

TEXAS AGRICULTURAL EXPERIMENT STATION

BULLETIN NO. 174

APRIL, 1915

DIVISION OF CHEMISTRY

The Effect of Organic Compounds in Pot Experiments



POSTOFFICE:
COLLEGE STATION, BRAZOS COUNTY, TEXAS



VON BOECKMANN-JONES CO., PRINTERS, AUSTIN, TEXAS

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BY

G. S. FRAPS,
Chemist in Charge; State Chemist.



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EFFECT OF ORGANIC COMPOUNDS IN POT EXPERIMENTS.

BY G. S. FRAPS, PH. D., CHEMIST IN CHARGE; STATE CHEMIST.

It has been known for a long time that certain kinds of plants do poorly, or do not grow at all on acid soils. It is believed that some of these acid soils contain organic acids. The addition of organic or inorganic acids has been known to affect injuriously the growth of plants, both those grown in soils and those grown in water culture solutions.

Schreiner and Shorey (Bulletin 53, Bureau of Soils, 1909) state that di-hydroxystearic acid is injurious to wheat seedlings in water culture solutions. As little as ten parts per million injured the growth. These results were secured with an acid isolated from soils. In Bulletin No. 70 of the Bureau of Soils, 1910, Schreiner and Skinner state that di-hydroxystearic acid injures the growth of wheat plants, both in distilled water alone, and in the presence of nutrient or fertilizer salts. It is the more harmful in those ratios of fertilizer elements not well suited for plant growth. The quantity of di-hydroxystearic acid used was 50 parts per million and the solution was changed every three days.

Schreiner and Skinner, in Bulletin No. 77 of the Bureau of Soils, 1911, state that cumaron is quite harmful to plants in water solution in minute amounts, a few parts per million having a noticeable effect on plant growth. Plants in the solution containing 250 parts per million were dying at the end of five days, and those in the solution containing 100 parts per million were dead at the end of eight days. Ten parts per million were found to be injurious. Vanillin killed the plants in 500 parts per million in nine days. Harmful results were secured with vanillin at the rate of 50 parts per million and even as low as one part per million.

Quinone was one of the most active poisons employed in another series of experiments. Wheat plants were killed in solutions containing 100 parts per million and seriously injured in solutions containing 50 parts per million. Ten parts per million produced harmful effects. The vanillin, cumaron, and quinone were found to be injurious to plants in water solution and various fertilizer salts assisted in overcoming the harmful effects of these substances. Schreiner and Skinner in Bulletin No. 87 of the Bureau of Soils, December, 1912, studies the effect of a number of other organic compounds upon plants in water solution. Some of these were found to be decidedly injurious. In Bulletin No. 108 of the Department of Agriculture, 1914, Schreiner and Skinner report some experiments in which salicylic aldehyde was found to be injurious to plants in water culture, and in field experiments. In Bulletin No. 36 of the Bureau of Soils, 1907, Livingston states that the water extract of certain soils contain injurious substances which are neutralized by pyrogallol, or carbon black.

POT EXPERIMENTS.

The experiments described in this bulletin were carried out with soils contained in galvanized iron pots, grown in the plant houses by the usual methods employed by us. The quantity of soil contained in these pots was not always the same. In the earlier experiments there was from 5000 to 6000 grams of soil, but in later experiments the quantity of soil taken was uniformly 5000 grams. The pots were watered three times a week, water being added each time sufficient to make the water content one-half of the water capacity of the soil. The pots were weighed and sufficient water added to bring the weight up to the required amount. The experiments reported were for the most part made in 1912.

The object of the first series of experiments on several soils was to test the harmfulness of the various organic substances in pot experiments, when added at the time of planting.

EXPERIMENTS WITH DI-HYDROXYSTEARIC ACID.

