

Table II. Recovery of Methoxychlor (P.P.M.)

Added	Recovered	Added	Recovered
0.050	0.050	0.100	0.095
	0.047		0.099
	0.065		
0.100	0.099	0.150	0.125
	0.080		0.140
	0.081		0.125

charring by the negative error due to incomplete recoveries. With samples containing over 0.1 p.p.m., the results reflect the true amount present with very

slight negative errors as the values increase. None of the values are corrected for variation in percentage of butter fat.

Methoxychlor is present in minute but detectable amounts in the milk of treated cows, and the concentration diminishes rapidly with successive samplings after spraying or dusting.

Acknowledgment

This investigation was conducted partially under research grants sponsored by Union Carbide Chemicals Corp. and E. I. du Pont de Nemours & Co., Inc., at Rutgers University. Edward

Gant and Fredi Jakob assisted in the collection and analysis of samples.

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Received for review May 8, 1957. Accepted December 5, 1957.

SEED DISINFECTION

Mechanism of Liquid Seed Treatment

Vapor Action and Adhesion, Radioactive Studies of Initial Liquid Distribution, and Investigations with Radioactive Panogen Formulations

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The mechanism of liquid seed treatment (Panogen process) was studied with physical and chemical methods. The mercurial distribution on treated seed is governed by the mixing process in the treater and by the vapor action of the fungicide. These processes were studied by means of volatile and nonvolatile tracers, and the distribution was characterized by statistical methods. High moisture content of the seed and poor mixing favor resorption which gives poorer initial distribution. A vapor pressure of 10^{-5} to 10^{-4} mm. of mercury gives sufficient vapor action to produce the final reasonably uniform distribution. In dust treatment, higher vapor pressures are required to give equivalent vapor action, as the effective surface of the dust is less than the seed surface treated with liquid. Panogen mercurials penetrate the fruit coat rapidly but diffusion stops at the endosperm. Liquid seed treatment may be improved further by reduction of the liquid volume.

FUNGICIDAL AND FUNGISTATIC AGENTS have been used for seed disinfection since the beginning of this century. Seed has been treated with solutions of formaldehyde or various dry or liquid copper and inorganic mercury preparations. The highly efficient organomercurial fungicides came into use at the end of World War I. Seed was soaked in a dilute aqueous solution of the mercurial and was dried in a subsequent operation. The necessity of drying was a great disadvantage, and the development work in this field during the twenties and thirties was therefore concerned mainly with this question.

The dry method, in which the seed was mixed with mercurial dust in special seed treaters, was introduced in the middle twenties. A few years later Gassner (7) developed the short wet treatment in which smaller amounts of liquid seed disinfectants were applied to the seed in a revolving drum. This method was superior in comparison with the dry method, especially as regards handling hazards. However, still comparatively

large liquid volumes were used, and the moisture content of the treated seed was increased to such a high level that the seed had to be sown within a couple of days after the treatment to prevent spontaneous heating of the stored grain.

Slurry treatment, a modification of short wet treatment—using slightly lower liquid dosages—was first performed with dusts in aqueous suspension but is now also used with true solutions.

Zade (27), working along similar lines as Gassner but independently of him, was able to develop a liquid seed disinfectant to be used in a much lower dosage—0.1 of the dosage in short wet treatment—so that the moisture content of the seed remained essentially unchanged. This disinfectant, Panogen, was put on the market in Sweden in 1938, where it received wide acceptance. In 1948, the Panogen process was introduced into the United States and Canada, and it has received much attention during the last few years.

Panogen has been subjected to worldwide testing for 20 years, with good re-

sults, experimentally and commercially. However, uniform distribution of the very small amounts of the liquid disinfectant used is still a problem as it was with the short wet treatment where much larger liquid volumes were involved.

The distribution problem has been discussed in the literature (15, 25) and, as late as in 1953, De Ong (10) remarked regarding Panogen that "even with the special applicator used for large scale work, there seems to be difficulty in securing a uniform distribution." The poor distribution of the dye used in Panogen to distinguish treated seed from non-treated seed is the main reason behind this and similar statements in the literature or in the field.

Ideally, just the necessary dose of the fungicide should be distributed over every site from which diseases may develop during germination, and the disinfectant should exhibit a specific vapor action in which the molecules of the fungicide are exclusively resorbed by fungi. As this scheme is not possible, uniform distribution over all sites from which plant

diseases may possibly develop when fungi are present is the next desirable goal. Uneven distribution results in spots with a high local concentration of fungicide, which does not give substantial gain in protection, and spots with less than the required amounts, which gives a limited amount of control. This necessitates an increase in the over-all amount of seed disinfectant used in order to give the deficient areas sufficient protection.

Zade, in his original development work, and others (7, 19) used biological techniques—i.e., agar-plate method—for the assay of mercurial on the individual kernels. The inhibition zones observed on the agar plate, however, are not primarily governed by the total mercurial content of the surface layer of the seed kernel. These zones indicate only the amount of fungicide which leaves the kernel and diffuses out in the surrounding medium.

To establish the distribution of fungicide directly in terms of concentration—i.e., the amount of fungicide per unit area of kernel surface—analytical methods with a sensitivity of 0.01 γ of mercury are required. This sensitivity can be obtained by the radioactive tracer methods described below. The radioactive determination does not affect the seeds; hence they can be subjected to biological tests after the nucleonic analysis.

The final distribution obtained in seed treatment depends on the outcome of the spreading and resorption processes. The spreading is accomplished in two phases: the initial mixing of the disinfectant with the seed in the seed treater and the vapor action of some of the slightly volatile mercurials, which are used as fungicides. These two mechanisms are conveniently studied by means of nonvolatile tracers and by tagged mercurials. The diffusion of the mercurial in the fruit coat of the seed can be studied also by means of tracer methods. Vapor action is directly observed by spectrophotometric determination of the mercurial concentration in the atmosphere surrounding the treated seed.

The seed treatment is for seed-borne and soil-borne diseases, and uniform distribution is essential in both cases. Though this investigation is concerned mainly with the physicochemical aspects of liquid seed treatment, a single biological test was performed with radioactive seeds to illustrate the relations between distribution and biological performance. This test had to be made with seed-borne diseases as soil infection is highly variable, while infection of stored seeds is fairly constant and reproducible over a period of time.

These studies were made using Swedish Panogen formulations and dosages. Moisture-content levels, 12 and 18%, of the seed tested were higher than the usual 9 and 15% found in the United States. However, this paper is concerned with the fundamental aspects of

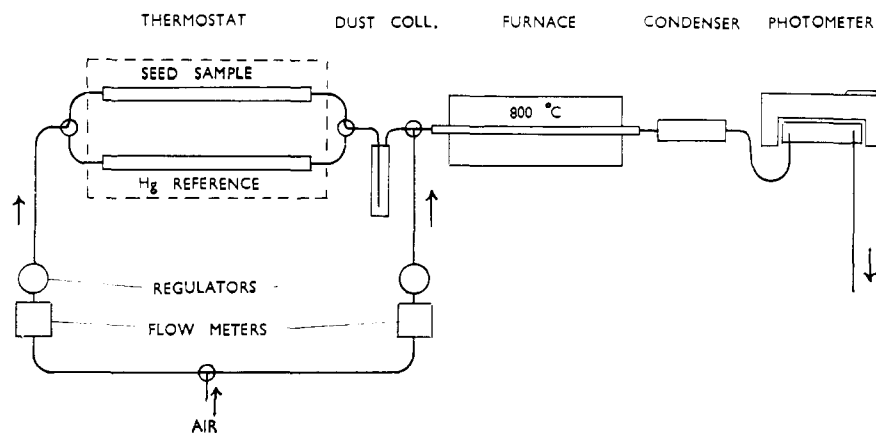


Figure 1. Setup for determination of mercurial vapor in atmosphere surrounding treated seeds

liquid seed treatment. Minor differences between practices and conditions in different countries have no influence on the general conclusions.

VAPOR ACTION AND ADHESION Materials and Methods

Seed-Treatment Procedure. Wheat seeds were treated with the following commercial disinfectants in the dosages recommended by the manufacturer: P₁, P₂, P₃, P₄, P₅, P₆, P₇, and P₈. (Explanations of symbols are in the Nomenclature section.) These treatments were performed in 100-gram batches in a laboratory seed treater (described later). The two seed moisture contents used, B₁ and B₂, were counted on the total weight of the seeds.

Mercurial Vapor Determination above Treated Seeds. The mercurial concentration in air in contact with treated seeds was determined by means of the setup shown in Figure 1. Air is passed through the seed sample, which is kept in a constant-temperature bath. The organic mercurial vapor is decomposed in the furnace and its concentration is then evaluated by means of the photometer (Kruger ultraviolet photometer Model 23, Harold Kruger Instruments, San Gabriel, Calif.), which was equipped with a special gas cell.

The sensitivity of this instrument is high. It has a dual range: 5 to 100 γ and 30 to 3000 γ of mercury per cubic meter. The instrument is provided with an internal standard for standardization and calibration. An external mercury vapor standard was also used as indicated in Figure 1. Air is passed through the tube with metallic mercury which is immersed in the constant temperature bath, diluted with extra air, and then passed through the furnace and the Kruger instrument. The vapor pressure of mercury at 25° C. corresponds to 19,900 γ of mercury per cubic meter. A dilution ratio of 1 to 20 was used to obtain a mercury concentration of about 1000 γ of mercury per cubic meter in the gas stream to be passed through the pho-

tometer. The photometer was checked frequently against this mercury standard and the deviation was always less than 15%.

In the experiments with treated seeds, undiluted gases were used, however, because of the low mercurial vapor concentration. The rate of flow through the cell was adjusted to 5.1 liters of air per minute. At lower rates of air circulation, errors were obtained, as was also reported by the manufacturer of the instrument.

Two slightly different techniques were used. In some experiments, the air stream was passed above the seed sample. The tube for the seed sample in Figure 1 was then substituted for a 250-ml. Erlenmeyer flask, with a 100-gram seed sample. The flask had an inlet and outlet for the air stream with the inlet 2 cm. above the seed. In other experiments, the setup was identical with that in Figure 1. Here air is passed through the tube (outside diameter, 3 cm.; length, 60 cm.) with the seed, which gives a good contact between the air stream and the seeds.

Mercury Analysis of Treated Seed and Wash Liquids. In the adhesion studies, the mercury contents of the dilute wash liquids were determined according to the procedure of Johansson and Uhrnell (14).

In the seed analysis, a slightly different digestion technique was utilized. Ten grams of seed were digested with 100 ml. of a 1 to 1 mixture of nitric acid-sulfuric acid under reflux, the outlet gases passing through a wash bottle with 40 ml. of concentrated sulfuric acid to take care of any evaporated mercury compounds. After complete digestion, 1 hour, nitrous gases were removed by boiling—the digestion flask and the wash bottle were processed separately. Any remaining nitrous gases were eliminated by boiling with potassium permanganate solution (4%) during 1 hour (100 ml. in the digestion flask, 50 ml. in the wash bottle). The mercury contents of the two solutions were determined sepa-

Table I. Evaporation and Diffusion of Mercury from Seed Kernels

Vapor Conc., $c_s, \gamma \text{ Hg/Cu. M.}$	Vapor Pressure, Mm. Hg	Time Required for Vapor Action		
		Evaporation I	Diffusion J	Diffusion, poor liquid distribution L
0.1	10^{-8}	1 hour	5 months	26 years
1	10^{-7}	6 min.	16 days	3 years
10	10^{-6}	37 sec.	2 days	96 days
100	10^{-5}	4 sec.	4 hours	10 days
1000	10^{-4}	0.4 sec.	24 min.	1 day
10,000	10^{-3}	0.04 sec.	2 min.	2 hours
100,000	10^{-2}	0.004 sec.	14 sec.	16 min.

rately by the procedure of Johansson and Uhrnell (14), after reduction of potassium permanganate with 15% hydrogen peroxide and finally, after adding a very small excess of potassium permanganate, by means of 0.5M oxalic acid solution. In general, no mercury was found in the wash bottle, but in some cases the presence of mercury justified the use of this tedious procedure.

Theory

Factors Governing Vapor Action.

Investigations by Army and Leben (2, 3, 4), Gassner (12), Pichler (20, 21), Purdy and Holton (23), and Lehman (17) have shown that some mercurial fungicides exhibit vapor action. Within the bulk of treated seed, evaporation and condensation of mercurial are simultaneously taking place from and onto the surfaces of the kernels. The rate of evaporation from a certain surface will be roughly proportional to the surface concentration of mercurial and the vapor pressure of the mercurial under the actual conditions. The actual vapor pressure of the mercurial is very much influenced by resorption processes and chemical reactions between the mercurial

and reactive compounds present in the fruit coat. The rate of condensation is governed mainly by the concentration of mercurial in the vapor phase. The net result of these processes is a transport of mercurial from rich to poor surfaces. When, and if, equilibrium is reached, rates of evaporation and condensation are equal.

