THE INFLUENCE OF ALCOHOL UPON THE GROWTH OF SEEDLINGS.

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I.

The Problem.

The object of the experiments here reported, which have been going on during the past winter, was to determine the influence of small doses of ethyl alcohol applied to the dry seed upon the subsequent growth of the seedling, kept under such conditions as to get no extraneous food whatever, but instead to be dependent solely upon the utilization of the stored food in the cotyledons. The results of such experiments were thought likely to have bearing upon certain general problems, regarding which work is going on in this laboratory. These are: (a) The influence of ethyl alcohol upon duration of life. It has been shown by Pearl (1) and by Stockard and Papanicolaou (2) that in fowls and guinea pigs, individuals regularly and moderately alcoholized by the inhalation method considerably outlived their controls. Pearl (3-6) has also shown that moderate drinking leads to no impairment of the expectation of human life. The present series of experiments are preliminary to an attack upon this same general problem with seedlings, a material thought likely to afford some critical approaches not possible with any material hitherto used for its study. (b) The selective action of ethyl alcohol upon somatic cells, similar to its action in this sense upon germ cells, first demonstrated by Pearl (7-10), confirmed by Danforth (11) and more recently by Stockard (12-14). (c) The form of the life curve produced under complete inanition (in this case relative to extraneous food only). We (15-16) have recently studied starvation life curves in Drosophila, and the biologically different conditions of starvation

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in seedlings introduces another and interesting element into the problem.

The present experiments are, in a sense, preliminary. A great deal of time has been spent in working out a critically accurate technique. The work will be continued, but as definite results have been obtained on one phase of the experimental program it is thought desirable to report them at this time.

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Methods.

All of the experiments here reported have been carried out with the seeds of the cantaloupe (*Cucumis melo*, var.). This form of cucurbitaceous seed, after various trials, proved to be excellently suited to the type of experiment we had in mind.

The general plan of all the experiments will be first described, and then points of detail will be discussed. From a large mass of seeds, samples, usually of 60 seeds each, were counted out at random; *i.e.*, taking the individual seeds as they came to hand without selection. Each batch of 60 seeds was then soaked for 3 hours in 50 cc. of one or another of the following series of solutions: 0 alcohol (distilled H_2O), 2, 4, 8, 12, 16, and 32 per cent alcohol.

The solutions were made up fresh each time, with the greatest care to secure accuracy, from absolute ethyl alcohol (Squibbs) and distilled water. At the end of the 3 hour period of soaking the seeds were removed from the solution and thoroughly washed in distilled water (or dried with filter paper) in order to remove any alcohol adherent to the surface. Then they were put in germinating pans (Pyrex glass pie plates covered on the bottom with filter paper soaked with distilled water). These plates were covered, to make each a moist chamber, and then put into an electric incubator, running at 37.5°C., to germinate. After 36 to 48 hours, when germination was complete the seedlings were carefully put upon a sterilized cheese-cloth platform, so that the sprout stuck through below the cheese-cloth and the cotyledons were above it. This platform was then suspended in a glass museum jar, containing just enough distilled water to reach up to the cheese-cloth platform, but not to go above it.

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Thus as the seedlings grew the roots were bathed in distilled water, while the stems were in the air of the moist chamber created by covering the jars. These jars were placed in an electric incubator at 37.5°C., with the heating bulbs covered with red paper: this prevented the seedling from forming chlorophyll and nourishing itself by photosynthesis. Each day the position of each jar in the incubator was changed, both horizontally and vertically, in order to equalize the effects of any differences of temperature in different parts of the incubator. Under these conditions growth ceased in about 8 to 10 days. After the seedling had finished their growth there soon supervened a breakdown of the tissues of the stem, which seemed to occur regardless of whether aseptic conditions had been maintained or not, but this latter point regarding the possible existence of a sterile autolysis has not been sufficiently investigated to make a positive statement possible. On the 9th or 10th day the seedlings were removed from the jars and divided with a sharp scalpel into three portions, cotyledons, stem, and root. Each portion was then pressed between filter papers to remove adherent water, and the fresh weight taken at once on an analytical balance to the nearest mg.

