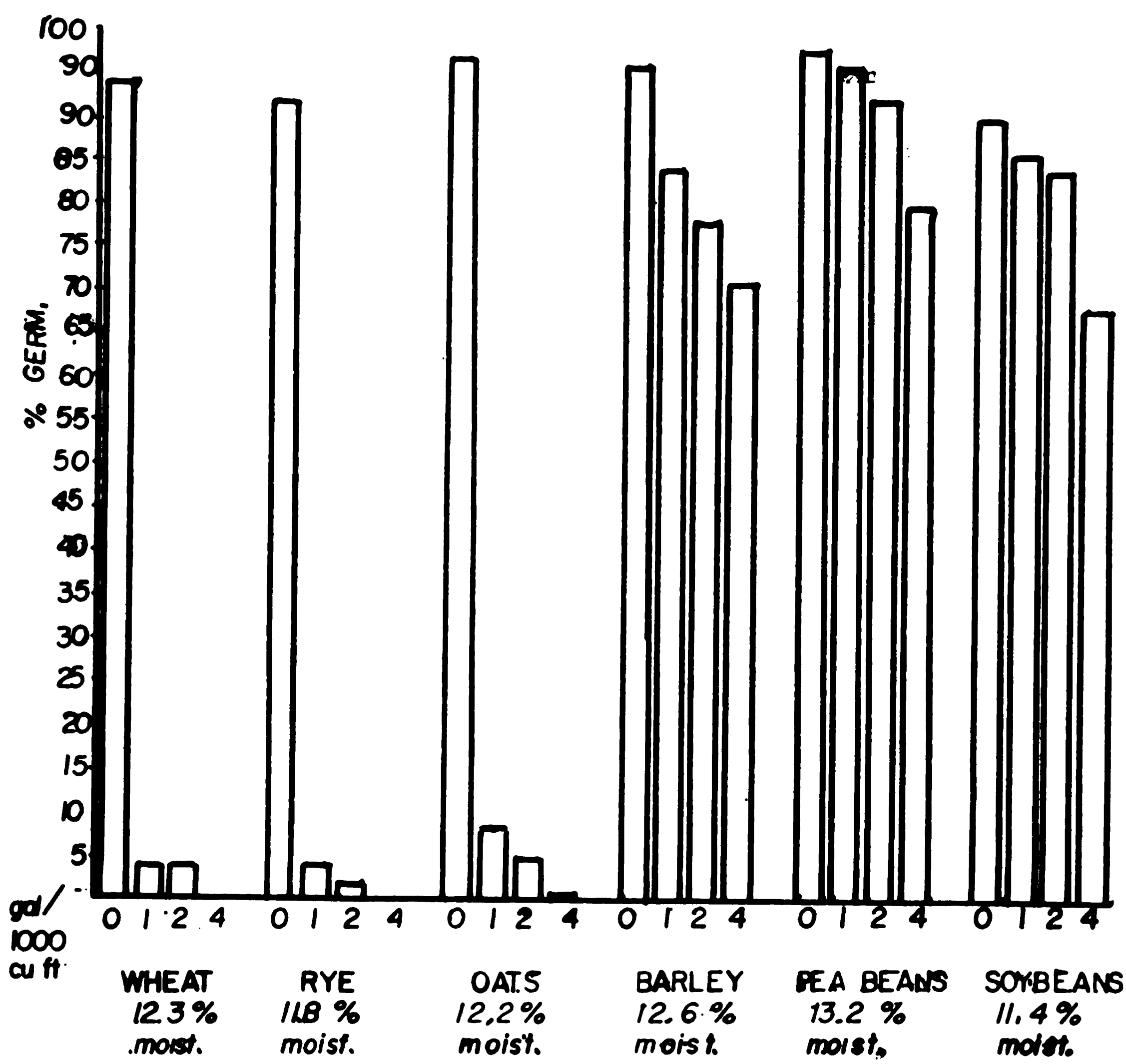
TABLE 3

The Effect of Treatment Duration on Germination of Seeds
(37% formaldehyde mist @ 1 gal./1000 cu. ft. 67°-82°F)

		Wheat @ 12.3% Moist.	Rye @ 11.8% Moist.	Oats @ 12.2% Moist.	Barley @12.6% Moist.	P.beans @ 13.2% Moist.	Soybeans @ 11.4% Moist.
	rep 1	17	13	22	88	96	88
6 Hours	rep 2	12	7	27	83	96	90
	rep 3	11	12	29	89	92	84
	Mean	13.3	10.7	26.0	86.7	94.7	87.3
	rep 1	4	4	8	84	96	88
12 Hours	rep 2	2	4	13	83	94	92
	rep 3	7	2	8	83	94	86
	Mean	4.3	3.3	9.7	83.3	94.7	88.7
	rep 1	1	2	8	72	92	86
24 Hours	rep 2	1	2	5	70	88	80
	rep 3	3	2	10	67	94	82
	Mean	1.7	2	7.7	69.7	91.3	82.7
	rep 1	2	0	2	70	86	80
36 Hours	rep 2	0	1	5	62	82	78
	rep 3	1	0	3	62	86	80
	Mean	1	.3	3.3	64.7	84.7	79.3

Results shown in Figure 2 substantiate the great difference between wheat, rye, and clipped oats and the much less affected barley, pea beans, and soybeans.

FIG. 2 GERMINATION AFTER 12-HR. EXPOSURE TO FORMALDEHYDE



Although there was little actual difference between the effectiveness of the one and the four gallon treatments on wheat and rye, there was a trend which was further evidenced by a 13% to 18% spread between these treatments in barley, pea beans, and soybeans.