The effective rate of evaporation is in general given by the kinetic equation:

$$dn(dt \times G) = e \times (p_s - p_a) / (2\pi \times m \times kT)^{1/2} \quad (1)$$

The effective rate is equal to the difference between the number of molecules evaporating from and the number of molecules condensing on the solid or liquid phase. The condensation coefficient, $e = 1$, means that every molecule, which impinges on the surface, will be captured (at least temporarily). This holds for liquids, and $e = 1$ is adopted in the present treatment.

To illustrate Equation 1 by a numerical example, consider a kernel, area = 0.6 sq. cm., covered with a thin layer of mercurial, total amount 0.77 γ of mercury, with the vapor pressure p_s mm. of mercury (corresponding to $c_s \gamma$ mercury per cubic meter) at 25° C. The molecular weight of the mercurial is assumed to be 400, and the actual vapor pressure in the atmosphere surrounding the kernel is put equal to half the saturation value. The time, I , which is required to decrease the mercury content of the kernel to half of its original value, the partial pressure of the mercurial remaining constant, has been calculated. Values for I are tabulated in Table I as a function of the corresponding saturation vapor pressures.

The rate of evaporation as indicated by the values for I is a very rapid process for all conceivable vapor pressures. Therefore, the rate-determining factor in vapor action seems to be the subsequent transport by diffusion and convection to deficient areas.

The mercurial diffuses from surface I to II (Figure 2). The mercurial vapor concentration near I is put equal to c_s , whereas the concentration in front of II is considered to be very small and accordingly the concentration gradient may be put equal to c_s per liter. The rate of transport by diffusion, d , from I to II

is governed by Fick's first law

$$d = (c_s/l) \times D \times G \quad (2)$$

where D is the diffusivity in molecules (of mercurial diffusing in air under the influence of unit concentration gradient) per square centimeter and per second. D is difficult to ascertain, but approximate calculations lead to the result that $D \sim 0.04$ for the molecular weight 400 and the collision diameter 7 A.

Consider in analogy with the above example the mass transfer from a kernel with a normal amount of mercurial to surrounding mercurial-free surfaces 0.1 cm. apart. By means of Equation 2, the time, J , is computed which is required to decrease the mercurial content of the kernel in question to half of its original value—i.e., from 0.77 to 0.38 γ of mercury. Values for J are shown in Table I. The transport by diffusion is, under the given set of conditions, very much less than the corresponding evaporation rate. Nevertheless the values show how mobile these minute amounts of substance are in spite of the very low vapor pressures.

To illustrate the conditions in case of extremely poor coverage, assume that the mercurial is to be distributed solely by evaporation from a few kernels—say three out of 100. Every "treated" kernel then carries, immediately after contact with the seed dressing, (100 per 3) times 0.77 γ or 25.6 γ of mercury, of which 24.8 γ have to be transported to surrounding kernels. As long as the kernels stay in the seed treater's drum, they are in continuous motion and the problem may be treated in the same simplified way as above—i.e., as a transport to deficient surfaces about 0.1 cm. apart. The time, L , is then required to reduce the mercurial content to the normal value (Table I). Under these conditions, very high vapor pressures are required if substantial equipartition is to take place within the drum (Table I). The time in the treating drum generally amounts to a few minutes or even less and this would require a vapor concentration of >1,000,000 γ of mercury per cubic meter or >10⁻¹ mm. of mercury.

After the seed has passed the treater, it is usually stored for some time. The few mercurial-rich kernels are then shielded from the majority of kernels, and the vapor has to travel some distance in order to find all deficient kernels. For the case of seed at perfect rest, the diffusion distance from a rich kernel to the farthest deficient kernel will probably amount to about 1 cm., and the values for L in Table I then have to be increased by a factor of 10.

These calculations demonstrate the importance of a reasonably good liquid distribution, which is needed in an efficient mixing procedure, as otherwise very high vapor pressures would be required to produce a uniform mercurial distribution.

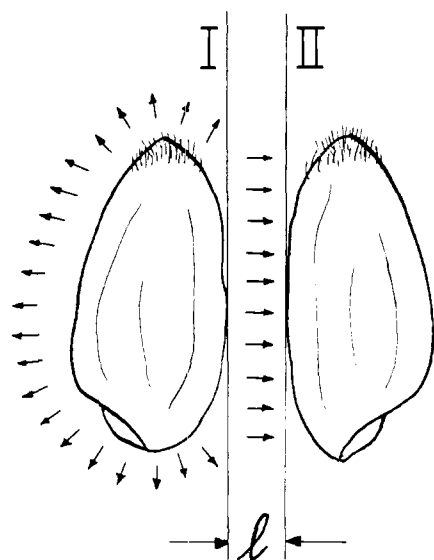


Figure 2. Schematic representation of vapor action. Mercurial is evaporating from left kernel and diffusing through planes I and II to kernel on right with no mercurial

Table II. Mercurial Vapor Concentration above Some Mercury Compounds at 20° C.

Compound	Physical State	Vapor Concn., c_s , γ Hg/m ³	Method of Determination
Cyano(methylmercuri)-guanidine	Crystalline, solid	270 (73)	"Transpiration" and distillation methods. Ionization vacuum gage. (Values later confirmed by nucleonic and spectrophotometric methods)
	Aq. solution, 0.8% Hg	100 (73)	Same as above
Methyl mercuric hydroxide	Crystalline, solid	10000 (73)	Same as above
	Aq. solution, 0.8% Hg	400 (73)	Same as above
Methyl mercuric chloride	Crystalline, solid	94000 (7)	Measurements of gas viscosity by vibrating quartz-fiber technique
Methyl mercuric bromide	Crystalline, solid	94000 (7)	Same as above
Methyl mercuric iodide	Crystalline, solid	90000 (7)	Same as above
Ethyl mercuric chloride	Crystalline, solid	8500 (7)	Same as above
Metallic mercury	Liquid	14140 (9)	"Transpiration" method
Mercuric chloride	Crystalline, solid	330 (76)	Not known
Phenyl mercuric acetate	Crystalline, solid	~10 (78)	Estimate from radioactive residue determinations

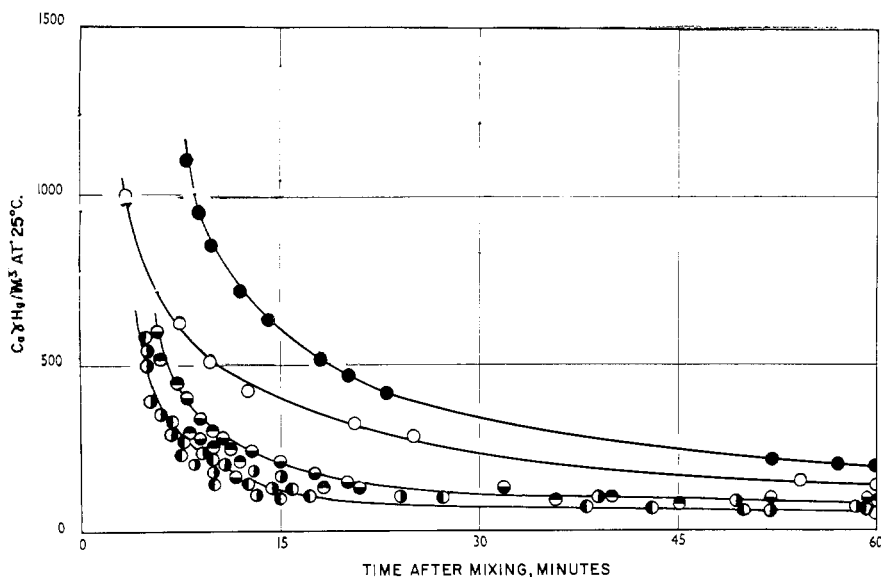


Figure 3. Mercurial in atmosphere above treated seeds as a function of time after seed treatment. Air stream passed above sample

● P₃P₁ ○ P₃P₂ ◐ P₁P₁ ◑ P₁B₂ ◒ P₂B₁ ◓ P₂B₂

When dusts are used, the kernel surface will carry a number of dust particles impregnated with the mercurial. The surface area of the dust effective in vapor action is smaller than the total treated surface area in case of liquid disinfectants. The "effective" surface area in case of a dust with a particle diameter as small as 2 microns probably amounts to only a few per cent of the kernel area and therefore the vapor action—i.e., the rate of mercurial transport to surrounding kernels—is reduced in a corresponding degree. (G is reduced in the expressions for evaporation and diffusion, Equations 1 and 2.) Vapor action is thus to be considered as a product of volatility and the effective surface area.

Vapor pressures for some mercurials are reported in Table II. Theden, Gerda, and Becker (24) demonstrated

that mercuric chloride used as a wood preservative exhibits a pronounced vapor action. The vapor pressure for mercuric chloride is similar to that for cyano(methylmercuri)guanidine.

Experimental Results

Mercurial Vapor above Treated Seeds. True saturation values for the vapor pressure of the mercurial above treated seeds can not be evaluated, as true equilibrium conditions are never obtained. Because of resorption, the vapor pressure will decrease continuously and at different rates on different parts of the kernels. A kind of dynamic equilibrium is set up which is shifted toward lower values for the observed mercurial in air concentration with time.

The mercurial concentration in air

above treated seeds was determined for various seed dressings by means of the described setup. The curves in Figure 3 refer to the following conditions: One hundred grams of wheat seeds were treated with 0.2 ml. of disinfectant and then transferred to the Erlenmeyer flask in the setup for the mercury vapor determination. The vapor pressure readings were then recorded. The data in Figure 3 refer to the three commercial liquid seed disinfectants: P₁, P₂, and P₃ at seed moisture contents B₁ and B₂.

Figure 3 shows a rapid decrease for the mercurial in air concentration, c_a γ of mercury per cubic meter, with time. This reduction is due to the resorption of mercurial by the kernels. The seeds are in a more reactive state at a high water content and therefore the decrease in c_a is more pronounced in these cases. No differences are observed between the two Panogen formulations, P₁ and P₂.

Some experiments were made to compare the dust preparation, P₄, known to exhibit a good vapor action with the liquid disinfectants, P₂ and P₃. Here a slightly different technique was utilized. The air stream was passed through a tube filled with the treated seeds, so that good contact between the seeds and the air stream was obtained (Figure 1). Higher values are obtained in this way (Figure 4) than in the experiments reported in Figure 3.

Five hundred grams of wheat with 12 to 13% moisture content were treated with the recommended amounts of disinfectants P₂, P₃, P₄, P₆, P₆, P₇, and P₈. The vapor action curves in Figure 5 show again a rapid decrease of vapor action with time. Each disinfectant exhibits an individual curve with respect to shape and position in the diagram.

Adhesion of Seed Disinfectants.

Adhesion between treated seeds and the seed dressing should be good in order to reduce the handling hazards and avoid losses of mercurial—e.g., in the sowing. The existence of vapor action may naturally cause doubts regarding residual action. The very rapid decrease with time in the vapor pressure above treated seeds indicates that residual action is developed rapidly.

Adhesion is most simply evaluated by determination of the amount of mercurial that can be removed from the seeds by subjecting them to a strong air stream or by rinsing them in water. The less mercurial that can be removed the better adhesion.

The experiment was planned as a Latin square with three factors on two levels: storage times, E₃ and E₄; disinfectants, P₃ and P₂; and moisture contents, B₁ and B₂. The Latin square design is represented by

$$\begin{matrix} E_3P_3B_1 & E_4P_3B_2 \\ E_3P_2B_2 & E_4P_2B_1 \end{matrix}$$

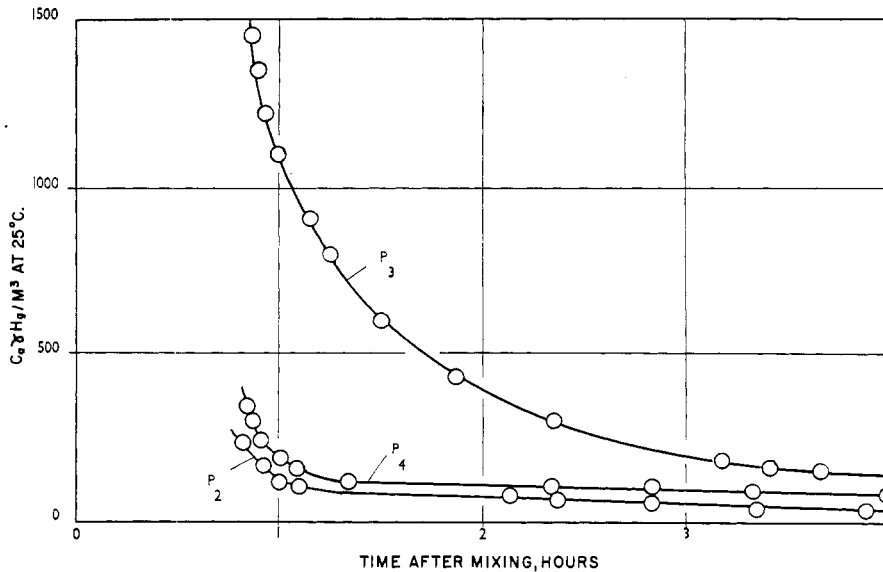


Figure 4. Mercurial in atmosphere above treated seeds as a function of time after seed treatment. Air stream passed through sample, which has normal moisture content

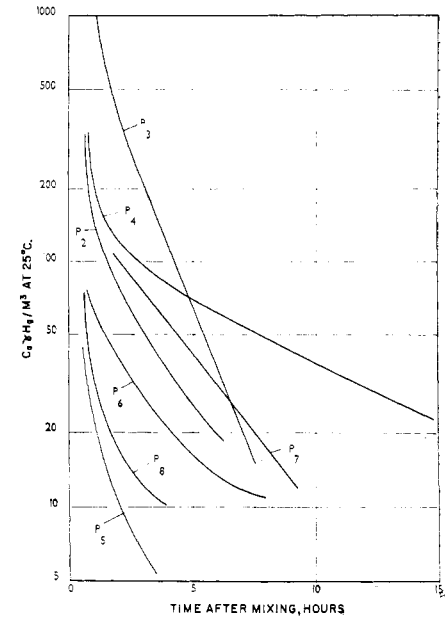


Figure 5. Mercurial in atmosphere above treated seeds as a function of time after seed treatment

Three methods to remove the mercurial were investigated: G_1 , air stream; G_2 , rinsing with water; and G_3 , air stream and rinsing. Mercury analyses were made on the seeds and on the rinsing water which was filtered and divided into a clear filtrate and a filtered-off portion.