Summarizing the whole procedure, what we have in these experiments is a record of the growth of cantaloupe seedlings, as measured by increase in fresh weight, in the entire absence of extraneous food intake, following an initial soaking of the dry seeds in various concentrations of ethyl alcohol for a period of 3 hours.

The experiments have been varied in minor particulars from the above outline, in the course of developing the technique, but without essential effect on the results. Thus the depth of distilled water in the growing jars was not found to influence the results. Nor does washing or superficially drying the seeds after their initial soaking discernibly affect the subsequent growth rates, nor does the number of seeds placed in one jar, between the limits of 5 and 15. The dimensions of the museum jars used for the growth are approximately $9 \times 14 \times 21$ cm.

One feature in the technique in the experiments here reported needs emphasis, because we believe it to be of importance in the interpretation of the results, for reasons which will presently appear. In the experiments to date the selection of dry seeds, and what is more important of the sprouted seeds from the germinating pans to go into the growing jars *has been at random*. That is to say, instead of following the usual procedure in plant physiological work of picking for the growing jars only the healthiest and best developed sprouted seeds, we have either put in all sprouted seeds, or else chosen them at random regardless of the length or healthiness of the seedling, when we had too many for the available jars.

In discussing the results it will be desirable to consider separately the two most important features, germination and growth. Since all the separate series of experiments gave similar results it will not be necessary to take the space to discuss each series separately, but instead we shall deal with the averaged data.

TABLE	I.
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<u> </u>		• n	1.
Gern	nınatı	on K	esults.

_			Alcohol	solution.		
Item.	H ₂ O only.	2 per cent.	4 per cent.	8 per cent.	12 per cent.	16 per cent.
No. of seeds	540	480	540	480	420	540
No. which germinated	454	362	411	363	280	348
Percentage germination		75.4	76.1	75.6	66.7	64.4
Probable error of percentage	± 1.1	±1.3	±1.2	±1.3	±1.6	±1.4

Germination.

The results of the germination in the different series of experiments are shown in Table I. These figures represent total germination. By this is meant that the pans were continued in the incubator until all seeds had germinated that ever would. In addition to the alcohol solutions noted in Table I a 32 per cent and a 64 per cent solution were tried, each in a single series. After the 32 per cent alcohol treatment only 1 seed in 60 germinated, and after the 3 hour soaking in 64 per cent alcohol no seed germinated. Following these trials 16 per cent was the strongest alcohol solution used. Inasmuch as germination experiments were tried from which seedlings were not subsequently grown, the totals of Table I are larger than those of Table III. The probable errors of the percentages, given in the last line of Table I indicate the order of variation to be expected in the results from simple random sampling alone. They are computed from the usual expression of the probable error of a percentage

$$\pm 67.449 \sqrt{\frac{pq}{n}}$$

where p = percentage germination; q = 100 - p; n = number of seeds.

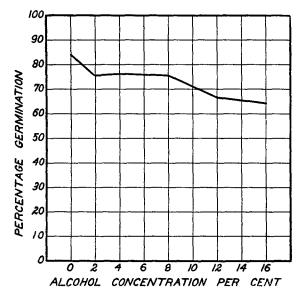


FIG. 1. Showing the deleterious effect upon germination of soaking the seeds of *Cucumis melo* in ethyl alcohol solutions.

The percentages of Table I are shown graphically in Fig. 1.

It is apparent that all the solutions of alcohol, even the weakest, affect adversely the germination of the seeds. The germination of those soaked in the 2 per cent solution is nearly 10 per cent lower than that of those soaked in water. After the initial drop of the curve, from 0 and 2 per cent alcohol, it continues at about the same level to and including 8 per cent concentration. This would suggest that the weak solutions of alcohol, those below 8 per cent, are not markedly different among themselves in their inhibiting effect upon

germination. Between 8 per cent and 12 per cent concentration there is again a marked drop of nearly 10 per cent in the germination. There is a still further drop in passing to 16 per cent.