Impure di-hydroxystearic acid was prepared by the oxidation of elaidic acid with permanganate according to the usual method. It was applied to the pots at the rate of 500 parts per million of soil, which should be amply sufficient to demonstrate its effects. On one pot, 1200 parts per million was added. Two soils were used. On one, No. 1600, no additions were made, except the di-hydroxystearic acid or the acid with carbonate of lime. The carbonate of lime was added in sufficient amount to just neutralize the acid, as ascertained by titrating some of the acid with caustic soda and phenolphthalein. On soil No. 1956, additions of acid phosphate, sulphate of potash and nitrate of soda were made. The results of the experiments are given in Tables 1 and 2.

TABLE 1.—DRY WEIGHTS OF CROPS GROWN ON SOIL NO. 1956.

Laboratory Number.		Corn.	Sorghum.	Total.	Average.
1—O	2.5 gms Acid Phosphate (P) 1.0 gms.	18.0	10.5	28.5	24.5
2—O	Nitrate of Soda K. 1.0 gm. Sulphate of Potash	5.5	15.0	20.5	
3—Ac.	Ditto and 2.5 gms. Dihydroxystearic Acid	15.1	10.8	25.9	25.5
4—Ac.	Ditto and 2.5 gms. Dihydroxystearic Acid	15.1	10.1	25.2	
5—Ac. Ca.	Same as 3, with 0.4 gm. Calcium Carbonate	16.2	7.4	23.6	22.8
6—Ac. Ca.	Same as 3, with 0.4 gm. Calcium Carbonate	13.8	8.3	22.1	
	Planted (1909)	May 3	July 5		
	Harvested	June 18	Aug. 16		
	*Injured by excess of water.				

In previous soil work we have allowed a variation of 10 per cent of the weight of the crops to go without consideration.

With soil No. 1600, 500 to 1200 parts per million of impure di-hydroxystearic acid had no injurious effect upon either the corn or sorghum. In fact, the crops grown on the pot receiving 1200 parts per million were larger than those grown in the check pots. On soil No. 1956, the di-hydroxystearic acid decreased the crop of corn slightly,

and possibly within the limit of variation. It had little effect on the sorghum crop. On both soils the addition of calcium carbonate to the dihydroxystearic acid decreased the growth of the crops.

TABLE 2.—WEIGHT OF CROPS IN GRAMS ON SOIL NO. 1600.

Laboratory Number.	Additions.	Corn.	Sorghum.	Total.	Average.
1—O	No addition.....	9.3	20.9	30.2	} 26.9
5—O	No Addition.....	10.1	13.5	23.6	
2—Ac	2.5 gms. of Dihydroxystearic Acid.....	8.8	19.0	27.8	
3—Ac Ca	Ditto and 0.4 gms. Calcium Carbonate.....	5.6	19.9	25.5	
4—2 Ac	6 gms. of Dihydroxystearic Acid.....	10.7	22.5	33.2	
	Planted (1909).....	May 3	July 5		
	Harvested (1909).....	June 18	Aug. 16		

According to these experiments, impure di-hydroxystearic acid has little injurious effect upon corn or sorghum when applied at the rate of 500 parts per million to the soil in pot experiments. In the water culture experiments previously cited, 10 parts per million injured the growth.

EXPERIMENTS WITH VANILLIN, QUINONE AND CUMARON.

A number of series of experiments were made with vanillin, cumaron and quinone. The basal quantity used was 100 parts per million, equal to 0.50 grams for a pot weighing 5000 grams. This quantity killed the plants in water culture solution in experiments of Schreiner et al., previously cited. The experiments were conducted as previously described.

Additions at Time of Planting.—In these experiments the additions were made at the time of planting the seed, and well worked into the soil. The weights of the dried crops are given in Tables 3 and 4. These weights, as all given in this bulletin, are based upon the weights of the plants after they had been harvested and dried for several days in a drying oven.