Five hundred grams of spring wheat were treated according to P_8B_1 , P_8B_2 , P_2B_1 , and P_2B_2 . P_8B_1 and P_2B_2 were stored for 1 week after the treatment, E_3 , and were then subjected to the treatments denoted by G_1 , G_2 , and G_3 and analyzed. P_8B_2 and P_2B_1 were stored for 3 weeks more, E_4 , and were then subjected to the same procedures (G_1 , G_2 , and G_3).

Every 500-gram batch was divided into five 100-gram samples, which were kept and processed further in 250-gram glass bottles. Three samples were investigated according to the following scheme:

Air Stream (G_1). The glass bottle had an inlet and outlet for compressed air. The bottle was shaken in a shaking machine for 10 minutes, while a strong air stream was passing through the bottle. The seeds were then analyzed.

Rinsing (G_2). The glass bottle was filled with water. A free air space of 20 ml. was left. The sample was shaken for 10 minutes in the shaking machine, after which the rinsing water was immediately separated from the seeds and filtered. Seeds, residue on filter, and clear filtrate were analyzed.

Air Stream and Rinsing (G_3). Treatment according to G_1 and G_2 in this order. Seeds, residue on filter, and clear filtrate were analyzed.

The analytical results are given in Table III. No attempts were made to determine the amount of mercurial removed by air stream. The portion re-

Table III. Adhesion of Liquid and Dust Seed Disinfectants

Code	γ Hg/10 G. of Seeds			Total amount ^a found
	Residue on treated seeds	Mercurial in clear filtrate of wash	Mercurial in filter cake of wash	
EXPERIMENTAL RESULTS				
$E_3P_2B_2G_1$	130	131
$E_3P_8B_1G_1$	90	109
$E_3P_2B_2G_2$	100	0.5	1.3	102
$E_3P_8B_1G_2$	32	6	60	98
$E_3P_2B_1G_3$	126	0.4	0.7	128
$E_3P_8B_1G_3$	62	4	42	127
$E_4P_2B_1G_1$	90	91
$E_4P_8B_2G_1$	86	105
$E_4B_2B_1G_2$	62	3	0.6	66
$E_4P_8B_2G_2$	55	7	50	112
$E_4P_2B_1G_3$	143	1.0	4	149
$E_4P_8B_2G_3$	88	3	36	146
AVERAGED EFFECTS				
E_3	90	3	26	116
E_4	87	4	23	112
P_2	108	1	1	111
P_8	69	5	47	116
B_1	80	4	26	107
B_2	98	3	22	121
G_1	99	109
G_2	62	4	28	95
G_3	105	2	21	138

^a To G_1 and G_3 combinations have been added 0.5 γ Hg per 10 grams for P_2 and 19 γ Hg per 10 grams for P_8 , which figures constitute average loss of mercurial by air stream treatment G_1 alone.

moved by the air stream is, however, also removed by washing alone. The average differences between the washed off fraction in the G_2 and G_3 treatments for the P_2 and P_8 combinations therefore constitute the portion of mercurial that is removed by air stream. Total mercurial data in Table III take care of this portion, which does not show up in the reported analyses.

The amount of mercurial applied to the seeds was 160 γ of mercury per 10 grams. The differences between "added and found" are due to residues on the walls of the bottle in which the treat-

Table IV. Summary of Adhesion of Liquid and Dust Disinfectants

	Nonadherent Portion, % ^a		Adherent Portion, %
	Air Stream, (G ₁)	Washed off, (G ₂)	
Panogen liquid ($P_2E_{3,4}B_{1,2}$)	0.5	3	97
Commercial dust ($P_8E_{3,4}B_{1,2}$)	16	53	47

^a Portion removed by air stream, also removed by washing ($G_2 + G_3 = 100\%$).

ment was performed. Analytical errors are also partly responsible. (A detailed calculation of the standard deviation, σ , to be expected for the total mercurial data gives the value $\sigma = 20 \gamma$ of mercury per 10 grams, which compares well with the observed value $\sigma = 24 \gamma$ of mercury per 10 grams.)

The data in Table III reflect a fundamental difference between dust and liquid disinfectants. Adhesion, poor in the case of dust, is very good with liquid treatment (summary, Table IV).

Discussion

Theoretical considerations showed that a vapor concentration of a few hundred micrograms of mercury per cubic meter is likely to produce a sufficiently good vapor action. This vapor concentration corresponds to an extremely low vapor pressure, about 10^{-5} mm. of mercury, which is still able to move the mercurial and distribute it homogeneously in a matter of hours.

However, a reasonably good initial liquid distribution is necessary; otherwise very high vapor pressures would be required to produce a good distribution.

Vapor action is to be considered as a product of the volatility of the disinfectant and the effective surface area. In case of dusts, as opposed to liquid treatment, a smaller effective surface is involved which would give a less favorable vapor action unless the mercurial has a higher vapor pressure.

The mercurial vapor pressure above treated seeds decreases rapidly with time, because of resorption. This process is faster at a higher moisture content. Within a few hours, low vapor pressures have been reached. Theoretical calculations, however, show that for vapor concentrations between 100 to 1000 γ of mercury per cubic meter, the equilibrating process should be essentially complete in a matter of hours. Vapor action will thus in general have sufficient time to do its job before the vapor pressure has been reduced to too low levels.

Only the unsubstituted alkyl mercurials seem to exhibit sufficiently high vapor pressures to produce the desirable fast vapor action (Figure 5).

There is a surprisingly good adhesion of the mercurial observed for seeds treated in the liquid process. The disinfectant is rapidly taken up by the seeds and cannot then be removed by washing or blowing with air. The mercurial of the dust disinfectant is firmly adsorbed on the dust carrier, part of which is easily removed from the seed.

RADIOACTIVE STUDIES OF INITIAL LIQUID DISTRIBUTION

Materials and Methods

Radioactive Nonvolatile Seed Dressing. An aqueous solution, A, with 5.2% thallium nitrate tagged with thallium-204 was used to track the

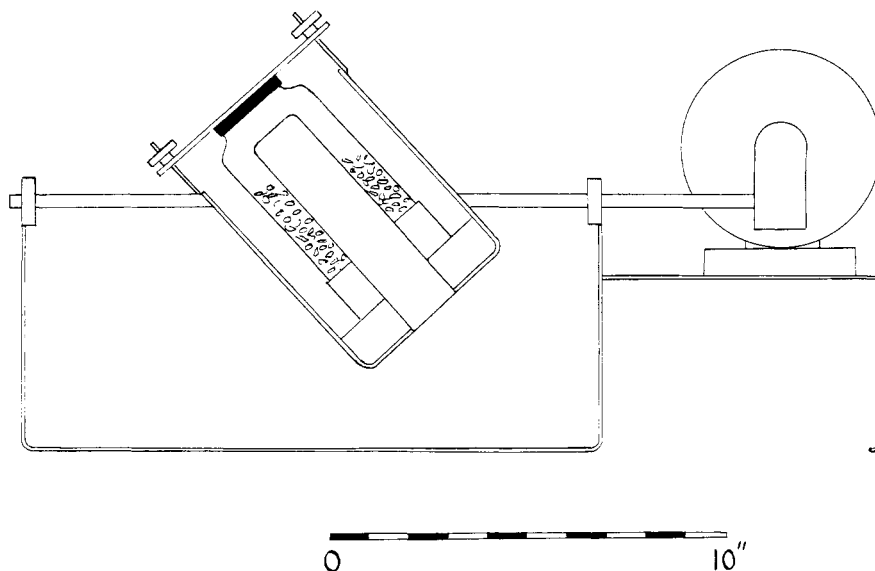


Figure 6. Equipment for seed treatment in laboratory. Inclined 500-ml. bottle is revolved at 40 r.p.m.

distribution of the liquid carrier in the process of seed treatment. Thallium nitrate is nonvolatile and its distribution would therefore be representative for nonvolatile fungicides with no vapor action. Thallium-204 is a strong beta emitter with the beta energy, 0.78 m.e.v., and the half life, 2.7 years (8). The normal dosage of 0.2 ml. per 100 grams of wheat gave an average activity of 850 c.p.m. per kernel.

Seeds. Several different seeds were tested: wheat, rye, oats, and sugar beets. The main investigations were, however, performed with wheat seeds. Mechanically damaged seeds were removed after visual examination. Two levels of the moisture content were used: B₁ and B₂.

Seed Treatment Procedures. Treatments were performed batchwise in the laboratory machine (Figure 6). The glass bottle is charged with 100 grams of seed plus the actual amount of disinfectant and is then immediately clamped into position in the machine and revolved for 4 minutes at 40 r.p.m. This will give the "rolling" movement producing the desired friction between the kernels. The friction forces depend, however, not only on the relative movement between the kernel surfaces, but also on the acting pressure. In full scale equipment, where large amounts of seeds are handled, this pressure is much larger than in this laboratory machine. Friction conditions are therefore more favorable in machine treatment, which is compensated for by the longer treatment time in the laboratory procedure. Army (5) has recently compared the effectiveness of machine and laboratory treatment; he found no essential differences in case of the standard materials Panogen and Ceresan M.

Two different techniques were used for the mode of addition of the disinfect-

ant: C₁, liquid poured directly on seeds—with mixing after about 10 seconds—and C₂, liquid poured on walls of the bottle and transferred to the seeds via the wall. C₁ constitutes an unfavorable mode of addition, as only a few kernels are soaked by the disinfectant. C₂ corresponds to treatment in modern commercial treaters, where the disinfectant is added via a spreader device.

The rolling type of seed movement in the treater was compared with "shaker mixing" in some experiments. These conditions are characterized by the symbols: D₁, 100 grams of seed treated 4 minutes in the laboratory machine, and D₂, 500 grams of seed vigorously shaken by hand in a 1000-ml. Erlenmeyer flask for 4 minutes.

Nucleonic Technique. The radioactive seed kernels were "counted"—i.e., their radioactivity evaluated—in standard equipment from Tracerlab, Inc., Boston, Mass. (Tracerlab SC-19 Utility Scaler, SC-90 Shielded Manual Sample Changer, and Geiger tube TGC-2 with the window thickness 1.9 mg. per sq. cm.). The setup was frequently checked by means of a cobalt-60 reference. Variations in geometry and counting efficiency were less than 2% over the experimental period. The seed kernel was placed between two thin sheets of Pliofilm and clamped into position on a sample disk by means of an outer ring (Tracerlab E-7A brass rings and disks) as shown in Figure 7. The kernel was placed in the middle of the disk with, in case of wheat seed, the furrow of the kernel facing the window of the counter tube. Beta rays emitted from the upper surface of the kernel are counted by the Geiger tube. Back-scattering does not influence the results to an appreciable degree.

Seed kernels were painted on one side with radioactive disinfectant. The ker-

Table V. Activities from Wheat Kernels with Tagged Mercurial on One Side Only

Location of Mercurial	Position of Furrow in Counting	Observed Activity, C.P.M.	Ratio
Furrow side	Down	54	0.012
	Up	4322	
	Down	67	
	Up	2292	
Back side	Down	18	0.006
	Up	2816	
	Up	340	0.091
	Down	3730	
	Up	346	0.079
	Down	4403	
	Up	69	0.046
	Down	1506	

nels were then counted in different positions on the sample disk. Table V shows that when the radioactive spot is turned down, the registered activity is only a small percentage of the activity observed with the active spot turned up.