As treated plants in the same lots had showed a marked difference in vigor, a trial was made to determine whether this difference might be due to a lack of effective penetration into the inner seeds in the bulk seed mass in the wire baskets. Results of this trial as indicated in Table 4 confirm the supposition that without air circulation or pressure, a relatively small amount of effective penetration can be expected. It may be noted that in the second replication containing smaller kernels in the outer layer of seeds the germination of the "core" seed tended to be somewhat higher.

TABLE 4

Penetration of Formaldehyde Fumes through One Inch of Bulk Seed of Wheat, Oats, and Barley-Using Clean Seed to Indicate Maximum Penetration at Atmos. Press. and 69°-79°F

	% germination Wheat		% germination Barley		% germination Oats	
	rep 1*	rep 2**	rep 1	rep 2	rep 1	rep 2
Inner kernels	23	29	81	87	60	64
Outer kernels	0	0	50	54	27	29

*rep 1 -- core of large kernels, outer layer small kernels
**rep 2 -- " " small " " " large "

Data given in Table 5 shows a decrease in germination from pre-soaking, particularly with barley, but also very evident with the legume seeds. This effect was reversed, however with seed lots of wheat and rye which were sitting on the floor of the box and partially immersed in water. The previously imbibed water appears to have acted as a protectant, preventing to some extent the absorption of formaldehyde.

TABLE 5

Effect on Germination of Two Hour Pre-soak Treatment on Seeds
(2 gal. CH_2O/1000 cu. ft. for 12 hours)

Position no.	Wheat		Rye		Oats		Barley		P beans		Soybeans	
	wet	dry	wet	dry	wet	dry	wet	dry	wet	dry	wet	dry
(1)*	40	22	22	12	22	28	18	68	20	58	20	70
(2)	0	8	0	0	0	6	0	77	28	90	10	86
(3)	0	3	0	2	2	5	2	70	12	96	22	82
(4)	0	0	0	0	0	6	0	79	20	84	18	78

*containers of wheat and rye were partially immersed in water

DISCUSSION OF PRELIMINARY STUDIES WITH FORMALDEHYDE

Results of preliminary studies with formaldehyde indicated that although seeds of wheat and rye, at moisture contents comparable to minimum moistures for harvesting, were rapidly de-vitalized by fumes of formaldehyde, seeds of barley, pea beans, and soybeans with similar moistures were injured very little even at relatively high concentrations.

Later work with oats showed much higher germinations under practically the same conditions, so a review of material used was made. It was found that although the lots used were the same variety and all lots had been processed for certified seed, the lot used in this experiment had been threshed harder or very lightly clipped, leaving a slight gap between the lemma and palea at the terminal end of most of the kernels. It is therefore suggested that these data for oats are not accurate with regard to usefullness in this experiment, but that they do emphasize the role of the "hull" or seed coat in restriction of passage of vapor into dry seed.

Including the previously discussed phenomenon regarding oats, the protection afforded by the lemma and palea or seed coa ts in those seeds studied appears to be a much greater obstacle than any inert tendency of living materials in the seed proper.

PART III TREATMENT OF SEEDS IN A COMBINE

EXPERIMENTAL METHODS AND MATERIALS

To verify the results of preliminary work, and to more accurately evaluate the practicality of this proposed method, investigations were carried out using the Massey Harris "35", 7-foot, self-propelled combine at the Farm Crops Experiment Station.

Prior to the first treatment the sieves were removed from the machine and all panels opened or removed including the slide over the cylinder housing, panels on each side opening into the body of the machine, and the trap under the elevator.

A rough cleaning operation was undertaken, without the aid of air, by tilting the machine at an angle four different ways and running at top speed to dislodge grain in the machine. This method of "cleaning" is one commonly used by farmers, including many seed growers, when going from one field to another. Areas in which material remained lodged after this thorough shaking were noted and samples of the remaining material taken from several areas to be analysed for weed seed and crop seed content. (Analysis is given in the appendix)

Open seed containers were made of heavy, perforated aluminum foil to fit into those major areas of seed lodging including:

1. A slot behind the header
2. Under the cylinder

3. On the grain rack

4. On a three-inch horizontal ledge extending the width of the body behind the beater and above and in front of the straw walkers

5. In the bulk tank under the auger

6. Under the auger in the grain pan

All positions with the exception of position one were in the direct flow of grain through the combine and could be considered as being nearly 100% of all the sources of contamination in this particular machine. Position number one, a two-foot long, V-shaped tube closed at one end, was used as it represented an area of the poorest possible accessibility to spray or fumes. Three other possible sources of contamination which were noted but not used were small one-inch wide horizontal strengthening seams which extended the length of the main body of the machine, the mat of material on the back of the cylinder bars, and the pan under the return elevator auger.

Containers of seeds to be treated were placed by hand and with the aid of tongs in the various positions. Before treating, exhaust and intake areas to the motor were sealed off and the combine was draped with a sheet of polyethylene. Treatments were made in a shed with a cement floor thus allowing an adequately tight seal to be made by pouring a layer of sand around and on top of the edges of the polyethylene.

Approximately twelve feet of extra polyethylene, that were accumulated at the front of the combine, served as a venting flue by stretching it into an open-end "tent" and allowing air movement into and out of the chamber through this large opening. At the time of venting of each treatment there was sufficient air movement so that most of the fumes were dissipated in two hours time. The plastic was then removed and the treated seeds taken from the machine.

Formaldehyde was applied full strength (37%) as a fine spray in the first three treatments and as a fine mist or fog in the fourth treatment.

Methods of Application

Application of 4 gallons of formaldehyde was made in the first treatment with a 3 gallon weed spray can. A John Bean Co. T-jet (potato spray) nozzle with 2/64 aperture and a whirling jet insert was used to achieve a fine spray. The capacity of the attachment at the 50 lb. pressure that was maintained was about 6 gallons per hour. A slot was cut in the polyethylene at the front, both sides, and back of the machine, allowing for the insertion of the nozzle into the body of the combine at these points.

