measurement bromothymol blue has a color-change interval of pH 6.0 to 7.6. At a dilution factor of 0.5 an error of 0.4 pH unit is shown. An experienced observer should be able to determine pH much closer than this with color standards. At a dilution factor of 0.2 this error is only 0.1 pH unit, which is well within the range of sensitivity of the visual method of measurement.

On the basis of these results, it is recommended that for measurement of the pH of an 8–24–0 liquid fertilizer with color-change indicators, the sample should be diluted with four volumes of distilled water. The buffer capacity of this system is sufficient to allow this degree of dilution as can be seen from true pH values of Figure 1. Of course, for closest control of the product the use of a pH meter is recommended.

A current technical sales bulletin (*6*) recommends the use of universal indicator papers and dilution of the sample with an equal volume of distilled water. As shown above, this is insufficient dilution to overcome the salt effect. The error of 0.4 pH unit thus introduced could reduce solubility of the salts at 32° F. some 20% as shown elsewhere in the same bulletin (*7*).

Acknowledgment

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GROWTH RETARDANTS AND PLANT VIGOR

Increasing Tolerance of Soybean Plants to Some Soluble Salts through Application of Plant Growth-Retardant Chemicals

PAUL C. MARTH AND J. RAY FRANK

Crops Research Division, Agricultural Research Service, U. S. Department of Agriculture, Beltsville, Md.

Plants that were altered through the use of plant growth retardants, such as Amo-1618, phosfon, or CCC, were more tolerant to toxic levels of salts placed around their roots than were plants grown without retardants. Young soybean plants retarded chemically were able to withstand, without visible injury, amounts of commercial 5–10–5 fertilizer applied to the surface of the soil that killed comparable plants not treated with the retardants. Retarded plants that withstood excessive amounts of fertilizer produced viable seeds.

Considerable interest has been shown recently in plant-regulating chemicals that retard the height of plants without injurious effects. In 1949 Mitchell, Wirwille, and Weil (*5*) reported the growth-retarding effects of six related nicotinium compounds. In 1950, six quaternary ammonium compounds reported to be growth retardants for bean plants (*10*) were later found to be very active on several other plant species (*4*). The most promising compound, 2-isopropyl-4-dimethylamino-5-methylphenyl-1-piperidine carboxylate methyl chloride, was designated Amo-1618 and methods of synthesizing it and related retardants were recently published (*2*).

A new group of growth-retarding chemicals, the phosphoniums, was reported in 1958 by Preston and Link (*6*). One of these, tributyl- 2,4-dichlorobenzyl phosphonium chloride (phosfon), affects some species of plants that do not respond to Amo-1618. Another group of growth retardants was reported more recently by Tolbert (*9*). One of these, (2-chloroethyl)-trimethyl ammonium chloride (CCC), is effective in retarding some plants not affected by Amo-1618 or phosfon (*7*).

Interest in our laboratory has been directed toward other types of responses besides the dwarfing induced by these chemicals. For instance, the leaves of treated plants were usually thicker and greener and, in a number of experiments treated bean and chrysanthemum plants lived 40 to 60% longer than did untreated ones (*4*). When water was withheld from plants in other experiments, the untreated plants wilted more quickly and severely than did the treated ones (*3*). Recently, Teubner and O'Keefe (*8*) reported that certain varieties of potatoes were more resistant to heat after treatment with CCC or related compounds.

The purpose of the present study was to learn whether chemically retarded plants could withstand higher concentrations of salts in the soil than unretarded ones.

Methods

Seeds of soybean (Lee variety) were sown in composted soil in 3-inch clay pots and allowed to germinate. The plants were thinned to two per pot and selected for uniformity when about 4 inches tall. In most instances the growth-regulating chemicals were applied when the primary leaves were almost fully expanded and the trifoliolate ones were about 1/2 inch long. One experiment was conducted with older plants 8 inches tall and with two sets of fully expanded trifoliolate leaves. Amo-1618 was applied either as an aqueous spray to the foliage or as a soil drench. Phosfon was applied as a soil drench to the roots, because it is known to be phytotoxic when applied to leaves. CCC also was applied to the soil, because it is not readily translocated out of leaves. No wetting agent was used in preparing the drenches, but all spray solutions contained 0.1% Tween 20. The drench was applied at the rate of 30 ml. per pot. This volume contained sufficient retardant to make the rate of application equivalent to 15 pounds per acre.

A commercial 5–10–5 fertilizer from a single lot was used throughout the experiments to make the soil-fertilizer mixtures. The nitrogen content of the

Table I. Injury Caused by Application of Toxic Amounts of 5–10–5 Fertilizer to Soil around Soybean Plants

Fertilizer Rate		Degree of Injury Due to Fertilizer Application[a]	
G./pot	Lb./acre	Unretarded plants	Retarded plants
4	7,848	+++	0
5	9,810	++++	0
6	11,772	++++	++
7	13,734	++++	+++
8	15,696	++++	++++

[a] O = no injury; + = slight; ++ = moderate; +++ = severe, plants recovered; ++++ = all plants killed.