The thickness of the Pliofilm cover amounted to 4.2 mg. per sq. cm. This film is used to eliminate radioactive contamination and also to keep the kernel in position during the measurement.

An exact value for the counting geometry cannot be calculated, as the seed kernel is not a defined geometrical entity. The surface area projected by the seed kernel on a close contact plane as indicated in Figure 7 was used for a rough estimate. If a photographic plate is put in this plane, the radioactive kernel produces a picture of itself on the plate. (This technique is described in more detail later.) The average projected surface area of the wheat kernels, determined by autoradiography, amounts to 0.1 sq. cm. The total macroscopic surface area of these wheat kernels is estimated to be 0.6 sq. cm.—kernel areas computed under the assumption that they constitute ellipsoids. The geometry involved is then calculated to give a counting efficiency of about 15%—i.e., 15% of the radiation emitted from the projected surface indicated in Figure 7 reaches the window of the Geiger tube.

Theory

Distribution Parameters. The number of counts obtained in a measurement is the sum of the activity from the radioactive kernel and the background activity. Subtraction of the background count gives the activity associated with the seed kernel. The set of, in general, 100 activities obtained in this way constitutes the primary material for evaluation of the distribution parameters, which characterize the effectiveness of the seed treatment procedure.

The histogram (Figure 8) has the shape of a normal distribution curve. A few kernels, however, carry higher activities than observed in the main group of kernels. Inspection of the diagram reveals that the borderline between these two

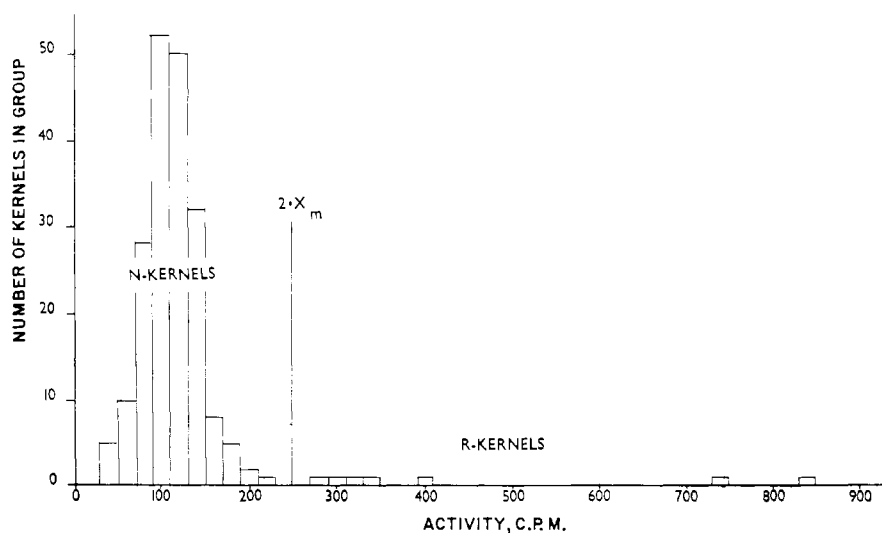


Figure 8. Histogram of kernel activities

populations is roughly twice the average kernel activity (designated by $2 \times X_m$).

The first step in the analysis of the experimental data is to separate these two populations. The borderline is always in the neighborhood of $2 \times X_m$. Differences between the two populations calculated by a more strict statistical procedure give but slightly differing borderlines from case to case.

The two populations will be characterized as R-kernels and N-kernels, with the latter referring to the main group of normal kernels. The group of R-kernels, only a small percentage of the total, have resorbed slightly larger quantities of fungicide because of mechanical damage or too long contact with a large quantity of the seed disinfectant.

The number of R-kernels divided by the total number of kernels is called the R-factor. The amount of mercurial which is carried by the R-kernels in excess of the mean value for the N-kernels is used uneconomically. This quantity divided by the total amount of mercurial is the S-factor. The T-factor is introduced as $T = S/R + 1$. The N-kernel population is characterized by its average and by its coefficient of variation.

The observed activity in a single meas-

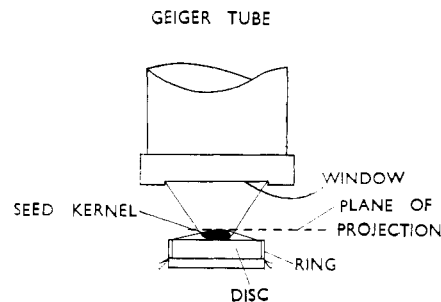


Figure 7. Radioactive counting on a treated seed kernel

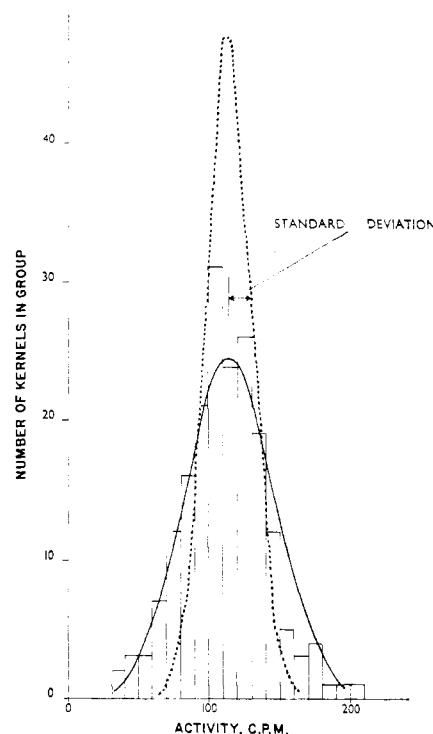


Figure 9. N-kernel distribution curve. Dotted curve shows distribution after elimination of some experimental errors present in the primary material represented by the histogram (Equation 5)

urement, X_i , is subject to several sources of variation. The kernel count may be written as

$$X_i = G \times Q \times q \times c \times K \times t \quad (3)$$

where

Q = γ of mercury per square centimeter, average concentration of mercurial on the surface G

q = over-all counting efficiency

c = millicuries per microgram of mercury, the specific activity of the mercurial

K = counts per minute per millicurie, conversion factor

t = minutes, counting time

The standard deviations associated with the various factors in the above expression are denoted by the sign σ . The coefficient of variation (σ_X/X) is governed by

$$(\sigma_X/X)^2 = (\sigma_G/G)^2 + (\sigma_Q/Q)^2 + (\sigma_q/q)^2 + (\sigma_c/c)^2 + (\sigma_K/K)^2 + (\sigma_t/t)^2 \quad (4)$$

The error due to the statistical fluctuations in the radioactive decay is the term $(\sigma_K/K)^2$. All other factors kept constant, $X = \text{constant} \times K$; and $\sigma_X = \text{constant} \times \sigma_K$; where σ_X here stands for the standard deviation due to counting statistics. Let b (in counts per minute) denote the background activity and σ_X may be written as $\sigma_X = \sqrt{X + 2bt}$, which yields $(\sigma_K/K)^2 = (X + 2bt)/X^2$ to be introduced in (4). The term which characterizes the mercurial distribution is isolated.

$$(\sigma_Q/Q) = \sqrt{(\sigma_X/X)^2 - [(\sigma_G/G)^2 + (\sigma_q/q)^2 + (\sigma_c/c)^2 + (X + 2 \times b \times t)/X^2]} \quad (5)$$

The quantity (σ_Q/Q) is termed the spreading error as it constitutes a numerical measure of the lack of uniform distribution. Confidence limits for (σ_Q/Q) are estimated by means of the standard deviation

$$\sigma(\sigma_Q/Q) = (\sigma_X/X)/\sqrt{2N} \quad (6)$$

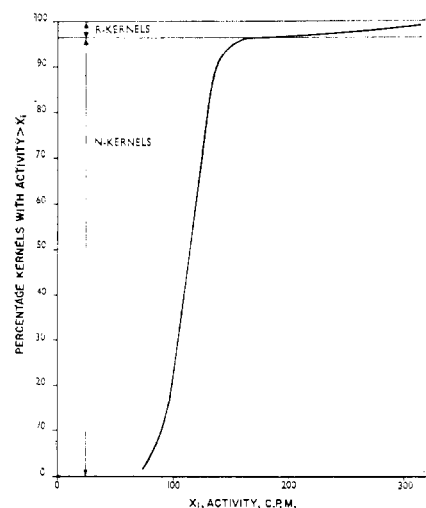


Figure 10. Integrated distribution curve for distribution parameters: spreading error $(\sigma_Q/Q) = 0.16$; $R = 0.035$; $S = 0.10$; and $T = 3.8$

where N is the number of analyzed kernels. Confidence limits of 95% are used throughout in this report. The terms which make up for the spreading error will be discussed separately. The term (σ_X/X) is evaluated from the experimental results in the usual way.

The dosage of fungitoxicant is preferably reported as micrograms of mercury per square centimeter of treated surface and not on a weight basis—e.g., parts per million of mercury. Seed treatment resembles spraying, as only the surface layer (the fruit coat) is treated.

The term (σ_G/G) takes care of the variation in kernel surface area. Assuming that the kernels are of an identical shape, (σ_G/G) will be two thirds of the coefficient for variation of kernel weight.

The geometrical error, σ_q/q , accounts for changes in the geometry of the equipment, variations in the transmission of the cover, and variations in beta absorption between different kernels. This term should also take care of the variations in shape besides the surface area variation accounted for by (σ_G/G) . The magnitude of (σ_q/q) cannot be calculated but it must be comparatively small.

The counting time was about 1 minute. The accuracy is estimated to be about 0.02 minutes and accordingly the term $(\sigma_t/t)^2$ is of no importance compared to $(\sigma_G/G)^2$. The last term under the square root sign of Equation 5 makes up for counting statistics.

The N-kernel population in Figure 8 has been subject to the above analysis.

In Figure 9, the histogram and the associated distribution curve constitute the primary experimental material, whereas the dotted curve shows the distribution obtained after elimination of some of the errors according to Equation 5. Figure 10 shows this material in integrated form.

All distribution measurements were performed with the furrow of the wheat kernel facing the window of the Geiger counter tube. That the distribution pattern is approximately the same whether the furrow is up or down was found in a separate experiment. Treated kernels (radioactive Panogen formulation P₉ described later) were picked up at random and counted on both sides. The results are reported in Table VI. "Furrow up" gives a slightly better distribution and a somewhat higher average count. The differences, however, are

small and not significant. The differences observed between furrow up and furrow down for each separate kernel were subjected to a statistical analysis. The mean square observed for the set of furrow up minus furrow down activities minus the observed over-all difference, 84 to 78 c.p.m. for the N-kernels, was calculated as 314 (c.p.m.)². This mean square is compared with the variation to be expected because of counting statistics, if the mercurial activities on each side had been identical. This gives $F = 314/279 = 1.13$ which is not significant ($F_{0.05[100,100]} = 1.39$). Therefore, the differences between the furrow up and furrow down activities in this experiment may be attributed to counting statistics.

A kernel with slightly higher furrow up activity thus exhibits a higher furrow down activity. This is not to be expected for R-kernels which may carry more mercurial on localized spots.

Results and Discussions

Initial Liquid Distribution in Seed Treatment. The final distribution obtained in seed treatment is a result of combined liquid spreading and vapor action. The liquid spreading necessarily takes place during the mixing in the drum of the seed treater, whereas vapor action also acts during the subsequent storage.

The distribution pattern obtained with nonvolatile compounds is likely to characterize the initial liquid distribution and how this is affected by various factors. Experiments on this were run with the radioactive thallium nitrate solution, A.

A 2 × 2 factor design was used to evaluate the effect of the moisture content and the mode of addition of the disinfectant. The addition of the liquid and the handling of the bottle during the very first seconds before the seed treater is started have a great influence on the final result—especially when no vapor action is involved. Two blocks were therefore run with two operators, F₁ and F₂ (Table VII). The averaged effects reported represent the average results for all combinations containing B₁, B₂, C₁, C₂, F₁, and F₂, respectively. R, S, and T are reported as arithmetic averages. The average spreading error, σ_Q/Q , is calculated as the square root of the mean square.

The rather large differences observed between the two operators show how

Table VI. Influence of Position of Furrow of Wheat in Nucleonic Distribution Measurements

	Distribution Parameters			Average Activity	
	R	S	T	X_m	X_m'
Furrow up	0.00	0.00	...	84	84
Furrow down	0.01	0.01	2.0	79	78

critical the very first moments of the mixing process are. Because of this, all subsequent work has been done by one operator, F_2 .

The higher moisture content level, B_2 , gives a less favorable distribution. That obtained is better than might be expected as every kernel received a portion of the dressing.

To prove that no vapor action was in play, the following simple check was run. Untreated seeds were placed on top of a layer of treated seeds but were kept separated from the treated seeds by means of a grid, through which any emitted vapors could freely pass. After 20 hours storage, there was no activity on the test seeds in case of A, whereas in case of radioactive Panogen, the test seeds had reached 22% of the activity for the treated seeds, owing to vapor action.