The second treatment was made using the same method except that 2 quarts of formaldehyde were used and a slightly finer spray was obtained by using a Spraying Systems "fog" nozzle with an approximate capacity at 50 pounds pressure of 3 gallons per hour.

Treatment number three used the same nozzle as treatment 2, but a one gallon amount was used.

Since the coverage of seeds by the spray proved to be very erratic in the first three applications an attempt was made in the last treatment to accomplish a more uniform coverage by using a much finer mist and applying from one point only. As no means of obtaining a large quantity of the fog type spray desired was available, the nozzle shown in Fig. (3) was made, modifying only slightly the specifications listed by Rowell and Olien (15). The capacity of this nozzle was calibrated at approximately one-half liter per hour per unit or a total of about 1½ gallons per hour for all six units combined. Since the volume of air required for dispersal of the mist was about 350 cubic feet, 2/3 gallon of extra formaldehyde was added to make a concentration of 2 gallons per 1000 cubic feet.

The nozzle was attached to the rear of the combine with the mist being directed into the body. In this treatment, 5 gallons of water as live steam were applied for one-half hour, and seeds allowed to soak in this atmosphere for three hours before the formaldehyde was applied. After the 3-hour soaking period and before formaldehyde application, fresh lots of seeds were placed in the machine in 4 of the six positions to give a comparison of pre-soaked with non-pre-soaked treatments.

Seed Storage

To duplicate as nearly as possible actual conditions under

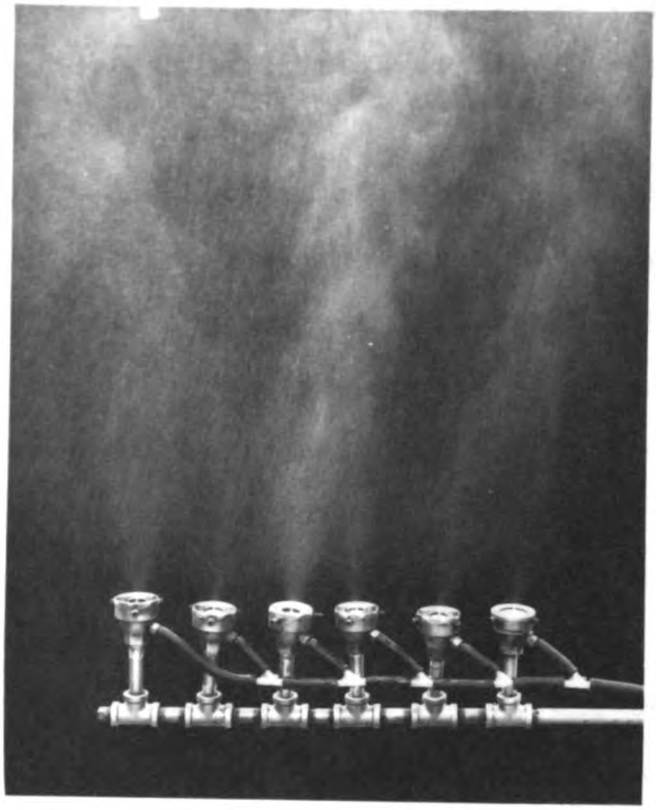
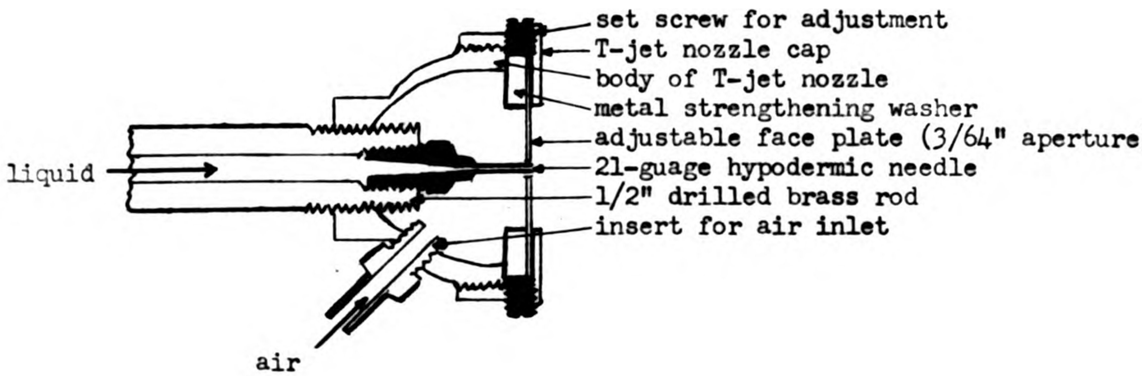


Fig. 3. Spray unit designed for "micro-mist" dispersion of formaldehyde



which these seeds might be stored; immediately upon removal from the machine they were mixed in a cloth bag with one pint of untreated seed, making a ratio of five parts of untreated to 1 part treated by volume.

To facilitate separation of treated seed from material in which it was stored, small grains were stored in pea beans, and treated soybeans and pea beans in wheat. Bags were hung on rafters of the shed for three days, then removed and piled in single spaced rows four bags deep in cardboard boxes and stored along with controls for the next three to four months at room temperature.

Moisture Conditions of Seeds After Treating

No moisture percentages were run at the time of removal of seeds from the combine, but visual observations were made and seeds were classified as follows:

Dry — No observable moisture on seeds or containers.

Moist — Small droplets of moisture on some seeds in the container, or in the bottom of the container.

Wet — Seeds visibly wet or partially or wholly immersed.

A series of germinations were run on portions of the treated seeds one to five days after treating and again after three to four months of storage.