It thus appears that of the original number of dry seeds taken at random from a large aggregation, a certain number are eliminated from any possibility of taking a part in the determination of the record of average growth, because they failed to germinate. Those so eliminated after soaking in water only may be fairly presumed to be essentially non-viable seeds, because of defective prior development or innate constitution. But in the case of seeds soaked in alcohol solutions a larger number fail to germinate than after treatment with water only. Regarding these additional seeds eliminated it may fairly

TABLE	II.
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	1	Alcoh	ol concen	tration.	
Item.	2 per cent.	4 per cent.	8 per cent.	12 per cent.	16 per cent.
No. of presumably viable seeds, which would have					
germinated if originally soaked in H2O only	404	454	404	353	454
No. which actually germinated	362	411	363	280	348
No. of presumably viable seeds which failed to ger-		1	[[1
minate following alcohol treatment	42	43	41	73	106
Percentage which seeds eliminated by alcohol is of		1			
total number of presumably viable seeds	10.4	9.5	10.1	20.7	23.3

Elimination of Seeds by Alcohol.

be presumed that they were not *essentially* non-viable. They would have germinated if they had been soaked in water only. The numerical relations of this extra elimination of seeds by alcohol from the possibility of participating in the growth records are shown in Table II.

In Table II the figures in the first line are derived by applying the germination percentage of the seeds soaked in water only (84.1 from Table I) to the total number of seeds put in each solution. The derivation of the remainder of the table is sufficiently self-evident.

From Table II it appears that the preliminary treatment with alcohol in concentrations up to and including 8 per cent eliminates, by the prevention of germination, about 10 per cent of the presumably viable seeds. At a concentration of the alcohol of 12 per cent nearly

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21 per cent of the presumably viable seeds are eliminated, and 23.3 per cent at a concentration of 16 per cent alcohol.

Experiments are now under way designed to extend and smooth the curve of germination under the influence of alcohol. The present material is not sufficiently extensive to enable a determination of the true mathematical relation between germination and concentration of alcohol.

It has long been known that alcohol exerts a deleterious influence upon the germination of seeds. We shall make no attempt here to review the literature on this point, merely calling attention to the fact that as long ago as 1876 Nobbe (*Samenkunde*) found that wheat soaked 3 days in 10 per cent ethyl alcohol germinated 54 per cent.

Growth.

The results as to the growth of the seedlings, under the conditions of complete starvation, so far as concerns extraneous food, are shown in Table III. The figures given are mean fresh weight per individual seedling, in mg. These figures represent completed growth under the conditions of the experiments.

The means of Table III are shown graphically in Fig. 2.

From the data of Table III and Fig. 2 the outstanding result is that all the series treated in alcohol solutions made a greater growth than was attained by those treated in water only. The conditions of the experiments should again be emphasized. The alcoholized seeds were merely soaked for 3 hours in the indicated alcohol solutions before germination. They then germinated and grew in distilled water and in darkness. There can be no question of the alcohol acting as a food in the metabolism of the seedlings in this case because they did not grow in the alcohol solutions. They merely received a preliminary soaking in them. The amount of alcohol which actually got into each individual seed must have been extremely small. But plainly this preliminary treatment with alcohol greatly increased the total growth of the seedlings, using only the stored food in the cotyledons.