A Latin square was run to evaluate the influences exerted by the type of motion in the seed treater; the mode of addition of the disinfectant; and the dosage. A_1 constitutes the normal dosage and A_2 , 0.1 of normal dosage. Results are given in Table VIII.

This Latin square design was run in duplicate and the variations between the replicates is rather small. There is a strong response for the mode of addition and also for the type of mixing in the seed treater. The low dosage seems to give at least as good distribution as the normal dosage. The dosage can be reduced to 0.1 of the normal dosage with no harm on the liquid distribution in this particular case (Table IX) because the solute, thallium nitrate, is not rapidly resorbed by the kernels and is therefore easily transferred from kernel to kernel during the mixing.

Wheat, one of the most important crops subject to seed treatment, was the seed most used in this study. In Table X distribution parameters are reported for some other seeds. The results are much the same as observed for wheat. Sugar beets, however, seem to give a poorer distribution of the nonvolatile thallium nitrate, because of the very irregular shape of these seeds.

INVESTIGATIONS WITH RADIOACTIVE PANOGEN FORMULATIONS

Materials and Methods

Radioactive Panogen Formulations.

Two radioactive liquid seed dressings were used in this study, P_2 and P_3 . P_2 is characteristic of the Panogen formulations in use today in different countries. Concentration and dosage vary slightly from country to country, but the amount of mercurial applied to the seed is about the same. In Sweden, a dosage of 200 ml. of Panogen with 0.8% mercury per 100 kg. of wheat is recommended, which is the normal dosage used in the present study. This recommendation gives a

Table VII. Distribution of Nonvolatile Thallium Nitrate Solution on Wheat as a Function of Moisture Content, Mode of Addition, and Operator

Code	Distribution Parameters			
	R	S	T	(σ_Q/Q)
EXPERIMENTAL RESULTS				
$A_1B_1C_1F_1$	0.06	0.14	3.3	0.46 ± 0.07
$A_1B_1C_2F_1$	0.03	0.06	3.0	0.39 ± 0.05
$A_1B_2C_1F_1$	0.07	0.30	5.3	0.64 ± 0.09
$A_1B_2C_2F_1$	0.09	0.31	5.0	0.68 ± 0.09
$A_1B_1C_1F_2$	0.07	0.15	3.1	0.39 ± 0.06
$A_1B_1C_2F_2$	0.06	0.08	2.3	0.27 ± 0.04
$A_1B_2C_1F_2$	0.06	0.11	2.8	0.44 ± 0.06
$A_1B_2C_2F_2$	0.07	0.16	3.3	0.37 ± 0.06
AVERAGED EFFECTS				
B_1	0.055	0.11	2.9	0.38
B_2	0.073	0.22	4.1	0.55
C_1	0.065	0.18	3.6	0.49
C_2	0.063	0.15	3.4	0.45
F_1	0.063	0.20	4.2	0.56
F_2	0.065	0.13	2.9	0.37

Table VIII. Distribution of Nonvolatile Thallium Nitrate Solution on Wheat as a Function of Dosage, Mode of Addition, and Type of Movement

Code ^a	Distribution Parameters			
	R	S	T	(σ_Q/Q)
EXPERIMENTAL RESULTS				
$A_1C_1D_2$	0.09	0.24	2.7	0.75 ± 0.10
$A_1C_2D_1$	0.12	0.22	2.8	0.68 ± 0.09
	0.00	0.00	...	0.19 ± 0.03
$A_2C_1D_1$	0.02	0.03	2.5	0.17 ± 0.03
	0.03	0.05	2.7	0.26 ± 0.04
$A_2C_2D_2$	0.03	0.05	2.7	0.26 ± 0.04
	0.05	0.20	5.0	0.34 ± 0.05
	0.05	0.15	4.0	0.33 ± 0.05
AVERAGED EFFECTS				
A_1	0.06	0.12	2.7	0.52
A_2	0.04	0.11	3.6	0.30
C_1	0.07	0.14	2.7	0.54
C_2	0.03	0.09	3.8	0.27
D_1	0.02	0.03	2.6	0.22
D_2	0.08	0.20	3.9	0.56

^a A_1 , 2 ml. and A_2 , 0.2 ml.

Table IX. Distribution of Nonvolatile Thallium Nitrate Solution as a Function of Dosage in Normal Seed Treatment ($B_1C_2D_1$) of Wheat

Dosage, Ml./Kg. of Wheat	Distribution Parameters			
	R	S	T	(σ_Q/Q)
10	0.00	0.00	...	0.25 ± 0.04
5	0.05	0.11	3.2	0.46 ± 0.08
2	0.034	0.08	3.3	0.33 ± 0.06
1	0.03	0.05	2.7	0.42 ± 0.06
0.5	0.05	0.11	3.2	0.46 ± 0.07
0.2	0.02	0.03	2.5	0.20 ± 0.04

Table X. Distribution of Nonvolatile Thallium Nitrate Solution in Normal Seed Treatment of Different Seeds ($B_1C_2D_1$)

Seed	Distribution Parameters			
	R	S	T	(σ_Q/Q)
Wheat	0.03	0.08	3.3	0.33 ± 0.06
Oats	0.02	0.02	2.0	0.28 ± 0.04
Rye	0.02	0.05	3.5	0.28 ± 0.05
Sugar beets	0.04	0.20	6.0	0.46 ± 0.08

very good disinfection of even highly infested seed. There is, however, a tendency among the growers to reduce the dosage in case seeds are known to be only slightly infested.

Formulation P_3 is 10 times more concentrated than P_2 and was used in 0.1 of the normal dosage or 20 ml. per 100

kg. of wheat seed. The same solvents were used as in the commercial product. The radioactive mercurials were synthesized by the method used in the industrial manufacture of these compounds.

The mercury isotope mercury-203 emits 0.21 m.e.v. beta and 0.29 m.e.v. gamma—24% of the gamma radiation

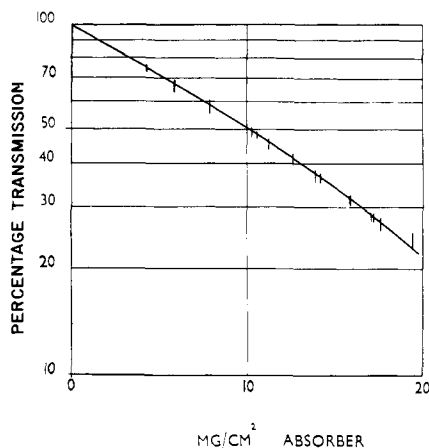


Figure 11. Beta absorption curve valid for conditions of distribution measurements

is transformed to internal conversion electrons (8). Values for the half life of mercury-203 mentioned in the literature vary considerably and, therefore, were checked by activity determinations on reference samples over an extended period of time. The value obtained, 47.5 days, is in harmony with some recent values: 47.9 (6) and 45.9 ± 0.5 days (26). The specific activity of P_2 was estimated to be 8.2 mc. per gram of mercury and of P_9 7.4 mc. per gram of mercury.

The beta radiation from mercury-203 is rather weak and is absorbed by a thin layer of material. An absorption curve which is valid for the conditions of these experiments is reproduced in Figure 11. The transmission of the 4.2 mg. per sq. cm. Pliofilm cover used in the distribution measurement is 76%. Part of the beta radiation is also absorbed in the surface layer of the seed kernels in case the mercurial is situated deep in the fruit coat. This feature will be used to estimate the penetration of the mercurial.

P_2 is water soluble to the extent of 21.7 grams of mercury per liter at room temperature (22), whereas P_9 can form highly concentrated aqueous solutions. The hydrophilic character of P_2 makes possible the use of nonphytotoxic solvents with good spreading qualities—among them, water. However, of course, the most important quality of P_2 and of P_9 is the high fungicidal activity combined with a low inherent phytotoxicity.

Besides the fungicide and the solvents, the commercial formulations usually contain a fluorescent dye and stabilizing agents, which were included in the radioactive formulations.

The dye used in Panogen is most rapidly adsorbed during the first seconds of the mixing process. This results in the slightly "patchy" look of the treated seeds. A poor dye distribution is, however, to be preferred, as the treated seeds are then more easily recognized. Because of this chromatographic effect,

the dye distribution is not representative of either the mercurial distribution or the initial liquid distribution.

Seeds. The wheat seeds were heavily infested by storage fungi and *Fusarium* species (*F. nivale*, *F. culmorum*, and *F. avenaceum*). Part of the *Fusarium* seemed to be situated deeply. Mold development due to storage fungi was severe after prolonged storage at the high moisture-content level. These storage conditions are not met with in practice. Untreated seeds germinated to only 8%. Two levels of the moisture content were used, B_1 and B_2 .

Treatment and Storage of Seeds. The seeds were treated by the technique described under Radioactive Studies of Initial Liquid Distribution and the same two different techniques were used for the addition of the disinfectant: C_1 and C_2 .

A certain storage time is believed to be required to get the full benefits of the vapor action. However, the desired storage time in case of the Panogen formulations studied was short—a few hours were sufficient. Four different storage times were used: E_1 , E_2 , E_3 , and E_4 . The seeds were stored in glass bottles at room temperature ($20-21^\circ\text{C}$.) in darkness. The bottles were completely filled with seed. No losses of mercurial could take place during the storage.

Germination Tests. The seeds, analyzed by the nucleonic distribution method, were subjected to a subsequent germination test. They were transferred immediately after the radioactive determination into the germination dish. Twenty-five kernels were planted in each dish in known positions. For this test, the standard method of the biological laboratory of AB Casco, Stockholm, Sweden, was used for evaluation of the protection against *Fusarium* fungus.

The 5×5 inch enamel dish is filled with 250 ml. of coarse crushed brick, previously washed and sterilized. The seed kernels are scattered on top of this brick dust layer. Water, 48 ml., is then added and the seeds are covered by an additional layer in a cold box at $10-11^\circ\text{C}$. After 3 days, the covers are removed because of the height of the seedlings. The sprouts are examined after 13 days. In general, only the amount of *Fusarium*-infected sprouts are noted but, in these special experiments, additional observations were made. The seeds were characterized as dead, infested with mold fungi, infested with *Fusarium*, cripples, over-treated, or healthy. The lengths of root and straw were measured for each separate seedling.

Autoradiographic Technique. The autoradiographs shown in this paper were processed in the following way.

The film (Ilford, Type C, industrial

x-ray film, size $11\frac{3}{4} \times 15\frac{3}{4}$ inches), was put in a tray still in its envelope. Seed kernels (1000) were spread out on the tube side of the film. Fine sand was spread over the kernels so that they were fixed on the surface of the envelope and pressed toward the film. The sand layer also took care of any mercurial vapors emitted from the kernels. The following empirical formula was used to calculate the necessary time of exposure, H .

$$H = \frac{G}{\bar{X}_m} \times 10^4 \text{ days}$$

The film was developed in Agfa-30 x-ray developer for 8 to 10 minutes at 20°C . After a water rinse, the film was fixed in Kodak F-5 for 20 minutes.

Results and Discussions

Distribution of Mercurial Fungicides. The vapor action associated with the volatile mercurial fungicides improves the initial distribution and may thus compensate in case of a less efficient mixing process. A most important function of vapor action, however, which does not show up in the distribution measurements is the ability of the vaporized mercurial molecules to reach deeply situated fungi.

A large number of measurements with Panogen formulations have demonstrated that the mercurial distribution is uniform and thus in harmony with biological and agricultural tests reported elsewhere. The distribution parameters for wheat under normal conditions are $R = 0.01$ to 0.02 , $S = 0.02$ to 0.04 , and the spreading error (σ_q/Q) ≤ 0.2 . Mercurial, 96 to 98%, is therefore used in the most economic way. The spreading error, Equation 5, contains some unknown errors, mainly (σ_q/q), which could not be estimated and are included in (σ_q/Q). Therefore, the actual mercurial distribution necessarily must be still better than is realized from the magnitude of (σ_q/Q).

In studies the experimental conditions were varied according to the principle "adding a little or a lot." The treatments were thus exaggerated—conditions in some cases much more severe than met with in the field—in order to get a more significant response. A Latin square was utilized according to

$$\frac{A_1B_1C_2 | A_2B_1C_1}{A_1B_2C_1 | A_2B_2C_2} \times E_1, E_2, E_3, E_4$$

where the effects of the dosage, moisture content, mode of addition, and storage time are studied. All experiments were run in duplicate and a total of $4 \times 8 \times 100 = 3200$ kernels were analyzed. The analyzed kernels were subjected to a germination test to investigate the emergence as a function of mercury content and distribution parameters.