Germinations

All germinations were run in wooden boxes of sterile sand. As previous work had showed a tendency for plants to initiate germination but fail to make any growth beyond an extremely spindly, weak shoot or root system, seeds were planted at a depth of $2\frac{1}{2}$ inches for pea beans and soybeans and $1\frac{1}{2}$ inches for small grains. At this planting depth those very weak seedlings failed to emerge. With the exception of those few plants which died within five days after emergence all plants were considered normal in germination counts.

EXPERIMENTAL RESULTS

Table 6 gives the results of germinations run on all treated seeds from the four treatments with the exception of those seeds subjected to a pre-steaming in treatment number four. It may be noted from this table that the highest germination values of wheat and rye positioned in the direct flow of grain through the combine are seven and four percent respectively.

The 12% and 17% germinations in wheat and rye obtained in placement number one represent a hypothetical situation in which air movement is very slight. No such position existed in the actual area of grain flow through this machine.

Germination of the other kinds of crop seeds show erratic results, indicating a very poor coverage, even in treatment number four where a very fine fog-like spray was used.

Table 7 summarizes the data according to degree of dryness. It is apparent that seeds of oats, barley, pea beans, and soybeans need to be thoroughly moistened to be de-vitalized completely.

Data in Table 8 show that pre-soaking resulted in decreased germination in every case with a maximum reduction, in the germination of barley in number (2) position, of 51%.

Table 9 gives the results of the investigation on the effects of storage time on germination. It may be noted that there is a very substantial decrease in all instances with the small grains, whereas treated pea beans and soybeans, although the results are quite erratic on these crops, tend to show only a very slight decrease in germination during storage.

TABLE 6

Treatment of Seeds In a Combine

Treatment no.	Placement in Combine* (1) cond#	(1) % germ	(2) cond	(2) % germ	(3) cond	(3) % germ	(4) cond	(4) % germ	(5) cond	(5) % germ	(6) cond	(6) % germ
WHEAT												
(1)	d	0	w	0	w	0	m	0	w	0	w	0
(2)	d	1	d	0	m	0	d	0	d	0	d	0
(3)	d	12	d	0	d	1	d	7	d	0	d	0
(4)	-	-	m	0	m	0	m	0	-	-	w	0
RYE												
(1)	d	0	w	0	w	0	m	0	w	0	w	0
(2)	d	0	d	0	w	0	d	0	d	1	m	0
(3)	d	17	d	0	m	0	d	4	d	2	d	0
(4)	-	-	d	1	m	0	m	0	-	-	w	0
OATS												
(1)	m	0	w	0	w	0	m	0	w	0	w	0
(2)	m	3	m	0	w	0	d	52	d	40	m	0
(3)	d	78	m	1	d	47	d	73	d	38	d	80
(4)	-	-	d	41	m	7	d	48	-	-	w	0
BARLEY												
(1)	d	52	w	0	w	0	m	8	w	6	w	0
(2)	d	58	m	30	w	0	d	57	d	40	m	0
(3)	d	75	m	17	d	71	d	65	d	58	d	79
(4)	-	-	d	51	m	30	d	63	-	-	w	0
PEA BEANS												
(1)	m	20	w	0	w	0	w	0	w	0	w	0
(2)	m	20	m	20	w	0	m	40	m	10	m	50
(3)	d	84	d	84	m	10	d	76	d	49	d	86
(4)	-	-	d	45	m	21	d	81	-	-	w	0
SOYBEANS												
(1)	m	20	w	40	m	8	w	0	w	0	w	0
(2)	d	77	d	58	m	26	m	35	d	89	d	59
(3)	d	86	d	86	w	0	d	80	d	76	d	89
(4)	-	-	m	27	m	12	d	68	-	-	w	0

*(1)-behind header auger (2)-under cylinder (3)-on grain rack
(4)-ledge behind beater (5)-in bulk tank (6)-in grain pan

w = wet m = moist d = dry

TABLE 7

Ranges in Germination Percents of Three Moisture Conditions (all placements and treatments)

	WHEAT	RYE	OATS	BARLEY	P BEANS	SOYBEANS
Wet	0	0	0	0-6	0	0-4
Moist	0	0	0-7	0-30	10-40	8-35
Dry	0-12	0-17	38-80	40-79	49-86	58-89

TABLE 8

Comparison of Germinations of Seeds Given a Three Hour Pre-treatment With Steam to Seeds Without Pre-treatment

	Placement							
	(2) Cylinder		(3) Grain rack		(4) Ledge		(6) Grain pan	
	Steam	No Steam	Steam	No Steam	Steam	No Steam	Steam	No Steam
WHEAT	0	0	0	0	0	0	0	0
RYE	0	1	0	0	0	0	0	0
OATS	0	41	0	7	0	48	0	0
BARLEY	0	50	0	30	25	63	0	0
P BEANS	12	45	0	21	37	81	0	0
SOYBEANS	5	27	2	12	40	68	0	0

TABLE 9

Effect of Four Months Storage Time on Germination of Seeds Treated With Formaldehyde

Position in Combine		2 Quarts/1000 cu. ft. Immediate Germination	2 Quarts/1000 cu. ft. 4 months Germination	4 Gal./1000 cu. ft. Immediate Germination	4 Gal./1000 cu. ft. 4 months Germination
WHEAT	(1)	13	1	0	0
	(2)	0	0	0	0
	(3)	0	0	0	0
	(4)	8	0		
	(5)	6	0		
	(6)	0	0		
RYE	(1)	2	0	0	0
	(2)	0	0	0	0
	(3)	0	0	0	0
	(4)	0	0	0	0
	(5)	30	1		
	(6)	0	0		
OATS	(1)	16	3	0	0
	(2)	0	0	0	0
	(3)	0	0	0	0
	(4)	88	52		
	(5)	44	40		
	(6)	0	0		
BARLEY	(1)	74	58	82	52
	(2)	46	30	36	8
	(3)	0	0	0	0
	(4)	70	57		
	(5)	80	43		
	(6)	72	48		
P BEANS	(1)	21	20	20	0
	(2)	28	20	0	0
	(3)	4	0	0	0
	(4)	40	40		
	(5)	20	10		
	(6)	68	50		
SOYBEANS	(1)	78	77	22	20
	(2)	70	58	4	4
	(3)	24	26	12	8
	(4)	80	35		
	(5)	88	89		
	(6)	56	59		