According to these results, vanillin and quinone at the rate of 100 parts per million of soil are injurious in only one of eight or nine soils. Quinone at the rate of 200 parts per million is not injurious in two soils, injurious in one. Vanillin added at the rate of 200 parts per million is injurious in each case. Cumaron is injurious in six out of nine experiments with 100 parts per million, five of seven with 200 parts, and one of two with 300 parts. The maximum depression observed is about 40 per cent with soils 4605 and 4689, and 30 per cent with soil 4797. As cumaron and quinone killed wheat in water culture solutions in eight or ten days, when present at the rate of 100 parts per million, it is obvious that these substances, when added to the soil, at the time of planting the seed, are much less injurious than in water culture, since even 300 parts per million did not kill the plants.

The substances were added to the pots shown in Table 5 at the rate of 500 to 2000 parts per million of the soil. Vanillin, while injurious, did not kill the plants, even in the single soil on which it was applied

at the rate of 2000 parts per million. Cumaron killed the plants, while quinone did not, both being tried on one soil each, and at the rate of 500 parts per million.

TABLE 3.—CORN IN GRAMS PER POT.

Laboratory Number.	Additions.	4605	4689	4747
1—O	None.....	11.2	13.2	26.8
5—O	None.....	11.9	11.3	30.5
9—O	None.....	12.5	14.4
Average—O	11.8	13.0	28.6
2—V	Vanillin 100 parts per million.....	10.1	13.1	26.3
6—2V	Vanillin 200 parts per million.....	9.1	8.9	22.8
10—3V	Vanillin 300 parts per million.....	14.2	10.0
Average.....	11.1	10.7	24.6
4—Q	Quinone 100 parts per million.....	12.0	15.1	26.6
8—2Q	Quinone 200 parts per million.....	12.3	14.6	20.9
12—3Q	Quinone 300 parts per million.....	13.5	8.0
Average.....	12.6	12.6	23.8
3—C	Cumaron 100 parts per million.....	7.1	10.2	20.2
7—2C	Cumaron 200 parts per million.....	9.9	10.4	22.5
11—3C	Cumaron 300 parts per million.....	14.3	8.0
Average.....	10.4	9.5	21.8
	Planted.....	April 10	April 10	April 10
	Harvested.....	June 26	June 26	June 26

TABLE 4.—SORGHUM IN GRAMS PER POT.

Laboratory Number.	Additions	4644	5648	1577	1594	3339	3346
1—O	No addition.....	15.8	11.6	20.5	16.3	19.5	16.2
2—V	Vanillin 100 parts per million.....	15.1	10.5	19.7	16.1	20.0	10.7
3—C	Cumaron 100 parts per million.....	14.8	11.7	21.2	16.9	19.4*
4—Q	Quinone 100 parts per million.....	14.3	11.1	17.7	12.6	20.7	15.7
5—2V	Vanillin 200 parts per million.....	13.1	10.5	14.9	13.0
6—2C	Cumaron 200 parts per million.....	13.3	10.8	21.7	10.9
	Date harvested.....	Aug. 3	Aug. 3	July 29	Aug. 20	Aug. 13	Aug. 23
	Date planted.....	June 7	June 7	June 7	June 7	June 7	June 7
	*Infected with worms June 22, dead June 29.

Disappearance of the Substance.—The odor of vanillin disappeared very rapidly from the pots. For this reason, at the end of the experiment, the vanillin and cumaron was estimated in pots to which 300 parts per million had been added. The method used is as follows:

Soil equivalent to 100 grams of the dry soil was heated with 200 c.c. water on the steam bath under the reflux condenser for thirty minutes. It was then filtered and washed three or four times with water. The aqueous extract was extracted with three successive portions of ether. This ether was allowed to evaporate spontaneously at room temperature in an evaporating dish, transferred to a weighed beaker with ether, allowed to evaporate at room temperature, dried in a vacuum desiccator and weighed. These soils had received 300 parts per million. The results are as follows in parts per million:

Soil 4605—Vanillin, 8 parts per million; cumaron, 9 parts per million.