The penetration into the kernels was followed by means of absorption measurements. The loss of mercurial from

Table XI. Distribution, Penetration, and Residual Activity for Wheat Seeds Treated with Panogen Formulations as Functions of the Dosage, the Moisture Content of the Seed, Mode of Addition, and Storage Time

(Experiments run in duplicates)

Code	Distribution Parameters				Penetration		Residual Activity	
	R	S	T	(σ_Q/Q)	$1 - \frac{X_m(E_i)}{X_m(E_1)}$	β to γ Ratio	Activity (E_i)	Activity (E_1)
EXPERIMENTAL RESULTS								
P ₂ B ₁ C ₂ E ₁ a	0.01	0.02	3.0	0.16 ± 0.03	(0.00)	50	(1.00)	
b	0.03	0.12	5.3	0.23 ± 0.04	(0.00)	63	(1.00)	
P ₂ B ₂ C ₁ E ₁ a	0.02	0.06	4.0	0.17 ± 0.04	(0.00)	33	(1.00)	
b	0.10	0.28	3.2	0.23 ± 0.04	(0.00)	50	(1.00)	
P ₃ B ₁ C ₁ E ₁ a	0.05	0.11	3.2	0.25 ± 0.04	(0.00)	48	(1.00)	
b	0.15	0.32	3.1	0.30 ± 0.05	(0.00)	48	(1.00)	
P ₃ B ₂ C ₂ E ₁ a	0.02	0.02	2.0	0.13 ± 0.03	(0.00)	71	(1.00)	
b	0.01	0.04	5.0	0.15 ± 0.04	(0.00)	38	(1.00)	
P ₂ B ₁ C ₂ E ₂ a	0.02	0.03	2.5	0.20 ± 0.03	0.08 ± 0.04	53	0.73	0.66
b	0.02	0.03	2.5	0.19 ± 0.03	0.11 ± 0.04	43	0.58	0.51
P ₂ B ₂ C ₁ E ₂ a	0.04	0.07	2.7	0.13 ± 0.05	0.08 ± 0.06	37	1.06	1.04
b	0.11	0.36	4.3	0.30 ± 0.05	0.18 ± 0.05	48	0.90	0.80
P ₃ B ₁ C ₁ E ₂ a	0.00	0.00	...	0.25 ± 0.04	0.26 ± 0.04	71	0.50	0.40
b	0.07	0.27	4.9	0.21 ± 0.04	0.00 ± 0.06	55	0.74	0.58
P ₃ B ₂ C ₂ E ₂ a	0.02	0.04	3.0	0.17 ± 0.03	0.20 ± 0.05	44	0.74	0.73
b	0.02	0.03	2.5	0.16 ± 0.04	0.17 ± 0.05	37	1.02	0.89
P ₂ B ₁ C ₂ E ₃ a	0.00	0.00	...	0.22 ± 0.04	0.40 ± 0.03	50	0.27	0.39
b	0.01	0.02	3.0	0.20 ± 0.03	0.12 ± 0.04	71	0.29	0.30
P ₂ B ₂ C ₁ E ₃ a	0.05	0.14	3.9	0.14 ± 0.04	0.24 ± 0.04	31	0.93	0.98
b	0.06	0.16	3.6	0.31 ± 0.05	0.25 ± 0.05	24	0.65	0.53
P ₃ B ₁ C ₁ E ₃ a	0.01	0.01	2.0	0.22 ± 0.05	0.52 ± 0.03	36	0.27	0.20
b	0.04	0.23	6.8	0.15 ± 0.03	0.00 ± 0.06	45	0.57	0.44
P ₃ B ₂ C ₂ E ₃ a	0.00	0.00	...	0.07 ± 0.03	0.23 ± 0.03	31	0.59	0.52
b	0.00	0.00	...	0.10 ± 0.05	0.24 ± 0.05	38	0.95	0.81
P ₂ B ₁ C ₂ E ₄ a	0.01	0.01	2.0	0.12 ± 0.04	0.56 ± 0.02	31	0.19	0.27
b	0.01	0.01	2.0	0.19 ± 0.04	0.23 ± 0.04	38	0.10	0.04
P ₂ B ₂ C ₁ E ₄ a	0.04	0.07	2.8	0.15 ± 0.04	0.18 ± 0.05	36	1.07	0.85
b	0.07	0.19	2.6	0.21 ± 0.04	0.38 ± 0.04	26	0.60	0.55
P ₃ B ₁ C ₁ E ₄ a	0.04	0.06	2.5	0.13 ± 0.05	0.64 ± 0.02	36	0.21	0.17
b	0.06	0.21	4.5	0.27 ± 0.06	0.40 ± 0.04	42	0.22	0.12
P ₃ B ₂ C ₂ E ₄ a	0.04	0.05	2.2	0.17 ± 0.04	0.28 ± 0.04	31	0.42	0.44
b	0.00	0.00	...	0.00 ± 0.04	0.18 ± 0.05	38	1.05	0.69
AVERAGED EFFECTS								
P ₂	0.038	0.10	3.6	0.20	0.23	38	0.60	
P ₃	0.033	0.09	3.6	0.19	0.26	42	0.56	
B ₁	0.033	0.09	3.8	0.21	0.28	45	0.37	
B ₂	0.038	0.09	3.5	0.18	0.22	36	0.79	
C ₁	0.057	0.16	3.8	0.22	0.26	38	0.60	
C ₂	0.014	0.03	2.9	0.16	0.23	42	0.55	
E ₁	0.049	0.12	3.5	0.21	(0.00)	48	(1.00)	
E ₂	0.038	0.10	3.6	0.21	0.14	45	0.74	
E ₃	0.021	0.07	4.3	0.19	0.25	37	0.54	
E ₄	0.034	0.08	3.4	0.17	0.36	35	0.44	

single kernels in contact with the free atmosphere was established. Results of the radioactive measurements are given in Table XI. The penetration of mercurial is reported in terms of the beta absorption—the absorbed portion is equal to the expression:

$$1 - \frac{X_m(E)}{X_m(E_1)}$$

where E₁ is E₂, E₃, or E₄. The 2-hour storage time activity here serves as the reference.

The decrease of the beta activity is due mainly to penetration of mercurial in the fruit coat. This view is supported by determinations of the ratio between the beta and the gamma radiation. The beta to gamma ratio was evaluated in this way: A number of kernels were placed on a disk and the activity was determined in the usual way. Thereafter, the disk was covered with a 0.5-mm. thick aluminum plate, which does

not transmit the beta radiation but a large proportion of the gamma. The Geiger tube used detects only 1% of the gamma radiation, and therefore the gamma measurements were not too accurate. Nevertheless, the trends of the averages of the beta to gamma ratio in Table XI support the view that the absorption values are connected with the penetration of mercurial and not caused by losses.

The column for residual activity in Table XI refers to measurements of the loss of mercurial from single seeds in contact with the free atmosphere. In this test, a number of kernels were cemented on a disk 2 hours after the seed treatment. The disk with the attached layer of kernels was exposed to the free atmosphere in a fume hood with good air circulation. Activity measurements were made at different times with the 2-hour values as references. No conclusions should be drawn regarding losses of mercurial from seeds under actual

conditions, where only a very small percentage of the kernels is exposed to the surrounding atmosphere. This experiment, however, shows vapor action as all evaporated mercurial molecules are lost and show up as a deficit.

Inspection of the average effects quoted in Table XI reveals the following features:

The distribution for the concentrated product, P₃, is essentially the same as the distribution obtained with normal Panogen used in a normal dosage, P₂.

The spreading errors are all about the same. Vapor action evidently compensates for the poor liquid distribution obtained in case of high moisture content as demonstrated previously.

The mode of addition still seems to have a significant effect on the number of R-kernels. In case of unfavorable addition where the disinfectant is dripped directly on the seed kernels and not via a spreading device, the number of R-kernels increases so that a larger portion

Table XII. Penetration and Evaporation of Mercurial for Wheat Seeds as Functions of Moisture Content and Storage Time

Code	Rel. Activity, Closed Storage $X_m(E_1)$	Rel. Activity, Open Storage Activity (E_1)	β to γ Ratio	Average Activity, Closed Storage, X_m , C.P.M.	Thickness of Absorbing Surface Layer, Mg./Sq. Cm.
B ₁ E ₁	(1.00)	(1.00)	50.0	229	5.6
B ₂ E ₁	(1.00)	(1.00)	43.5	166	9.6
B ₁ E ₂	0.89	0.59	52.6	202	7.0
B ₂ E ₂	0.84	0.90	40.0	140	11.0
B ₁ E ₃	0.74	0.34	47.6	162	9.9
B ₂ E ₃	0.76	0.75	29.4	126	12.9
B ₁ E ₄	0.54	0.17	35.8	125	13.0
B ₂ E ₄	0.74	0.71	32.3	124	13.1

of the disinfectant is used uneconomically as reflected in the increase of the S-factor from 0.03 to 0.16. The T-factor, which is a measure of the relative dosage of the R-kernels, however, remains essentially constant. In laboratory treatment, according to C₁, about 10 seconds elapse before the mixing is started.

A storage time of 2 hours is sufficient to give a good distribution. There is a further small improvement at increased storage time but the gain is of no practical importance.

Comparing the distribution reported in Table XI with the values obtained for the nonvolatile A dressing, in case of good mixing conditions (Tables VII and IX), the distributions of these particular non-volatile and volatile compounds turn out to be much the same. Under more severe conditions, however, the nonvolatile compound is very unevenly distributed, whereas treatment with the volatile mercurial results in a good N-kernel distribution.

The data for absorption, residual activity, and beta to gamma ratio are further clarified in Table XII, in which the combinations of storage time and moisture content are isolated. The evaporation of mercurial from seeds of a high moisture content is practically zero, as the relative activities for closed and open storage are about the same. At the 12% level, some evaporation of mercurial is taking place even after a long storage time.

Seeds are not stored in practice as was done in these experiments where single kernels were exposed to a continuous air stream. The resorption of mercurial takes place at a higher rate when the moisture content is high. Within 2 hours, the mercurial seems to be fully tied up in the kernel. However, the vapor action has done its job within that time (Table XI).

Figure 12 shows that the reduction of beta activity on storage is due to diffusion of mercurial. The average activities for closed storage (Table XII) are here plotted as a function of the beta to gamma ratio. As the gamma activity should remain practically constant, a linear relationship between the average

activity and the beta to gamma ratio is to be expected. This ratio, in case of no penetration, was determined to be 91 and the maximum average activity in case of no diffusion is then estimated to be 340 c.p.m.—beta and gamma determination on thin layer of straight disinfectant. The maximum diffusion observed in these experiments corresponds to the activity 120 c.p.m. or $120/340 \times 100\% = 35\%$ transmission. Hence, the mercurial is situated behind a layer with the mean thickness of 13 mg. per sq. cm., or roughly 100 microns, considering the fact that the measurements are not performed on a flat surface. The thickness of the fruit coat of wheat kernels is actually about 100 microns; therefore, the whole fruit coat is penetrated by the fungicide.

The depths of diffusion have been calculated in a similar way for the combinations in Table XII. These values are plotted in Figure 13 as a function of storage time. The rapid diffusion of mercurial, particularly at 18% moisture content, is demonstrated very clearly in this diagram.

The duplicates in Table XI do not agree too well as regards penetration and residual activity. In all series, however, the penetration increases with time, whereas the residual activity is decreased

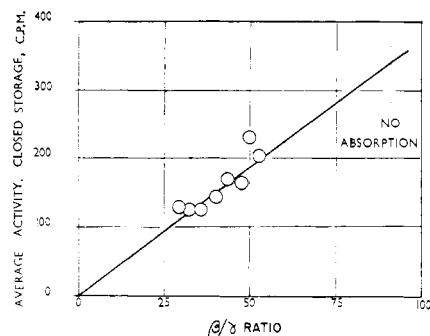


Figure 12. Relationship between average activity and β to γ ratio

or remains constant. The differences between the duplicates are not believed to be due to experimental errors but considered to reflect differences between the seed samples. One explanation could be that the seeds are in different states of biological activity. This completely randomized experimental design was run over a period of time during which changes may have occurred in the activity of the kernels. In Figure 14, values for relative activity in closed storage are given as a function of the date for treatment. All curves exhibit minimum activity around September 10. The seeds treated around this date therefore appear to be in more reactive state (faster diffusion) than seeds treated before or after this date.

Germination Tests on Radioactive Seeds. Germination tests were run on all of the 3200 seeds investigated in the large distribution experiments reported above. In this case the activity of each separate kernel was known. Results are reported in Table XIII. Values for nontreated seeds at normal moisture content are included. When these badly infected seeds were stored at the high moisture-content level, there were no healthy seedlings at all because of attack of storage fungi under these severe storage conditions.