Type of Injury

The type of injury to the embryonic and developing plant by formaldehyde was somewhat varied. A common injury to both monocots (small grains) and dicots (pea beans and soybeans) was repression of radicle growth. The grasses, having seminal roots to "take over" where the radicle fails, were not so much affected by this injury and one-hundred percent of those grass seedlings which emerged in germination trials survived, whereas the legumes - dependent on a "tap root system" - suffered as high as 35% fatality within the first five days after emergence.

Injury to the side of the hypocotyl region closest to the seed coat, coupled with an apparent partial to complete reversal of normal geotropic response for a short duration gave the basal hypocotyl area of even slightly affected legume seedlings the characteristic "hooked" appearance evident in Figures 4 and 5. Another characteristic effect on the legumes was a blackening of the cotyledons of soybeans. All treated soybeans, regardless of germination percent, showed very definite blackening of the cotyledons in contrast to the normally green ones of the control plants.

The over-all vigor of the legume plants from treated seeds was greatly reduced even when germination and survival were practically one-hundred percent of the control. This difference is clearly seen in Figure 4 (top photo) where germination in one case was within five percent of the control.

The vigor of the grass seedlings however was actually increased in several cases as shown by the barley in Figure 6, even when germination was only 60% of control.

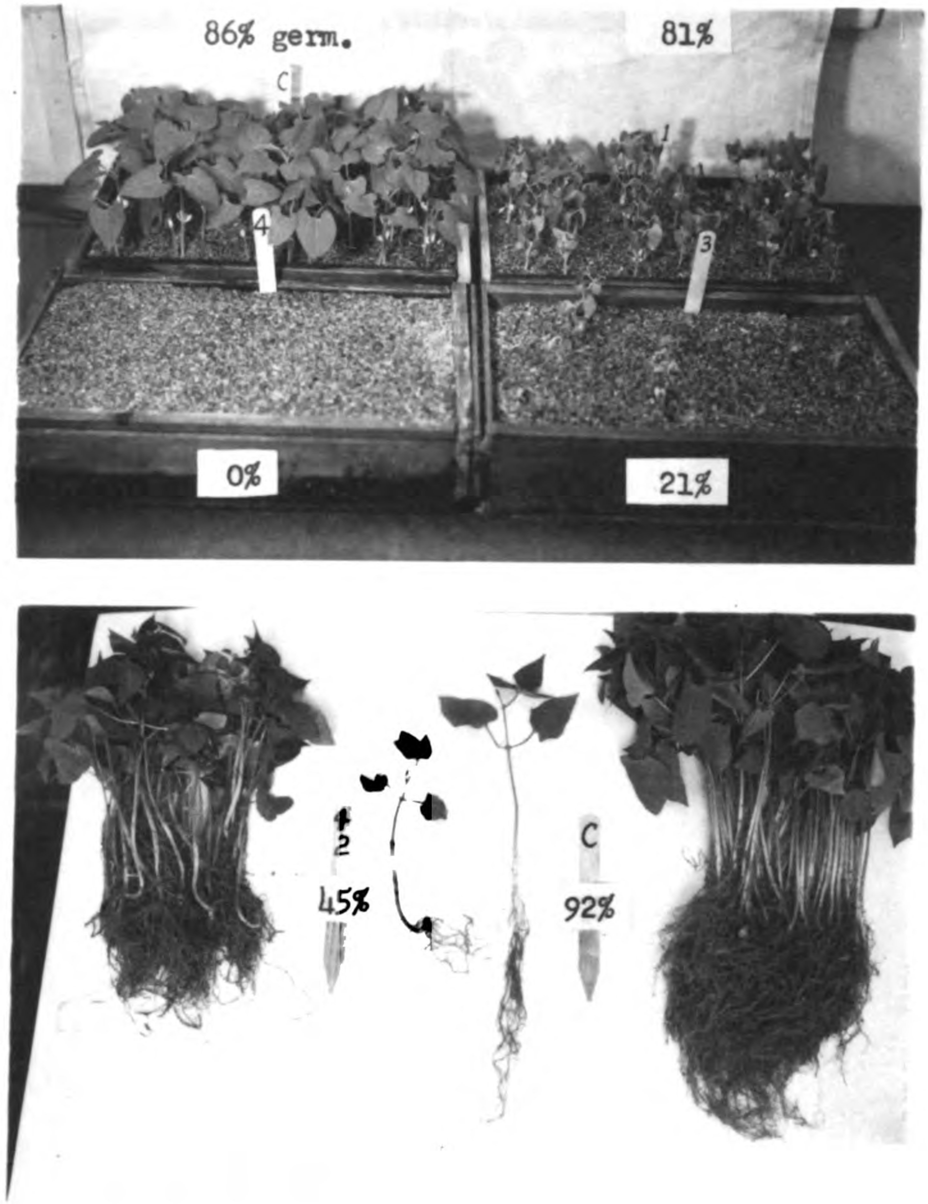


Fig. 4. Effect of formaldehyde on germination and seedling growth of pea beans.

Top photo shows characteristic stunting (lot 1) and range in effects with same treatment but different positions (lots 1 and 3). Lot 4 shows complete de-vitalization.