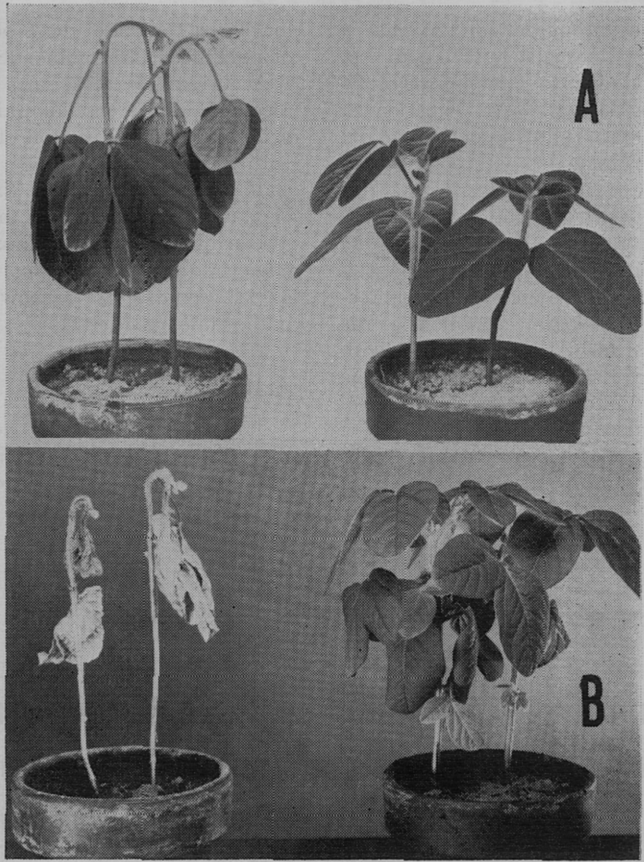

Figure I. Effect of application of 5 grams of 5–10–5 fertilizer to soybean plants

Left. Unretarded plants. *Right.* Chemically retarded plants *B.* Same plants 3 weeks after application

fertilizer was supplied by ammonium nitrate (3%), ammonium sulfate (1.5%), and sodium nitrate (0.5%); the phosphorus by superphosphate; and the potassium by potassium chloride.

Weighed fertilizer mixture was spread evenly over the soil surface. The soil surface was about $^1/_2$ inch below the rim of each pot. Thirty milliliters of water was required to fill this space above the soil. It was necessary to water twice daily to maintain optimum soil moisture.

In all instances the treated plants were allowed to develop visible growth responses to the regulators, which required 7 to 12 days, before the fertilizer mixture was applied. The intensity of visible growth responses varied with the kind and amount of growth regulator used and the age of the plant.

Results and Discussion

Wilting became apparent on the unretarded plants within 24 hours after application of the fertilizer. Even the lowest rate of fertilizer application (4 grams per pot) produced this effect. Unretarded plants that received 4 grams of fertilizer per pot subsequently lost about 50% of their leaves but later recovered and grew to maturity. Wilting of the other unretarded plants increased in proportion to the amount of fertilizer applied (Figure 1, *A*). All the unretarded plants that received 5 or more grams of the fertilizer ultimately died as a result of the fertilizer treatment (Table I and Figure 1, *B*).

In contrast, plants that were first retarded with Amo-1618 and then received 4 or 5 grams of fertilizer per pot showed no injury. These plants developed dark-green foliage and the deep color persisted throughout the experiments. Each plant produced an average of 12.5 pods. Only moderate symptoms of injury developed on retarded plants that received 6 grams of fertilizer per pot and these plants averaged 8 pods.

Two experiments were carried out in which young soybean plants were treated with Amo-1618, phosfon, or CCC at 1000-p.p.m. concentration as an aqueous drench to the soil 12 days prior to application of 5 grams of 5–10–5 fertilizer. None of the plants showed injury due to any of the three retardants. In both of these experiments all unretarded plants (80) were killed by the fertilizer application. Plants retarded with Amo-1618 did not show fertilizer injury and by the end of the experiment had yielded an average of 11.5 pods. The plants treated with phosfon also showed relatively little injury from the fertilizer. Eleven of the 80 retarded plants, however, developed moderate to severe foliar burn, but these injured plants later recovered and produced an average of 10.5 pods. All plants treated with CCC prior to fertilizer application developed slight to severe symptoms of foliar burn; six of them died but the remainder recovered slowly and produced an average of 5 pods. Amo-1618 induced the greatest visible growth response. The response was less when phosfon was used and least with CCC. Had the concentration of the retardants been adjusted to induce identical plant responses, the resistance to injury by fertilizer might then have been approximately equal for all three.

Mature seeds were harvested from the Amo-1618–, phosfon-, and CCC-retarded plants, respectively, that withstood application of 5 grams of fertilizer per pot. Twenty-five seeds selected at random from each lot were planted in soil. All of these seeds were viable.