Soil 4689—Vanillin, 9 parts per million; cumaron, 12 parts per million. Average, 9 parts per million ether soluble material recovered.

We would judge from this that the vanillin and cumaron are rapidly oxidized in the normal soil.

In order to study further the loss of vanillin and cumaron in the soil, the following experiment was carried out:

Five hundred grams of soil were mixed with 0.5 gram cumaron or 0.5 gram vanillin, water added equal to one-third of the saturation capacity, again mixed, and allowed to stand two weeks. At the end of that time one-fifth of the weight (equal to 100 grams dry soil) was weighed out, and heated with 200 c.c. water on a steam bath under a reflux condenser for about thirty minutes, filtered and washed with hot water. The aqueous extract was extracted with ether, allowed to evaporate spontaneously at room temperature, and transferred with ether to a weighed beaker. The ether was allowed to evaporate, the residue dried in a vacuum, and weighed.

A similar experiment was made with two of the original soils, using, however, 100 grams soil, 0.1 gram material, and extracting at once with water.

The results are presented in Table A. It is evident that these substances are rapidly oxidized in the soil.

TABLE A.—LOSS OF VANILLIN AND CUMARON

	3346	4605	4747
Vanillin Added (grams).....	0.1	0.1	0.1
Recovered from original soil.....	0.801	0.835
Recovered after two weeks.....	0.557	0.0082	0.0231
Vanillin lost (grams).....	0.0244	0.0718	0.0604
Cumaron added (grams).....	0.1	0.1	0.1
Recovered from original soil.....	0.0781	0.0733
Recovered after two weeks.....	0.0613	0.0363	0.0027
Cumaron lost (grams).....	0.0168	0.0237	0.0706

Additions Made After Planting.—On account of the disappearance of the substance from the soil, in subsequent experiments, the organic material was added after the seed had come up, and the plants had begun to grow, which was a week or ten days after planting. In the results given in Table 6, the addition was repeated for each subsequent week until the plants died, or were harvested. Thus, six additions were made to soils 1739 and 5939, seven to soil 3337. Each of these additions consisted of 100 or 200 parts per million or a total of 600 to 1400 parts per million. Applied even in these large quantities, the vanillin was not decidedly injurious except on soil 3337 at the rate of 200 parts per million. The cumaron was injurious on all the soils, and killed the plants at 200 parts per million on soil 3337. The quinone was decidedly injurious on two soils, and to some extent on the third. It

will be recalled that the additions were made every week for the purpose of giving the organic substances an opportunity to exert their injurious action upon the plants before they were oxidized by the soil.

In the experiments of Schreiner et al., previously cited, 100 parts per million of cumaron or quinone killed the plants in water culture in eight to ten days. Vanillin at the rate of 500 parts per million killed the plants in nine days. In the experiments described above, six successive additions of 100 parts per million of vanillin, cumaron, or quinone, did not kill any of the plants, and only with cumaron did successive additions of 200 parts per million kill the plants. After making due allowance for the fact that different plants were grown on the soil from those used in water solution, it yet appears that these organic substances are much less toxic in the soil than in water solution.

TABLE 5.—WEIGHT DRY MATTER GRAMS PER POT WITH ADDITIONS

Laboratory Number.	Addition.	Sorghum. 5647	Corn. 2409	Corn. 3343
1—O	No addition.....	7.4	20.1	21.2
2—5V	Vanillin 500 parts per million.....	6.9	15.9	17.9
3—10V	Vanillin 1,000 parts per million.....	7.7
4—20V	Vanillin 2,000 parts per million.....	5.5
5—5C	Cumaron 500 parts per million.....	0
6—10C	Cumaron 1,000 parts per million.....	0
5—Q	Quinone 500 parts per million.....	19.7
	Planted.....	June 25		
	Harvested.....	Aug. 23		

TABLE 6.—SORGHUM IN GRAMS PER POT.