Bottom photo shows stunting, characteristic hooked effect of hypocotyl, and repression of root development.

Fig. 5. Effects of formaldehyde on germination and seedling growth of soybeans.

Top photo shows "hooking" of hypocotyl, darkened cotyledons, and lack of root development.

Bottom photo shows delayed emergence and extreme stunting.

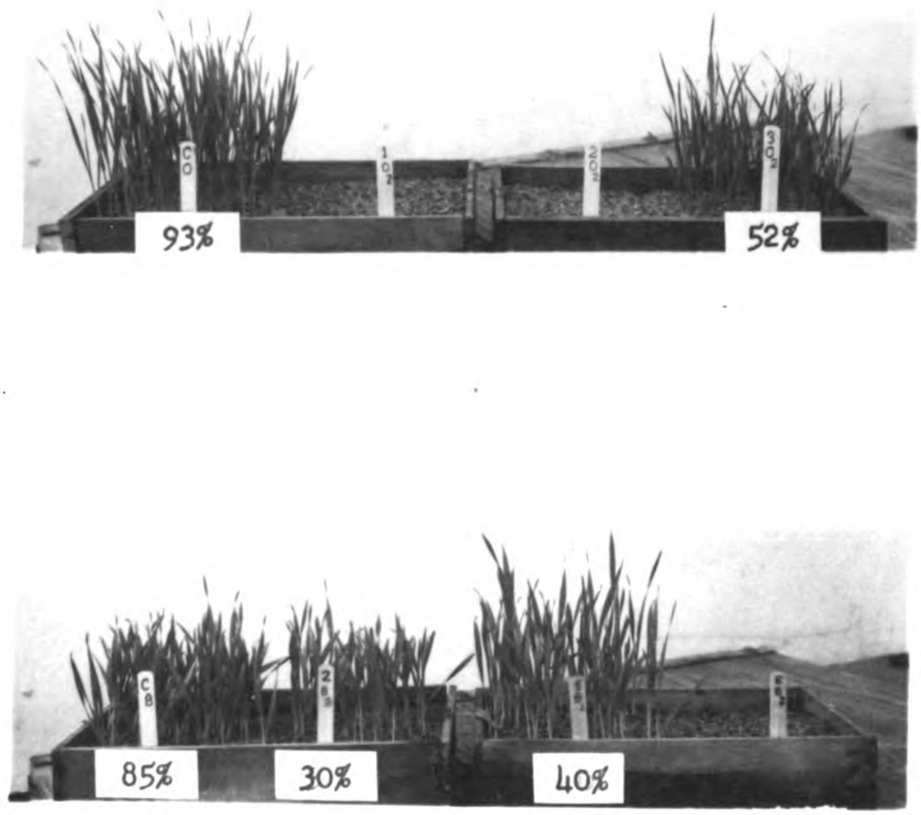


Fig. 6. Effect of formaldehyde fumes and mist on germination and seedling development of oats (top) and barley (bottom).

Top photo shows variability of germination from the same treatment on oats, but in different positions in the combine.

Bottom photo shows a decreased germination of barley resulting from treatment, but an increased vigor of the remaining seedlings. A slight difference in color of the treated lots is also evident.

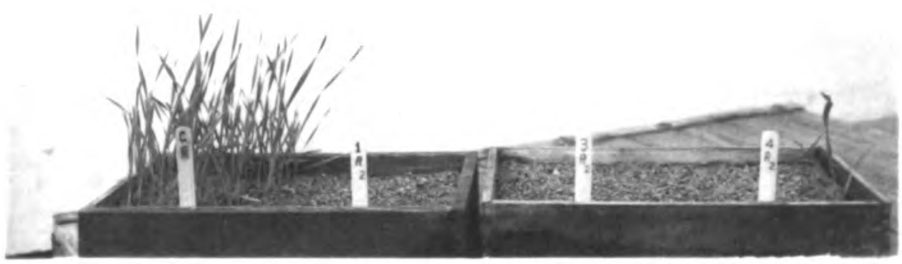


Fig. 7. Effect of formaldehyde fumes and mist on germination and seedling development of wheat (top) and rye (bottom)

Top and bottom photos show practically complete de-vitalization of wheat and rye. The one seedling which did emerge in each case is only slightly less vigorous than the controls.

<u>Effect Of Treatment On Machine Metals</u>

Visual observations were made for rusting after every treatment. The only metals affected at the end of all treatments were bare iron or steel, such as the sickle guards, and the cutter bar. At the end of the first three treatments, corrosion was only evident as almost indiscernible scattered small flecks, but it is estimated that the steaming in the fourth treatment caused at least one-hundred percent more corrosion than did the small amount of formic acid in the first three treatments combined. Rust flecking appeared to be independent of deposit areas of the solid white polymers of formaldehyde.

GENERAL SUMMARY AND DISCUSSION

In setting up this problem for investigation the proposition was advanced that it might be a very easy and uncomplicated matter to cover a combine, and using a simple fumigation technique, kill any seeds which might be inside. The data obtained in these experiments indicate that the problem is not easily solved in this way.

Under normal harvest moisture and temperature conditions, seeds of wheat, although apparently the most easily affected by chemical fumigation, were damaged only slightly by methyl bromide concentrations three times higher than dosages used in commercial fumigation work.

Seeds of wheat and rye succumbed rapidly to fumes of formaldehyde but techniques used were not sufficient to completely kill seeds of other major combine-harvested crops. A pre-soaking treatment appeared to offer a good possibility for de-vitalization of beans, soybeans, barley, and oats, but this line of investigation was not pursued further because of increased corrosion. Corrosion from formaldehyde in the three "dry" treatments was negligible.