An experiment was carried out with more mature soybean plants than were used previously. These larger plants were restricted in growth by the limited amount of soil in which each was grown. The leaf areas of the retarded plants were, therefore, approximately equal to those of the unretarded plants. Amo-1618, phosfon, and CCC again were applied as a 30-ml. drench per pot, using 1000-p.p.m. aqueous concentration of each chemical. Six grams of 5–10–5 was applied to each pot 7 days after treatment with each of the three retardants.

Even with these older plants the entire population of unretarded ones (40) was killed by fertilizer application while the retarded ones remained uninjured. Leaves of the plants retarded with phosfon before fertilizer treatment developed slight injury and those treated with CCC were moderately to severely damaged but recovered.

The ability of the retarded plants to withstand excessive amounts of fertilizer apparently did not depend entirely on leaf area because, in the experiment with relatively mature plants, the retarded ones showed markedly greater tolerance to the salts although they had about the same leaf area as the unretarded ones.

Sacks (*7*) reported that the numbers of dividing cells in the terminal buds of chrysanthemum were reduced by Amo-1618. Such a reduction in cell number might result in reduced absorption of salts. In the present experiments, the relatively mature plants studied showed no outward evidence of retardation of growth, but plants that received the retardant exhibited increased tolerance to application of excessive fertilizer.

Although increased tolerance to excessive fertilizer did not obviously depend on reduced leaf area and growth rates, the outwardly apparent responses mentioned indicate that the retarded plants did develop marked chemical and physical changes. Further evidence of chemical and physical changes that resulted from use of the retardants was supplied by the fact that mites, which are sucking insects, multiplied more rapidly on leaves of the unretarded plants than on the retarded ones.

Further research is needed to determine whether some retardants can be used to increase the resistance of certain crop plants to alkaline and saline soils.

Acknowledgment

Amo-1618 was supplied by the Rainbow Color & Chemical Corp., Northridge, Calif.; phosfon, by the Virginia-Carolina Chemical Corp., Richmond, Va.; and CCC, by the American Cyanamid Co., Stamford, Conn.

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INSECT ATTRACTANTS

New Attractants for the Mediterranean Fruit Fly

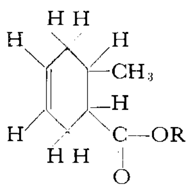
MORTON BEROZA, NATHAN GREEN, and S. I. GERTLER

Entomology Research Division, U.S. Department of Agriculture, Beltsville, Md.

L. F. STEINER and DORIS H. MIYASHITA

Entomology Research Division, U. S. Department of Agriculture, Honolulu, Hawaii

Insect attractants have proved to be invaluable tools for the control and eradication of certain insect species (*1, 2, 7, 8*). The important roles played by angelica seed oil (*12*) and *sec*-butyl 6 - methyl - 3 - cyclohexene - 1 - carboxylate (I*b*) (*2, 4, 6*),

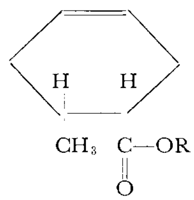


I*a*, R = H

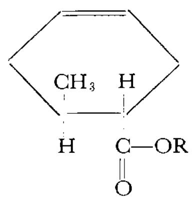
I*b*, R = $CHCH_2CH_3$ (CH_3 on the CH)

now known as siglure, in the eradication of the Mediterranean fruit fly [*Ceratitis capitata* (Wied.)] or medfly, from Florida in 1956–57 have been recorded (*1*). These lures attract only the male of the species.

An observation that certain commercial lots of siglure gave inferior performance led to the discovery that *cis-trans*-isomerism affected the attractiveness of the product (*11*).



cis-I

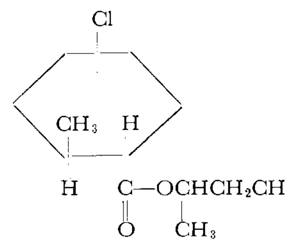


trans-I

The *trans*-product was much superior to its *cis*-analog. Later, an infrared spectrophotometric method was developed to determine the *cis-trans*-content of the ester, and a means of preparing the desired all-*trans*-product was devised (*6*).

As part of this investigation, the authors studied the conditions that led to epimerization of the *trans*-acid, mainly to avoid such conditions in the course of the synthesis. An unexpected bonus accrued when a means was found to add the theoretical amount of hydrogen chloride to the double bond of the acid moiety of siglure. Past attempts to effect this addition by conventional means were unsuccessful. With the hydrochlorinated acid available, a series of esters was prepared and sent to Hawaii for testing as medfly lures. In laboratory and field trials, some of the esters were far superior to siglure in both attractiveness and lasting quality. The excellent performance of the hydrochlorinated products led to the preparation of the hydrogen bromide addition product of (I*a*) and its esters.

Of all the esters prepared, the best were the *sec*- and *tert*-butyl esters of the hydrogen chloride adduct of (I*a*) and the ethyl ester of the hydrogen bromide adduct.



II (medlure)

sec-Butyl *trans*-4(or 5)-chloro-2-methylcyclohexane-1-carboxylate (II), now known as medlure, has already been produced commercially and is used by the U. S. Department of Agriculture,