Examining the results in detail it is seen that:

1. The mean fresh weight of the cotyledons at the end of the growth under starvation conditions is about the same in all the series. It is

GROWTH OF SEEDLINGS

Growth of Se	Growth of Seedlings of Cucumis melo. Mean Fresh Weight per Seedling. Dry Seeds Soaked 3 Hours in Alcohol Solutions.	Seedling.	Dry See	ls Soaked	3 Hour	s in s	Alcohol Sc	olutio	ns.
Ĩ		H ₂ O	2 per cent alcohol.	4 per cent alcohol.		<pre>8 per cent alcohol.</pre>	12 per cent alcohol.	¥	i per cent alcohol.
Lart	Ttem.	No. Total weight in mg.	No. weight in mg.	t No. weight in mg.	No.	Total weight in mg.	No. Weight in mg.	No.	Total weight in mg.
	Absolute totals	174 6, 38	8 135 5, 12	174 6,383 135 5,127 163 5,826	124	4,642	124 4, 642 73 2, 720 162	0162	6,022
Cotyledons.	Mean weight (in mg.) per seedling Percentage increase over H ₂ O	36.6 0	38.0 +3.8	35.7 -2.5	[∞] +	37.4 +2.2	37.3 +1.9	 +	37.2 +1.6
	Absolute totals	174 13,43	135 12,76	7 163 14,22	7 124 1	1,704	73 7,489	162 1	3,545
Stem.	Mean weight (in mg.) per seedling Percentage increase over H ₂ O	72.2 0	94.6 +22.5	87.3 +13.1	94.4	94.4 22.3	102.6 +32.9	∞∓	83.6 +8.3
	Absolute totals	174 3,88(135 3, 75	174 3,880 135 3,750 163 4,189	124	3, 633	124 3, 633 73 2, 301 162 4, 042	162	4,042
Root.	Mean weight (in mg.) per seedling Percentage increase over H ₂ O	22.3 0	27.8 +24.7	25.7 +15.2	29.3	29.3 31.4	31.5 +41.3	+ 11	24.9 11.7
Stem + root.	Mean weight (in mg.) per seedling Percentage increase over H ₂ O	99.5 0	122.4 +23.0	113.0 +13.6	123.7 +24.3	1 6	134.1 +34.8	101 101 101 101 101 101 101 101 101 101	108.5 +9.0

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TABLE	

higher, by the insignificantly small amount of 1 to 2 per cent in the 8, 12, and 16 per cent alcohol-treated series than in the H_2O series. But in the series treated with 4 per cent alcohol the mean fresh weight of the cotyledons is 2.5 per cent lower than in the water series. Taking all the data together it is plain that no significant effect upon the final

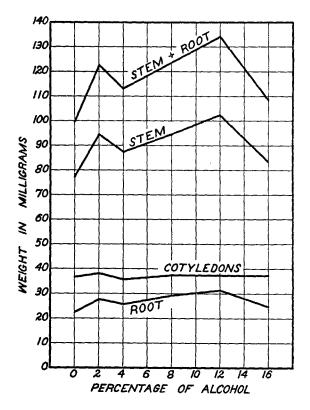


FIG. 2. Mean fresh weight of seedlings of *Cucumis melo* after complete growth under conditions of starvation.

fresh weight of the cotyledons after the completion of growth under starvation conditions was produced by a preliminary treatment of the dry seeds with alcohol.

2. The case is different with the stems. The final mean weight of this part of the seedling was higher in every case if the dry seed had been treated for 3 hours with alcohol than if it had been soaked in

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water only. The amount of the increased growth over that exhibited by the water-treated seeds varied from 8.3 per cent in the case of the 16 per cent alcohol treatment to 32.9 per cent in the case of the 12 per cent alcohol treatment. These are substantial increases.

3. The increased growth in the alcohol-treated seeds is even more marked in the case of the roots than in the stems. Here the percentage increases over the water series range from 11.7 per cent to 41.3 per cent. In other words soaking the dry seeds in a 12 per cent solution of ethyl alcohol for 3 hours prior to germination led to the production, in completed growth under starvation, of roots nearly half as large again as those produced by seeds treated in every respect identically except that they were soaked in water instead of 12 per cent alcohol.

4. Taking the weight of stems and roots together, as measuring the total real growth of the seedlings, since the cotyledons at this stage and under the conditions of these experiments are essentially nothing but food reservoirs, the same result emerges. The increases in the alcohol series over the water range from 9 to nearly 35 per cent.