Uniform distribution of liquid formaldehyde spray was impossible to obtain, although aided to a degree by the mist-nozzle used in the last treatment, The uneven distribution did indicate however, that seeds other than wheat and rye would have to be thoroughly wet before they would be killed in this 12-hour treatment duration.

Seeds of small grains showed a greater decrease in viability over a period of storage time when treated with formaldehyde than those treated with methyl bromide. Why the same magnitude of decrease in germination of pea beans and soybeans does not take place is not known, nor is the great amount of variation in germination decrease in soybeans understood.

It is felt that the very apparent increase in vigor and size of barley seedlings from formaldehyde treated seed was probably due to the combined earlier emergence - noted in every case with barley-and the fungicidal actions of the formaldehyde on those seed-borne saprophytes or parasites present. It is also feasible that a preconditioning may take place, either in the form of making the seed coat more permeable to water, or in enzyme stimulation or a combination of the two factors.

Although it is possible that with proper temperatures, pressures, moisture contents of seeds, or combinations of these, a 100% chemical sterilization could be accomplished using these materials, re-modeling a combine would probably be a great deal less expensive, no more time consuming, and could accomplish practically the same results.

It is felt that the value of this work lies in the demonstrating of the feasibility of this proposed method of "combine cleaning"; in the stimulation of thought as to other more usable methods; in the indication of the vast difference between germination drops due to fumigation, and total de-vitalization by the same means; and in further demonstration of the inadequacy of available equipment for special seed harvesting work.

CONCLUSIONS

1) Concentrations of 15 lbs/1000 cu. ft. of methyl bromide under ordinary temperature and pressure conditions are not sufficient in the 12-hour period used, to de-vitalize seeds of wheat and oats at harvest moisture contents.

2) Seeds of wheat and rye at harvest moisture contents are practically 100% de-vitalized by fumigation with 37% formaldehyde for 12 hours at concentrations above 1 gallon / 1000 cu. ft.. It is felt that this might represent a usable method to supplement mechanical cleaning. It should be emphasized, however that in view of the limited penetration when used without pressure and for a limited duration this method could not be used as an alternative to mechanical cleaning, even in wheat and rye, but only as a supplement.

3) Seeds of pea beans, soybeans, barley, and oats were not sufficiently injured by any treatments made, with the exception of actual soaking, for this method to be considered successful on these crops.

LITERATURE CITED

1. Anonymous. 1954. Rules For Testing Seeds, "Germination Tests". Proceedings of The Association Of Official Seed Analysts.

2. Atwood, W. M. 1922. Physiological Studies of Effects of Formaldehyde on Wheat. Bot. Gaz. 74: 233-63.

3. Bakke, A.L., and Plagge, H.H. 1919. Studies on the Absorption and Germination of Wheat Treated With Formaldehyde. Ia. Acad. Sci. 26: 365-375.

4. Braun, Harry. 1920. Presoak Method of Seed Treatment. Jour. Agr. Res. 19: 363-392.

5. Cobb, Rodney D. 1958. Seed Germination After Fumigation With Methyl Bromide for Khapra Beetle Control. The Bulletin. Calif. Dept. Agr. Jan, Feb, March.

6. Cranefield, F. 1902. The Influence of Formaldehyde on the Germination of Oats. Wisc. Sta. Rept. 268-272.

7. Dorset, Marion. 1918. Some Common Disenfectants. U. S. D. A. Farmers Bull. 926.

8. Droste. 1916. Formaldehyde for Soaking Grain. Pharm. Zentralhalle. 57: 587-90. C. A. 11: 2340.

9. Horn, D. W., and Osol, A. 1929. Formaldehyde as a Fumigant. Amer. J. Pharm. 101: 741-778.

10. Hurd, Annie M. 1920. Injury to Seed Wheat Resulting from Drying After Disinfecting With Formaldehyde. Jour. Agr. Res. 20: 209-44.

11. Lewis, S. E. 1948. Inhibition of S H Enzymes by Methyl Bromide. Nature (London) 161 (4096)::692-693.

12. Lindgren, D. L., Vincent, L. E., and Krohne, H. E. 1955. Fumigation of Seed as Affecting Germination. News Leter No. 72. Univ. of Calif. Agr. Expt. Sta.

13. Lubatti, O. F., and Harrison, A. 1944. Determination of Fumigants XVIII. Comparison of The Sorption of Hydrogen Cyanide, Ethylene oxide, Trichloroacetonitrile, and Methyl Bromide by Wheat. Soc. Chem. Industry J. 63: 353-359.

14. Rabald, Erich. 1951. Corrosion Guide. 236-251 and 388-395.

15. Rowell, J. B., and Olien, C. R. 1957. Controlled Inoculation of Wheat Seedlings With Urediospores of Puccinia Graminis var. Tritici. Phytopath. 47: 650-655.

16. Skinner, J. J. 1913. Effect of Formalin and Bluestone Pickle on the Germination of Wheat. J. Dept. Agr. Victoria. 11: 473.

17. Stephens, F. L. 1908. Experiments Upon the Effect of Formalin on the Germination of Oats. N. C. Agric. Expt. Sta. Rept. 30-36.

18. Stewart, Robert, and Stephens, John. The Effect of Formalin on The Vitality of Seed Grain. Utah Expt. Sta. Bull. 108.

19. Walker, 1953. Formaldehyde. 2nd Ed.

20. Will, H., and Landtblom, F. O. 1919. Effect of Different Disinfecting Agents on Metals. Z. ges. Brauw. 42: 81-2. C.A. 14:11764.

APPENDIX

In order to get a rough estimate of the amount of carry-over of seed from one harvesting operation to another a sample of dirt, chaff, and seed taken from the combine used in this study was analysed. The sample was taken from two locations after the "rough cleaning" operation described in experimental methods in part II. Approximately one quart of material was taken from the horizontal ledge above and behind the cylinder housing and beater (designated as no. 4 position), and another pint was taken from under the auger in the bulk tank.