Laboratory	Addition.	1739	3337	5939
O	No addition.....	22.5	18.9	10.2
V	Vanillin 100 parts per million.....	14.1	8.7
C	Cumaron 100 parts per million.....	2.1	5.2
Q	Quinone 100 parts per million.....	0.8	8.4
2V	Vanillin 200 parts per million.....	30.9	8.8	9.5
2C	Cumaron 200 parts per million.....	19.6	0	7.9
2Q	Quinone 200 parts per million.....	3.2
	Planted.....	July 1	July 1	July 1
	Harvested.....	Aug. 13	Aug. 20	Aug. 13

EFFECTS OF FERTILIZERS UPON THE INJURIOUS ACTION.

It has been ascertained, in a previous experiment, that the vanillin, cumaron and quinone had little action when added at the time of planting (probably being rapidly destroyed by the soil), and, on the other hand, when added after the plants were up, they had some injurious action as they had opportunity to affect growing plants before they were oxidized in the soil. The next series of experiments has for their object to see if fertilizers in connection with the injurious substance, decreased or overcome its injurious effect. The results are given in tables which follow.

The organic substance was always added after the plant had come up and begun to grow.

In Table 7, phosphoric acid and nitrogen do not overcome the in-

injurious action of 200 or more parts per million of vanillin. One hundred parts is injurious to two of the three soils.

In Table 8, 200 parts per million of cumaron is shown to kill the plants in spite of addition of ammonium nitrate.

In Table 9, 100 parts per million of quinone is seen to kill the plants, in spite of addition of acid phosphate or nitrate of soda.

In Table 10, there is no evidence of phosphoric acid or nitrogen overcoming the injurious action of vanillin, except, possibly, in soil 5938.

These experiments do not offer any good evidence that the addition of phosphoric acid or nitrogen to the soil, assists in overcoming the injurious effects of the organic substances, or at least that this is not the primary function of these substances in the soil. Of course, as a general principle, a strong plant should be less susceptible to injurious influence than a weak one.

THE EFFECTS OF CARBON BLACK AND PYROGALLIC ACID.

It has been claimed that the aqueous extract of soils contains substances which are injurious to plant growth, and that these substances are removed or neutralized by treatment of the soil by carbon black, or by pyrogallie acid. It has also been claimed that the action of fertilizers is not due to the plant food that they carry, but to their neutralizing toxic substances in the soil.

TABLE 7.—GRAMS, SORGHUM PER T.

Laboratory Number.	Additions.	5101	5649	1579
1—PN	2 gms. Acid Phosphate, 1 gm. Amm. Nitrate.....	11.8	9.2	10.5
2—PNV	2 gms. Acid Phosphate 100 parts per million Vanillin.....	5.1	8.7	6.9
3—PN2V	2 gms. Acid Phosphate 200 parts per million Vanillin.....	0	3.4	3.7
4—PN5V	2 gms. Acid Phosphate 500 parts per million Vanillin.....	0	0.6	2.2
5—PN10V	2 gms. Acid Phosphate 1,000 parts per million Vanillin.....	0	0	0
6—PN20V	2 gms. Acid Phosphate 2,000 parts per million Vanillin.....	0	0	0
	Planted.....	July 19	July 19	July 19
	Harvested.....			

TABLE 8.—GRAMS, SORGHUM PER POT.

Laboratory Number.	Addition.	5650
1—N	1 gm. Ammonium Nitrate.....	3.8
2—NC	1 gm. Ammonium Nitrate and 100 parts Cumaron per million.....	4.1
3—N2C	1 gm. Ammonium Nitrate and 200 parts Cumaron per million.....	0
4—N5C	1 gm. Ammonium Nitrate and 500 parts Cumaron per million.....	0
6—PN	1 gm. Ammonium Nitrate and 2 gms. Acid Phosphate.....	9.5
7—PN5C	1 gm. Ammonium Nitrate, 2 gms. Acid Phosphate and 500 parts Cumaron per million.....	0

TABLE 9.—WEIGHT, SORGHUM IN GRAMS.