5. There is a fairly steady rise of the curves up to 12 per cent alcohol, and a distinct drop in the 16 per cent series. The results so far obtained indicate that, under the conditions of these experiments, a 3 hour treatment with 12 per cent alcohol is near the optimum for subsequent growth. We do not, however, regard this as in any sense a final conclusion. The determination with precision of the optimum alcohol treatment demands considerable further experimentation. But the present results encourage one to look for it somewhere in the general region of concentrations of 10 to 15 per cent. The relatively high value at 2 per cent concentration, and relatively low one at 4 per cent we are for the present disposed to regard as probably due to chance errors, and likely to disappear in longer series. It seems probable that the influence of alcohol upon growth here observed is nearly a linear function of concentration up to volume concentrations of about 12 per cent. Again, however, we hope by further experiments to settle more definitely than is now possible the mathematical form of this functional relationship.

6. In spite of the greater effect of the alcohol treatment upon the

root than upon the stem, already noted, the stem/root ratio is not markedly altered in the different series. Thus:

													Ste	m u	eigh	t/Ro	ot we	ight
H_2	0				 		 •••	 		 	 	 				3.4	16	
2	per	cent	alcoho	ol	 	•••	 	 		 	 • •	 		• •		3.4	40	
T	"	"	"		 		 	 		 	 	 				3.4	40	
8	"	"	"		 		 	 		 	 	 				3.2	22	
12	"	"	"	•••	 	•••	 	 		 	 	 		•••		3.2	26	
16	"	"	"		 	•••	 	 • •	••	 ••	 ••	 •••		• •	•••	3.3	36	

In general the roots tend to become heavier relative to the stem in the alcohol series, and as the concentration increases up to 12 per cent at least, but the differences must be regarded as suggestive rather than conclusive until supported by further data.

Dextrose.

The following question occurs to one at this stage. Is the effect upon growth which has been demonstrated in the preceding section, to be regarded as specifically due to the treatment with ethyl alcohol as such, or is it simply a result of the different osmotic pressures of the several alcohol solutions in which the dry seeds were soaked? To get light on this point a series of experiments was tried with dextrose solutions isotonic with certain of the alcohol solutions, the conditions in every other particular being precisely the same as those in the alcohol experiments already described.

Through the kindness of Dr. W. B. D. Penniman, consulting chemist to this laboratory, and of Professor J. C. W. Frazer, chairman of the department of chemistry of the Johns Hopkins University we have the following data as to the osmotic relations of ethyl alcohol and dextrose.

Volume concentration of alcohol, per cent.	Mols/1000 gm. H_2O .	Osmotic pressure. (atmospheres).	Volume concentration of dextrose.
			per cent
2	0.33	7.9	5.94
4	0.706	16.45	12.71
8	1.528	36.0	27.50
16	3.32	79.5	59.76

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TABLE IV.	Mean Fresh Weight per Seedling. Dry Seeds Soaked 3 Hours in L
	Growth of Seedlings of Cucumis melo.