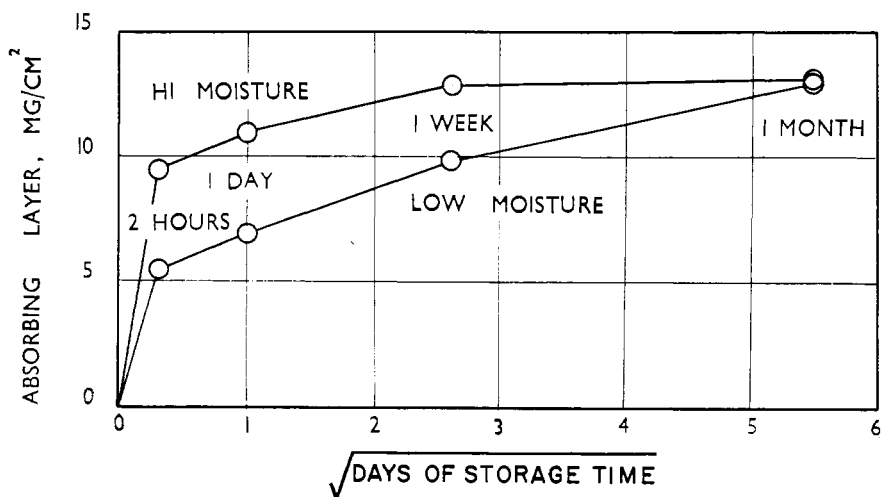


Figure 13. Diffusion of mercurial into kernels of wheat as a function of storage time and moisture content

Table XIII. Germination of Highly Infested Wheat as a Function of Liquid Dosage, Moisture Content of the Seed, Mode of Addition, and Storage Time

(Experiments run in duplicates)

Code	Healthy, No.	Mold, No.	Fusarium, No.	Cripples, No.	Overtreated, No.	Dead, No.	Straw, Cm.	Root, Cm.
EXPERIMENTAL RESULTS								
P ₂ B ₁ C ₂ E ₁ a	81	0	7	1	0	11	12.8	9.1
b	85	0	8	3	0	4	15.0	9.0
P ₂ B ₂ C ₁ E ₁ a	75	0	1	5	4	15	10.0	5.3
b	81	0	2	2	3	12	13.6	7.7
P ₉ B ₁ C ₁ E ₁ a	88	0	7	0	1	4	13.3	10.3
b	84	0	9	3	1	3	16.7	9.8
P ₉ B ₂ C ₂ E ₁ a	78	12	5	1	0	11	16.0	8.0
b	83	5	4	2	0	10	12.8	8.0
P ₂ B ₁ C ₂ E ₂ a	92	0	2	1	0	5	14.6	9.0
b	89	0	2	2	0	7	13.3	9.7
P ₂ B ₂ C ₁ E ₂ a	75	0	3	11	3	8	8.9	5.4
b	77	1	3	1	6	12	11.4	6.8
P ₉ B ₁ C ₁ E ₂ a	89	0	4	1	0	6	14.1	10.8
b	90	0	1	1	0	8	16.0	9.7
P ₉ B ₂ C ₂ E ₂ a	79	8	1	3	0	14	15.0	6.0
b	80	5	2	1	1	12	11.0	7.5
P ₂ B ₁ C ₂ E ₃ a	89	0	1	2	1	8	12.6	9.3
b	92	0	1	0	3	4	14.5	8.8
P ₂ B ₂ C ₁ E ₃ a	64	1	5	8	2	21	14.4	6.7
b	78	0	1	0	4	13	11.5	5.9
P ₉ B ₁ C ₁ E ₃ a	85	0	3	3	0	9	12.8	9.0
b	92	0	1	1	0	6	14.2	8.9
P ₉ B ₂ C ₂ E ₃ a	78	6	2	2	0	20	15.6	7.8
b	75	11	3	2	0	17	10.3	6.7
P ₂ B ₁ C ₂ E ₄ a	79	0	0	4	4	13	11.0	7.6
b	83	0	5	1	2	9	13.5	8.1
P ₂ B ₂ C ₁ E ₄ a	16	69	8	3	4	48	12.9	5.7
b	70	0	1	3	7	19	10.7	7.0
P ₉ B ₁ C ₁ E ₄ a	88	0	0	2	0	10	11.4	8.8
b	92	0	1	1	1	4	12.4	8.9
P ₉ B ₂ C ₂ E ₄ a	71	15	0	2	0	24	12.4	7.7
b	46	32	3	5	0	37	12.2	7.4
AVERAGED EFFECTS								
P ₂	77	5	3.1	2.9	2.7	13	12.5	7.6
P ₉	81	6	2.9	1.9	0.3	12	13.5	8.5
B ₁	87	0	3.3	1.6	0.8	7	13.7	9.2
B ₂	70	10	2.8	3.2	2.1	18	12.4	6.9
C ₁	78	5	3.1	2.8	2.2	12	12.8	7.9
C ₂	80	6	2.9	2.0	0.7	13	13.3	8.1
E ₁	82	1	5.4	2.1	1.1	9	13.8	8.4
E ₂	84	1	2.3	2.6	1.3	9	13.0	8.1
E ₃	82	1	2.1	2.3	1.3	12	13.2	7.9
E ₄	68	7	2.3	2.6	2.3	21	12.1	7.7
Nontreated (B ₁)	8	0	63	24	...	27

The concentrate used in the ultra-low dosage, works as well as commercial Panogen in its normal dosage as shown by the distribution measurements reported in Table XI.

The number of overtreated kernels is reduced in case of the concentrate to the same extent as the dosage is reduced, mainly by a factor of 10. The number of overtreated kernels also shows the greatest response to high moisture content and improper addition of liquid C₁. The outcome of the seed treatment, as regards the frequency of overtreated kernels, is governed by the initial liquid distribution and the amount of liquid used. These damages may be generated when particularly sensitive parts of the kernel, most probably the embryo, come in contact with the disinfectant.

The effects of moisture content and storage time are further investigated in Table XIV. Here storage times E₁ and E₂ are pooled as well as E₃ and E₄. The kernels are classified into groups accord-

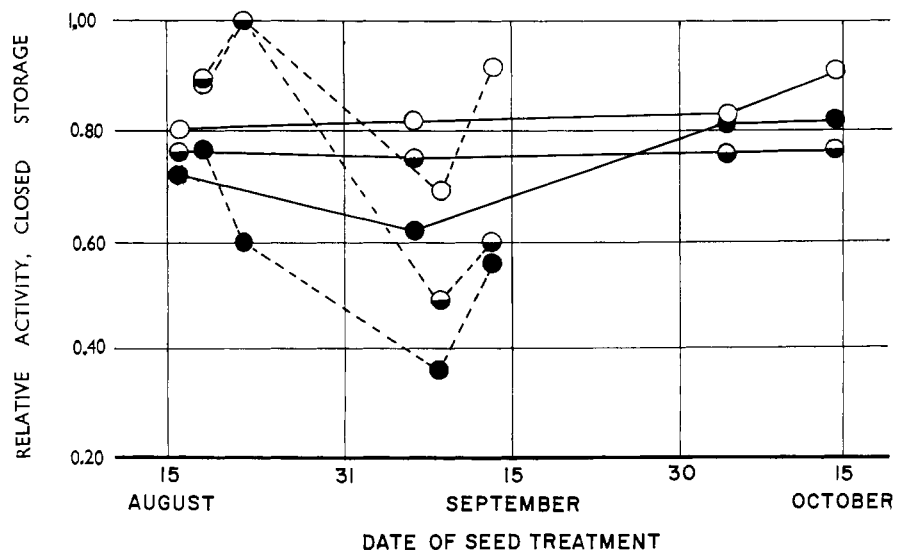


Figure 14. Graph illustrating possible variation in biological activity of seeds. Figures for the relative activities on the y-axis were obtained by division of the actual activity with the reference activity observed 2 hours after treatment

--- B₁ — B₂ ● E₂ ● E₃ ○ E₄

Table XIV. Germination of Highly Infested Wheat as a Function of Mercurial Content of Individual Kernels, Storage Time, and Moisture Content

Code	Activity	Fraction	Healthy	Mold	Fusarium	Cripples	Overtreated	Dead
B ₁ E _{1,2}	<X _m	494 (0.618)	441 (89%)		23 (5%)	7 (1%)	1 (0.2%)	22 (4%)
	X _m -2X _m	280 (0.350)	234 (84%)		16 (6%)	5 (2%)		22 (8%)
	2X _m -4X _m	18 (0.023)	14 (78%)		1 (6%)			3 (17%)
	4X _m -8X _m	5 (0.006)	3 (3/5)					1 (1/5)
	>8X _m	3 (0.004)	3 (3/3)					
B ₁ E _{3,4}	<X _m	495 (0.619)	456 (92%)		4 (1%)	7 (1%)	5 (1%)	23 (5%)
	X _m -2X _m	289 (0.361)	235 (81%)		8 (3%)	8 (3%)	6 (2%)	34 (12%)
	2X _m -4X _m	13 (0.016)	7 (54%)			2 (15%)		4 (31%)
	4X _m -8X _m	1 (0.001)	1 (1/1)					
	>8X _m	2 (0.002)	0 (0/2)					2 (2/2)
B ₂ E _{1,2}	<X _m	476 (0.595)	394 (83%)	7 (1%)	15 (3%)	16 (3%)	4 (1%)	41 (9%)
	X _m -X _m	297 (0.371)	222 (75%)	23 (8%)	6 (2%)	10 (3%)	10 (3%)	39 (13%)
	2X _m -4X _m	20 (0.025)	10 (50%)	1 (5%)			2 (10%)	7 (35%)
	4X _m -8X _m	6 (0.008)	2 (2/6)					4 (4/6)
	>8X _m	1 (0.001)	0 (0/1)				1 (1/1)	
B ₂ E _{3,4}	<X _m	466 (0.583)	320 (69%)	66 (14%)	11 (2%)	8 (2%)	7 (2%)	98 (21%)
	X _m -2X _m	314 (0.393)	173 (55%)	65 (21%)	12 (4%)	17 (4%)	10 (3%)	92 (29%)
	2X _m -4X _m	18 (0.023)	5 (28%)	3 (17%)		1 (6%)	4 (22%)	8 (44%)
	4X _m -8X _m	1 (0.001)	0 (0/1)				1 (1/1)	
	>8X _m	1 (0.001)	0 (0/1)					1 (1/1)
B _{1,2} E ₁₋₄	<X _m	1931 (0.604)	1611 (83%)	73 (4%)	53 (3%)	38 (2%)	17 (1%)	184 (10%)
	X _m -2X _m	1180 (0.369)	864 (73%)	88 (7%)	42 (4%)	40 (3%)	26 (2%)	187 (16%)
	2X _m -4X _m	69 (0.022)	36 (52%)	4 (6%)	1 (1%)	3 (4%)	6 (9%)	22 (32%)
	4X _m -8X _m	13 (0.004)	6 (46%)				1 (8%)	5 (38%)
	>8X _m	7 (0.002)	3 (3/7)				1 (1/7)	3 (3/7)
B _{1,2} E ₁₋₄		3200 (1.000)	2520 (79%)	165 (5%)	96 (3%)	81 (3%)	51 (2%)	401 (13%)

ing to their radioactivity. The percentage figures given in parenthesis after the number of kernels refer to the percentage of kernels with this special diagnosis within the actual group.

The percentage of dead kernels is seen to be roughly proportional to the mercury content. A large percentage of dead kernels at the high moisture-content level are caused by mold development and should be subtracted to give a more realistic figure. However, even the very few kernels with a high activity have a good chance of surviving.

A very short storage time and low moisture content will give a slightly higher percentage of *Fusarium*-infected kernels. The entire fruit coat is not completely penetrated by the mercurial under these conditions and deeply situated fungi may then have a small chance of development before they are reached by the mercurial. At the high moisture content, on the other hand, resorption is very rapid and control is obtained after 2 hours' storage.

Kernels with a higher mercurial content do not exhibit less *Fusarium* residues (Table XIV). Actually the emergence picture is slightly inferior. This may be explained in three different ways: A slight increase in mercurial content will offer less protection; a high mercurial content may, to some extent, damage the kernels so that the effect of fungicidal protection will be, to a slight extent, counteracted; and kernels that already are damaged or have acquired larger infestations of fungi are also likely to take up larger amounts of mercurial.

The first possibility is not reasonable in case of *Fusarium* fungi. The other two

Table XV. Mercurial Uptake of Damaged and Intact Kernels

	Storage Time	Distribution Parameters				Average Activity	
		R	S	T	(σ _q /Q)	X _m	X' _m
Damaged kernels, mean weight 32.9 mg.	4 hours	0.03	0.07	3.3	0.26 ± 0.05	287	258
	4 days	0.03	0.20	7.6	0.39 ± 0.06	348	269
Intact kernels, mean weight 40.2 mg.	4 hours	0.01	0.02	3.0	0.25 ± 0.04	207	201
	4 days	0.01	0.01	2.0	0.38 ± 0.06	226	222

possibilities are both reasonable but there is much evidence in favor of the third.

Thirty grams of damaged kernels mixed with 70 grams of intact kernels were treated with P₂ and counted at random in an ordinary distribution measurement. The results are shown in Table XV. The R-kernel population is less favorable in the case of the damaged kernels than in the case of intact kernels.