Chaff and some dirt were removed with a South Dakota blower, and several size ranges of seed were obtained by screening. An analysis of this material showed some seed from every crop harvested during the season. Since this was the first season for the combine there was no possibility of carry-over from previous years. Some sound seeds of each of the following crops were found: wheat, oats, barley, lupines, vetch, red clover, alfalfa, and trefoil. Weed seeds isolated from the samples are listed as follows:

1. Quack grass --------------- Agropyron repens
2. Redroot pigweeds ----------- Amaranthus sp.
3. Toothpick plant ----------- Ammi visnaga
4. Dog fennel ---------------- Anthemis cotula
5. Yellow rocket-------------- Barbarea vulgaris
6. Field mustard-------------- Brassica arvensis
7. Indian mustard------------- Brassica juncea
8. Black mustard ------------- Brassica nigra
9. Small-seeded false flax---- Camelina microcarpa
10. Shepards purse------------- Capsella bursa-pastoris
11. Mouse-eared chickweed------ Cerastium vulgatum
12. Hare's-ear mustard--------- Conringia orientalis
13. Ox-eye daisy -------------- Chrysanthemum leucanthemum

14. Wild carrot -------------------- *Daucus carota*
15. Crabgrass ---------------------- *Digitaria sanguinalis*
16. Barnyard grass ----------------- *Echinochloa crusgalli*
17. Wormseed mustard --------------- *Erysimum cheiranthoides*
18. Field peppergrass -------------- *Lepidium virginicum*
19. White cockle ------------------- *Lychnis alba*
20. Black medic -------------------- *Medicago lupulina*
21. Sweet clover ------------------- *Melilotus sp.*
22. Yellow oxalis ------------------ *Oxalis stricta*
23. Witch grass -------------------- *Panicum capillare*
24. Timothy ------------------------ *Phleum pratense*
25. Buckhorn plantain -------------- *Plantago lanceolata*
26. Common plantain ---------------- *Plantago major*
27. Rugels plantain ---------------- *Plantago rugelli*
28. Prostrate knotweed ------------- *Polygonum aviculare*
29. Wild buckwheat ----------------- *Polygonum convolvulus*
30. Lady's thumb ------------------- *Polygonum persicaria*
31. Purslane ----------------------- *Portulaca oleracea*
32. Cinquefoil --------------------- *Potentilla sp.*
33. Sheep sorrel ------------------- *Rumex acetosella*
34. Curled dock --------------------*Rumex crispus*
35. Yellow foxtail ----------------- *Setaria lutescens*
36. Green foxtail ------------------ *Setaria viridis*
37. Forked catchfly ---------------- *Silene dichotoma*
38. Night-flowering catchfly ------- *Silene noctiflora*
39. Tumbling mustard --------------- *Sisymbrium officinalis*
40. Horsenettle -------------------- *Solanum carolinense*
41. Common chickweed --------------- *Stellaria media*
42. Dandilion ---------------------- *Taraxacum sp.*
43. Pennycress --------------------- *Thlaspi arvense*
44. Alsike clover ------------------ *Trifolium hybridum*
45. White clover ------------------- *Trifolium repens*
46. Common mullein ----------------- *Verbascum thapsus*

A sample was taken from the bulk tank again after the first treatment was made. This material, which had been thoroughly soaked with formaldenyde was dried and seeds were again isolated. Germinations were run on some of the control and treated weed seeds. Germination methods used were those suggested by Everson (2), Steinbauer and Grigsby (4), Steinbauer *et al* (3), and *Rules for Testing Seeds* (1). Results of these germinations are listed in Table 1. Final readings were made at 30 days.

Numbers of weed seeds present varied from one seed of several kinds of weeds to over 5 grams of yellow rocket seed in one 10-gram sample analysed. This illustrates the efficiency and tremendous capacity of the combine as a weed carrier and spreader.

TABLE I

PERCENT GERMINATION OF SOME WEED SEEDS AS AFFECTED BY A 12-HOUR SOAKING PERIOD IN 37% FORMALDEHYDE

		Control	Treated
Amaranthus sp.	Red root pigweeds	61	3
Barbarea vulgaris	Yellow rocket	64	0
Chenopodium album	Lambsquarters	23	0
Echinochloa crusgalli	Barnyard grass	72	0
Digitaria sanguinalis	Crabgrass	44	0
Lychnis alba	White cockle	40	0
Panicum capillare	Switch grass	37	0
Phleum pratense	Timothy	49	0
Plantago lanceolata	Buckhorn plantain	50	0
Potentilla sp.	Cinquefoil	70	0
Rumex crispus	Curled dock	96	50
Setaria lutescens	Yellow foxtail	3	0
Setaria viridis	Green foxtail	18	0
Solanum carolinense	Horsenettle	26	0

REFERENCES:

(1) Anonymous, (1954), *Rules For Testing Seeds* - "Germination Tests", Proceedings of The Association of Official Seed Analysts, pp. 44-46.

(2) Everson, L. (1949), *Preliminary Studies to Establish Laboratory Methods for The Germination of Weed Seed*, *Proceedings of The Association of Official Seed Analysts*, pp. 84-89.

(3) Steinbauer, G. P., et al, (1955), *A Study of Methods For Obtaining Laboratory Germination of Certain Weed Seeds*, *Proceedings of The Association of Official Seed Analysts*, pp. 48-52.

(4) Steinbauer, G. P., and Grigsby, B., (1957), *Dormancy and Germination of the Seeds of Four Species of Plantago*, *Proceedings of The Association of Official Seed Analysts*, pp. 158-164.

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