	TABLE IV.							
Growth of See	Growth of Seedlings of Cucumis melo. Mean Fresh Weight per Seedling. Dry Seeds Soaked 3 Hours in Dextrose Solutions.	Dry S	seeds	Soaked	1 3 H	ours in	Dextrose So	lutions.
Part	Item.	0°H		5.94 per cent dextrose.		12.71 per cent dextrose.	27.5 per cent dextrose.	59.76 per cent dextrose.
		L w.ii	Total weight in mg.	No. weight in mg.	cht No. Ig.	D. weight in mg.	No. weight in mg.	No. weight in mg.
	Absolute totals		45 1,773	45 2,0	100	5 1,941	45 2,001 45 1,941 45 1,756 45 1,960	45 1,960
Cotyledons.	Mean weight (in mg.) per seedling Percentage increase over H ₂ O	39.4 0	·	44.5 +13.0		43.1 +9.4		43.5 + 10.4
	Absolute totals		45 4, 552	45 4,6	560 4	45 4,660 45 4,699	45 4, 344	45 4,818
Stem.	Mean weight (in mg.) per seedling Percentage increase over H ₅ O	10	10.12 0	103.6 +2.4		104.4 +3.2	96.5 -4.6	107.1 +5.8
	Absolute totals		, 283	45 1,3	340 4	45 1,283 45 1,340 45 1,411	45 1,251	45 1,321
Root.	Mean weight (in mg.) per seedling Percentage increase over H ₂ O	28.5 0	ъ	29.8 +4.6		31.4 +10.2	27.8 -2.6	29.3 +2.8
Stem + root.	Mean weight (in mg.) per seedling Percentage increase over H ₂ O	129.7 0	7	133.3 +2.8		135.8 +4.7	124.2 -4.2	136.4 +5.2

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Solutions of C. P. dextrose of the indicated concentrations were made up with distilled water. The highest concentration is near the point of saturation for dextrose in water, and complete solution at room temperature was only attained after the addition of the dry seeds to the solution (possibly as result of an adsorption effect on the surface of the seeds). The seeds as before were soaked in these dextrose solutions for 3 hours, then washed off in distilled water, and from that point on treated exactly as in the alcohol experiments. The results for completed growth are shown in Table IV.

The absolute values of the means in the table are generally higher than those in Table III, presumably because the experiments in the dextrose series were done with a variety of cantaloupe having a larger seed (and seedling) than the variety which was chiefly used in the experiments detailed in Table III. Since we are here only interested in relative values within the series, however, the greater absolute size has no bearing upon the results.

It is obvious that the picture presented by Table IV is essentially different in character from that seen in Table III. There is no steady, systematic increase in the total growth associated with increasing concentration of the dextrose solutions. In the different concentrations the differences, plus or minus, between the different series in respect of total growth are of the order of 2 to 5 per cent change from the results shown by the controls soaked in distilled water only. The contrast between these and the 8 to 35 per cent increases shown in Table III is striking.

We are compelled to conclude that the changes observed in the alcohol series cannot be accounted for merely as the results of differences in osmotic pressure in the several solutions.

DISCUSSION.

The general result of the experiments reported in this paper may be summarily stated in this way. In a series of experiments so managed as to deprive seedlings of *Cucumis melo* of all food except that derived from their own cotyledons, the total growth (as measured by fresh weight) is markedly greater if the dry seeds are soaked for a period of 3 hours, before germination, in solutions of ethyl alcohol of from 2 to 12 per cent volume concentration than if they are soaked in distilled water. Parallel experiments with solutions of dextrose isotonic with the alcohol solutions demonstrate that the effects on growth observed in the alcohol series cannot be explained as due to osmotic differences. How then are these results to be interpreted? To this question we are not yet prepared to give any final answer. More experimental work is necessary and is in progress. It will, however, be profitable to consider at this time the chief alternative explanations which suggest themselves.

There are broadly two such alternatives. The first is that the excessively small amounts of alcohol which penetrated to the embryo and cotyledons, under the conditions of these experiments, had a true stimulating effect upon growth, either by influencing cell division directly, or indirectly through improving the metabolism of the stored cotyledonary food. The other alternative explanation is that the alcohol acted as a selective agent and eliminated weak seeds which, under any conditions, would have made a poor growth, leaving to grow in the alcohol series only the strongest and most vigorous seeds. The experimental data available at present are not entirely sufficient to enable a final decision between these alternatives to be made. It will be well, however, to examine the existing evidence in favor of both hypotheses, considering them separately.