The damaged kernels carry more mercurial than the intact kernels—25% more when X_m is used for the calculation or 47% when X'_m is used. These figures refer to the amount of mercurial per kernel. If the difference in average kernel weight is taken into consideration, the damaged kernels carry 51% (78%) more mercurial per unit weight than the intact kernels. The spreading error increases with storage time, whereas the opposite is true for a normal sample (Table XV). During the storage, because of the vapor action, there will be a poorer distribution of the mercurial, in this case. The damaged kernels are intimately mixed with the intact kernels and these two populations have different

properties as regards mercurial resorption. If mercurial is transported from an intact kernel to a damaged kernel, this will show up not only as a high concentration of mercury on the damaged kernel, but also as a correspondingly lower concentration of the involved intact kernel. Therefore the spreading errors of the two populations will be affected in the same way.

Autoradiographs. Autoradiographs were taken of the radioactive seeds after storage for 1 week. The time of exposure, H, varied between 7 and 29 days, depending on the activity of the sample. During this comparatively long storage time, vapor action was taking place and, in some cases, was giving diffuse photographs. When vapor action was absent—i.e., with seeds of a high moisture content—clear photographs were obtained.

The pictures (Figure 15) compare well with the numerical results in Table XI. When kernels are spread on a surface they usually fall furrow down. Most of the kernels on these pictures therefore have the furrow facing the film.

The resorption of mercurial should

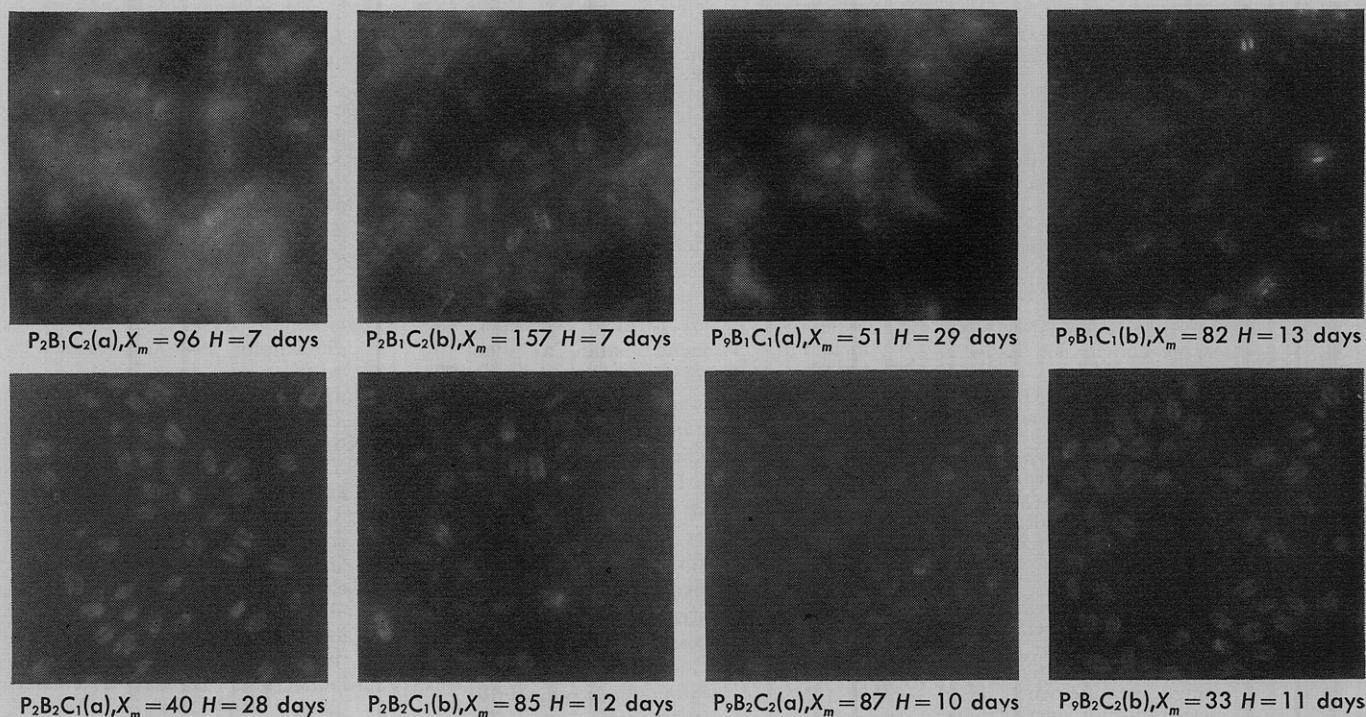


Figure 15. Autoradiographs of wheat seeds reported in Table XI, where code is explained

take place on the same transport ways that are used in the transport of water during the germination; however, autoradiographs show no mercurial taken up by the furrow. This may indicate that the furrow has no such pore system for transporting water into the kernel.

The effect of vapor action is very clearly illustrated in the samples with a low moisture content. The diffuse zones around each kernel are due to evaporated mercurial molecules. The seed kernels in these experiments are covered by sand and the evaporated molecules have to travel through this heavy layer. The most important feature demonstrated by these pictures is the very even distribution of the mercurial with the exception of the furrow for reasons discussed above.

Figure 16 shows autoradiographs for the intact and damaged seeds reported in Table XV. The high mercurial content of the damaged kernels and the poor distribution in this case are easily observed.

Nomenclature

- P_1 = Panogen, liquid, 0.8% mercury, cyano(methylmercuri)-guanidine, Swedish formulation used several years ago
- P_2 = Panogen, liquid, same mercurial and concentration as P_1 . Present Swedish formulation (slightly different solvents compared to P_1)
- P_3 = Betoxin F, liquid, 0.8% mercury, solubilized ethyl mercuric chloride
- P_4 = Ceresan M, dust, 3.2% mercury, *N*-(ethylmercuri)-*p*-toluenesulfonanilide
- P_5 = Certosan T, dust, 1.5% mercury, alkoximethyl mercuric compound (not further specified)
- P_6 = Lunasan, dust, 0.54% mer-

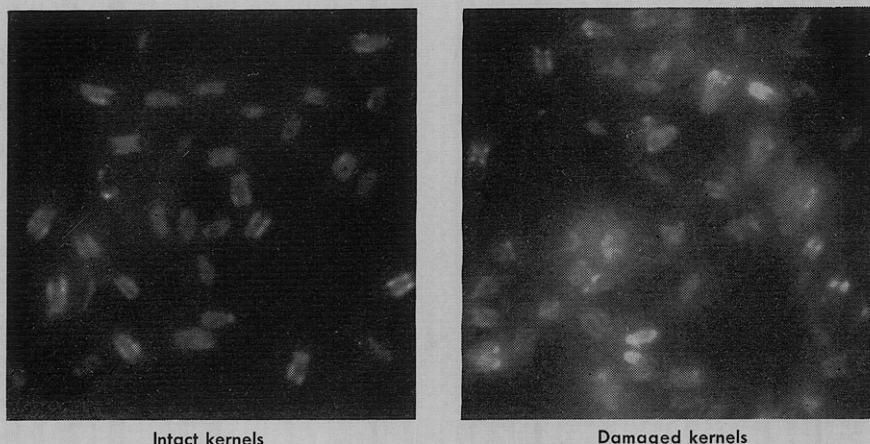


Figure 16. Autoradiographs of damaged and intact seeds treated simultaneously according to $P_2B_1C_2$ (Table XV)

- P_7 = Agrosan, dust, 1.0% mercury, 0.15% as ethyl mercuric chloride and 0.85% as phenyl mercuric acetate
- P_8 = Betoxin TD, dust, 0.8% mercury, ethyl mercuric chloride magnesium bromide, plus 40% Thiram
- P_9 = experimental-type Panogen, 8.0% mercury as methyl mercuric hydroxide
- B_1 = 12% seed moisture content (normal)
- B_2 = 18% seed moisture content (high)
- G = surface area in square centimeters
- dn = number of molecules evaporating from G
- dt = time of dn
- p_a = actual vapor pressure
- p_s = saturation vapor pressure
- e = condensation coefficient factor
- k = Boltzmann's constant
- m = mass of one molecule
- T = absolute temperature
- c_s = mercurial vapor concentration, saturation value
- I = time required to decrease mercury content of kernel to half its original value in evaporation process
- d = diffusion rate
- D = diffusion constant
- J = time required to decrease the mercurial content of kernel to half of its original value in diffusion process
- L = time required to reduce mercurial content to normal value from extreme overdose
- c_a = mercurial vapor concentration, actual value
- E_1 = 2 hours storage time after treatment
- E_2 = 24 hours storage time after treatment
- E_3 = 1 week storage time after treatment
- E_4 = 1 month storage time after treatment
- G_1 = air stream method for removal of mercurial
- G_2 = rinsing with water to remove mercurial
- G_3 = air stream and rinsing to remove mercurial

σ = standard deviation (sigma)
 A = 5.2% thallium nitrate tagged with thallium-204 (aqueous solution)
 C₁ = liquid poured directly on seeds, with mixing after about 10 seconds
 C₂ = liquid poured on walls of bottle and transferred to seeds via wall
 D₁ = normal rolling-type motion of mechanical mixing (Figure 6)
 D₂ = vigorously shaken by hand in flask for 4 minutes
 X'_m = average activity for N-kernels
 X_m = average activity for R- + N-kernels
 2 × X_m = borderline activity between kernels with high and normal activity
 R-kernels = higher activity kernels
 N-kernels = normal activity kernels
 R-factor = number of R-kernels divided by total number of kernels
 S-factor = excess mercurial carried by R-kernels divided by total mercurial
 T-factor = (S/R + 1) S-factor divided by R-factor plus 1, relative dose on R-kernels
 X_i = kernel count (activity in a single measurement)
 Q = micrograms of mercury per square centimeter, average concentration of mercurial on G
 q = over-all counting efficiency
 c = millicuries per microgram of mercury, the specific activity of mercurial
 K = conversion factor, counts per minute per millicurie
 t = counting time, minutes
 (σ_X/X) = coefficient of variation for kernel count
 (σ_K/K) = error due to statistical fluctuations
 b = denotes background activity (c.p.m.)
 (σ_q/Q) = spreading error
 N = number of analyzed kernels
 (σ_G/G) = variation in kernel surface
 (σ_q/q) = geometrical error
 F₁ and F₂ = operators

A₁ = normal dosage of disinfectant A
 A₂ = 0.1 of normal dosage of disinfectant A
 H = necessary time of exposure of treated kernels on film
 E_i = E₂, E₃, or E₄

Acknowledgment

This long-term study has been supported by the manufacturers of the Panogen liquid seed disinfectants. The author wishes to express his most sincere thanks to J. M. Blegvad, AB Casco, Stockholm, Sweden; and to Pete Hellman, of Panogen Inc., Ringwood, Ill., for continuous interest and encouragement and for the permission to publish these investigations. Thanks are also extended to D. C. Arny, R. L. Frank, Åke Hedén, Margareta Lambert, Sigurd Lindberger, Henrik Markkula, and R. P. Seven for valuable discussion. The skillful technical assistance of Sven Bodin, John Carlsjö, Erik Ljusberg, and Tommy Åkerlund is gratefully acknowledged. Particular thanks to Kajsa Weinar, for the tedious radioactive measurements and the biological tests.

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Received for review February 7, 1957. Accepted January 24, 1958.

FERTILIZER MATERIALS

Phosphoric Acid of High Concentration

THE PHOSPHORIC ACID sold as phosphatic fertilizer solution contains about 55% phosphorus pentoxide or 75% orthophosphoric acid. The acid, produced either by the wet process or from phosphorus obtained by the electric-furnace method (11), is used for the production of solid and liquid fertilizers. As a part of its objective to develop methods for producing better and lower cost fertilizers, the Tennessee Valley Authority (TVA) has studied the production and properties of furnace acid of higher concentrations than that available to the fertilizer industry.

In 1937 Durgin, Lum, and Malowan (3) published information on the characteristics of strong phosphoric acids and stated that an acid containing 84% phosphorus pentoxide, called tetraphosphoric acid, was being produced commercially. In 1941 Walthall and Stripplin (12) described the pilot-plant development of a method, involving the combustion of phosphorus with dried air, for the production of phosphoric acid of about this concentration.

In recent work by TVA (9), acid containing 76% phosphorus pentoxide, which is equivalent to 105% orthophos-

phoric acid, has advantages over acids both of lower and higher concentrations. Acid containing about 60 to 75 and about 77 to 83% phosphorus pentoxide crystallized at ordinary temperatures, whereas that containing about 75 to 77% phosphorus pentoxide remained liquid at temperatures low enough to be shipped and stored in tanks exposed to the weather. Acid containing about 84% phosphorus pentoxide also remained liquid, but it was much more viscous than the acid containing about 76% phosphorus pentoxide, which has been called "superphosphoric acid," and has been

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