Laboratory Number.	Additions.	3657	3975	5099
1—O	No addition.....	0.7	0.7	0.3
2—Q	100 parts Quinone per million.....	0	0	0
3—PN	2 gms. Acid Phosphate and 1 gm. Nitrate of Soda.....	9.9	10.6	3.1
4—PNQ	2 gms. Acid Phosphate, 1 gm. Nitrate of Soda and 100 parts of Quinone per million.....	0	0	0
5—N	1 gm. Nitrate of Soda.....	0.9	1.6	0.4
6—NQ	1 gm. Nitrate of Soda and 100 parts Quinone.....	0	0

[illegible]

TABLE 12.—WEIGHT OF SORGHUM IN GRAMS.

Laboratory Number.	Additions per 5000 gm. pot.	5710	5945	5947	5948
1—O	No addition.....	2.2	2.6	4.1	2.1
2—P	1.0 gm. Acid Phosphate.....	6.6	1.0	4.1	2.2
3—PN	1 gm. Acid Phosphate, 1 gm. Ammonium Nitrate.....	8.2	9.3	7.7	5.4
4—PNK	1 gm. Acid Phosphate, 1 gm. Ammonium Nitrate and 1 gm Sulphate Potash.....	8.6	10.9	9.5	6.3
5—B	8.7 gms. Carbon Black.....	2.7	2.7	2.9	2.4
6—Y	.87 gms. Pyrogallic Acid.....	2.8	2.6	2.6
7—PB	1 gm. Acid Phosphate and 8.7 gms. Carbon Black.....	8.0	2.4	3.9	1.8
8—PN2V	Acid Phosphate, Ammonium Nitrate, 200 Vanillin.....	6.2	3.9	10.6	4.0
9—B2V	Carbon Black, 200 Vanillin per million.....	3.3	0.7	2.6	0.1
10—PNK2V	Acid Phosphate, Ammonium Nitrate, Vanillin.....	3.6	4.2	8.3	0
11—P2V	Acid Phosphate, Vanillin.....	5.4	0	3.9	4.5
12—2V	Vanillin, 200 parts per million.....	3.6	0.1
	Planted.....	July 22	July 22	July 22	July 22
	Harvested.....	Aug. 24	Aug. 24	Aug. 24	Aug. 24

ACKNOWLEDGMENT.

Assistance in the conduct of these experiments was given by Messrs. J. B. Rather and J. B. Kelly, assistant chemists.

SUMMARY AND CONCLUSIONS.

1. Impure di-hydroxystearic acid has little injurious effect upon corn or sorghum grown in pot experiments, when applied before planting at the rate of 500 parts per million of soil. In water culture experiments of other workers, 10 parts per million injured the growth of seedlings.

2. Vanillin and quinone, applied to the soil before planting at the rate of 100 parts per million of soil, injured the growth of only one of eight crops. In water culture experiments by other workers, cumaron and quinone killed the plants in eight or nine days, when present at the rate of 100 parts per million.

3. Vanillin and cumaron are oxidized in the soil, a considerable portion disappearing in two weeks, and little remaining at the end of the pot experiments in which the tests were made.

4. Six successive additions of cumaron, vanillin, or quinone at the rate of 100 parts per million, did not kill the plants.

5. Vanillin, cumaron or quinone are much less injurious in the soil, than they were in water culture experiments of others.

6. There was little evidence that fertilizers overcome the injurious action of cumaron, vanillin or quinone.

7. Pyrogallic acid and carbon black showed no beneficial action in pot experiments, while acid phosphate or other fertilizer are decidedly beneficial to the soils and produced decided increases. The conclusion is that these poor soils need the plant food supplied by the fertilizers, and that the action of the fertilizer is to supply plant food and not to overcome toxic substances.

8. Results of pot experiments may be quite different from results of water culture experiments.