The results we have obtained with alcohol in this work are in some respects strikingly analogous with those recorded in a mass of horticultural literature which exists regarding the use of ether vapor in the forcing of plants. In 1900 Fischer (17) reported for a number of species that if, in their true resting stage, the plants were placed in a closed chamber filled with ether vapor for a period of 48 hours, and then removed to a warm room, they would be forced into blossom considerably earlier than in the absence of this treatment, or by any other known method of culture. Jannock (18), Rüde (19), Maumené ((20) and various other papers), Bellair ((21) and various other papers), Charmeux (22), Bolle (23), Lochat (24), Drude, Naumann, and Ledien (25), Aymard (26), Marble (27), Stuart (28), Lewis (29), Taubenhaus (30), Howitt (31), Ledien (32), and various other persons, obtained essentially the same results, using a wide variety of species. Furthermore it appeared in this work that in many cases not only was the plant forced into earlier blooming, but actually a sturdier

and more vigorous growth was induced. Cook and Wilson (33) found that small quantities of ether in liquid culture media have a stimulating effect on the growth of *Endothia parasitica*. Howard and Whitten (34) reported, after extensive experimentation, that etherization tended to stimulate seeds into early growth and to increase the percentage of germination. All of the earlier horticultural work on forcing by etherization was entirely empirical and merely established the fact of stimulation. Indeed the physiology of the matter is still far from being fully understood. Perhaps the most significant lead in this direction is that the treatment alters the metabolism by affecting the quantitative relations of the enzymes. Thus McCool (35) showed that in bulbs and tubers diastatic activity, and the activity of oxidases and peroxidases was greater following etherization, whereas catalase activity was diminished.

One important respect in which the analogy of the present experiments with this etherization work, which has been briefly reviewed, seems to us to be close is found in the fact that in both cases the amount of substance (alcohol or ether) which comes actually into contact relations with the living cellular elements of the plant must be extremely minute. These cases are very different from those in which the agent is continuously in contact with the plant during growth. It has long been known that a great range of toxic substances may, in proper dilutions, act as stimulating agents to plant growth. The classical work here is that of Bokorny (36). The same idea has lately been revived by Popoff ((37) and various other papers). But generally in work in this field the agent has acted continuously during the growth period observed.

In reviewing this earlier literature it has seemed to us that in many cases the possibility of a simple selective action of the deleterious agent was entirely overlooked both in the conduct and in the interpretation of the experiments. The work of Pearl (7-10), Danforth (11), and Stockard (12-14) seems to have demonstrated conclusively that ethyl alcohol has just such a selective action both on germ cells, and on somatic cells (in the fowl). It has been shown that a dosage is attainable which kills off constitutionally weak and defective cells, without injuring sound and vigorous cells. The whole action is a graded one, and the zone in which a clean-cut, purely selective, action

occurs is generally not wide. As one passes beyond this restricted zone of dosage towards higher concentrations the tendency is not only to eliminate the constitutionally weakest, but also at the same time to injure more and more the strong.

We incline at present to the opinion that such a selective action is the most probable explanation of the results here reported. Soaking the seeds for 3 hours in 12 per cent ethyl alcohol eliminates about 20 per cent of the viable seeds by preventing their germination. It seems a reasonable inference that the seeds so eliminated are constitutionally the weakest, least resistant, and least vigorous of the whole population. If this is so it follows necessarily that the 80 per cent which do germinate have a higher average of strength and vigor (or sounder organization) than the whole population of viable seeds taken together. Furthermore it will scarcely be questioned that a constitutionally sound, normally organized seed will grow more vigorously than a weak, abnormal seed. It must then follow that the average growth of the seeds surviving the alcohol treatment at the appropriate level of concentration (apparently about 12 per cent) will be greater than the average growth of all viable seeds, as exhibited in the water controls. The present experiments show that so, in fact, it is. Treatment for 3 hours with 16 per cent alcohol would seem to have passed the optimum zone of efficient selective action. More seeds are eliminated by 16 per cent than by 12 per cent. But apparently the survivors are in some degree injured.

SUMMARY.

In this paper it is shown that if the dry seeds of the cantaloupe (*Cucumis melo*) are soaked for 3 hours in solutions of ethyl alcohol of concentration ranging from 2 to 16 per cent by volume, and then germinated and grown in distilled water in the dark, the total growth attained is greater by amounts ranging from 9 to 35 per cent than is that made by seeds treated in every way identically except that they are initially soaked in distilled water instead of alcohol. It is shown that this result is not due simply to differences in osmotic pressure in the different alcohol solutions. It is probably due to a simple selective action of the alcohol which eliminates the constitutionally weak and defective seeds.

BIBLIOGRAPHY.

- 1. Pearl, R., J. Exp. Zool., 1917, xxii, 165.
- 2. Stockard, C. R., and Papanicolaou, G. N., J. Exp. Zool., 1918, xxvi, 119.
- 3. Pearl, R., Am. J. Hyg., 1922, ii, 463.
- 4. Pearl, R., Alcohol and mortality, in Starling, E. H., The action of alcohol on man, London, 1923.
- 5. Pearl, R., Proc. Nat. Acad. Sc., 1924, x, 231.
- 6. Pearl, R., Brit. Med. J., 1924, i, 948.
- 7. Pearl, R., Proc. Am. Phil. Soc., 1916, lv, 243.
- 8. Pearl, R., Proc. Nat. Acad. Sc., 1916, ii, 675.
- 9. Pearl, R., J. Exp. Zool., 1917, xxii, 241.
- 10. Pearl, R., Eugenics Rev., 1924, xvi, 9.
- 11. Danforth, C. H., J. Exp. Zool., 1919, xxviii, 385.
- 12. Stockard, C. R., Brit. Med. J., 1922, ii, 255.
- 13. Stockard, C. R., Proc. Am. Phil. Soc., 1923, lxii, 311.
- 14. Stockard, C. R., Am. J. Med. Sc., 1924, clxvii, 469.
- 15. Pearl, R., and Parker, S. L., Am. Naturalist, 1924, lviii, 193.
- 16. Pearl, R., Nature, 1924, cxiii, 854.
- 17. Fischer, J., Am. Gardener, 1900, xxi, 358, 372.
- 18. Jannock, T., Gardener's Chron., xxxiv, 3 ser. 1903, 240.
- 19. Rüde, K., Möllers deutsch. Gärt. Ztg., 1904, xix, 50.
- 20. Maumené, A., Jardin, 1904, xviii, 42.
- 21. Bellair, G., Rev. Hort., 1904, lxxvi, 84.
- 22. Charmeux, F., Jardin, 1904, xviii, 188, 189.
- 23. Bolle, J., Z. Landw. Versuchsw. Oesterr., 1904, vii, 182, 183.
- 24. Lochat, J., Rev. Hort., 1904, lxxvi, 250.
- 25. Drude, A., Naumann, A., and Ledien, F., Jahresber. Flora, 1902-03, vii.
- 26. Aymard, J., Les anesthésiques et le forçage des plantes, Paris, 1904, 68.
- 27. Marble, F. L., Garden Mag., 1905, ii, 64, 65.
- 28. Stuart, W., Vermont Agric. Exp. Station Rep., 1904, 418.
- 29. Lewis, C. I., Cornell Countryman, 1906, iii, 190, 191.
- 30. Taubenhaus, J., Cornell Countryman, 1907, iv, 254.
- 31. Howitt, J. E., Cornell Countryman, 1906, iii, 187, 188.
- 32. Ledien, F., Möllers deutsch. Gärt. Ztg., 1906, xxi, 530.
- 33. Cook, M. T., and Wilson, G. W., Bot. Gaz., 1915, lx, 412, 413.
- 34. Howard, W. L., and Whitten, J. C., Missouri Agric. Exp. Station Bull. 117, 1914, 427.
- 35. McCool, M. M., Science, 1914, xxxix, 261.
- 36. Bokorny, T., Biochem. Z., 1913, l, 1-118.
- 37. Popoff, M., Biol. gen., 1925, i, 52.