

a sensitive instrument, such as the Coleman photo-fluorometer, Model 12, must be used.

By means of an apparatus which was developed three years ago by Mr. George S. Liebeck, of the American Telephone and Telegraph Company, and which has been in constant use since, from 0.1 to 6 microgram quantities in 5 cc of extract can be determined. This range is made possible by very high stable electronic amplification and a variable shunt which increases or decreases the sensitivity of the microammeter with which the readings are obtained. At one-half of the maximum sensitivity of the instrument, 0.1 to 1.0 micrograms in 5 cc of standard solution, at increments of 0.1 micrograms, gave the following corrected (blank, 1.6) galvanometer deflections: 2.9, 5.9, 8.2, 10.1, 13.1, 17.3, 19.2, 22.7, 25.4, 29.5. One galvanometer deflection, therefore, indicated the presence of 0.0068 microgram of nicotinic acid per cc of extract.

The procedure has given results with wheat flours, cornmeal and animal and green plant tissues which generally check closely with those obtained by the microbiological method. The results by the latter method were obtained on acid digests. In a few instances, for example, with soybean meal, the fluorimetric method has given definitely higher results than the microbiological or colorimetric methods. This effect is not due to other fluorescent substances such as riboflavin, or amine-, or CNBr-reactive materials. It appears to be due to pyridine-like substances which react with *both* CNBr and the amine. Whether or not these substances belong in the category of physiologically active pyridine compounds studied by Elvehjem and coworkers¹¹ is now being determined in this laboratory.

The procedure and its application to biological materials will be described in detail in a later publication.

Summary: In a study of the König reaction, it was found that nicotinic acid, on reacting with CNBr and certain substituted aromatic amines, yields glutamic dialdehyde derivatives which fluoresce with a greenish-yellow light in visible violet light of about 440 millimicrons wave-length. A procedure was described which is applicable to 0.1 to 6 micrograms in 5 cc of solution or extract. It is suggested that the principle of preparing fluorescing "tagged" derivatives be applied to other vitamins of the B-complex.

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¹¹ W. A. Krehl, C. A. Elvehjem and F. M. Strong, *Jour. Biol. Chem.*, 156: 13, 1944.

THE INHIBITION OF POLLEN PRODUCTION IN RAGWEED BY THE USE OF CHEMICAL SPRAYS¹

DURING the course of a pollen survey, made for the Michigan Department of Health in the summers of 1940, 1941, 1942, and 1944, it became apparent that, while data in regard to the number of ragweed pollen grains in the air at any time are valuable to physicians working in the field of pollen allergy, there is a much more important problem in those regions where ragweed grows freely. The more fundamental problem is not the accumulation of numerical data concerning pollen incidence, but rather the development of some method of control for this menace to the health and well-being of many people.

A program designed to eradicate ragweed from large areas frequently faces some opposition because of the possibility that successful eradication of these plants may tend to increase soil erosion in certain regions and because the weeds have some value as cover and food for wild-life.

The ideal solution of this pollen problem would be the development of a method by which large areas may be treated with some agent which will prevent flower formation, or at least pollen production, and not destroy the vegetative portion of the offending plants. The chief obstacles to the use of chemicals for this purpose are the cost of materials and the possible danger to cultivated crops and livestock. Observations on weed control in vegetable crops by the use of selective herbicides, however, have shown that certain chemicals are available which are not a hazard to animal life and may be useful in this connection.

In the period of August 20 to September 16, 1944, a number of test sprayings were made on areas having a heavy growth of ragweed. Ragweed on these plots began pollinating the last week of August and spray treatments were begun as soon as pollen release was evident. All sprays were applied at 100 pounds pressure and at the rate of 100 gallons per acre.

RESULTS

G-412² (di-nitro-secondary-butyl-phenol) in kerosene gave a complete kill of ragweed within a period of six hours. The vegetative portion of the plant, as well as the flower spikes, turned brown, and pollen release was stopped. Water solutions of this material killed more slowly, and frequently killing was not complete.

G-410² (penta-chlor-phenol) gave a 75 per cent. kill in twelve hours, but some stems remained alive and continued growth until frost. Pollen was not again produced by these plants.

¹ Journal Article No. 755 (n.s.) from the Michigan Agricultural Experiment Station.

² Supplied by the Dow Chemical Company.

Sinox (in kerosene) killed only the younger leaves and flower spikes. Axillary buds developed and young flower shoots were evident at the time of frost.

Kerosene alone killed the younger leaves of ragweed and any flowers that were ready to open at the time of spraying, but the effect was temporary, and the plants recovered within a short time and continued to release pollen.

DISCUSSION

These results indicate that it is possible to stop pollen production in ragweed with chemical sprays. Greenhouse tests and some field observations, however, indicate that most of the materials used in this series of tests are more or less toxic to cultivated crops at the concentrations used in these experiments. Very few crops will tolerate kerosene, and thus the method of weed control described would be of little value in areas where crops are being grown.

Recent reports of work with growth-regulating substances^{3, 4} have shown that ragweed, as well as many other annual weeds, can be killed by spraying with "2-4-D" and other similar materials. Limited greenhouse tests, conducted during the winter, have

shown that low concentrations of these substances have a very pronounced effect on ragweed. When young ragweed plants, six inches tall, were sprayed, growth of terminal buds was stopped. No further elongation occurred during the observation period of two months, nor were there any flower spikes evident. Leaves present at the time when sprayed remained green, and the older parts of the stem appeared normal.

Confirmation of these results will be sought as soon as plants are available under natural conditions. Details of concentration of "2-4-D" and time of application remain to be explored, but the information already available indicates that it will be possible to develop a program of treatment that will prevent the production of pollen by common ragweed without having the undesirable features of complete destruction of vegetation.

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DISCUSSION

THE CONTROVERSY ON CHOLINESTERASES

For about two years a controversy, in the form of articles and counter articles in *SCIENCE*, has continued between Mendel and Rudney and Alles and Hawes concerning claims of priority for the discovery of two separate enzymes capable of hydrolyzing choline esters on the one hand, and the use of the term "pseudo-cholinesterase" on the other. The former point should be resolved simply by referring to the facts, and the latter should be considered without delay since, if the term is disadvantageous, it ought to be dropped from the scientific literature as soon as possible. The writer, as a completely disinterested individual, relative to sides in the matter, has undertaken this objective discussion in the hope that it might help to clarify the controversial issues.

It would appear that the significant facts are as follows:

(1) Alles and Hawes¹ were the first to point out, by experiments on human blood, that two apparently distinct enzymes exist that are capable of hydrolyzing acetylcholine. They arrived at this conclusion, which

was reiterated by Hawes and Alles,² on the basis of differences in the enzymatic properties of the blood cells and serum as regards activity-pH, activity-sodium chloride and activity-substrate concentration relationships, as well as the differences effected by introducing methyl groups into the choline portion of the substrate esters. These investigators found that, whereas serum exhibits only a slight enzymatic hydrolysis of acetyl- β -methylcholine, the cells produce an enzymatic scission at a rate which is of the same order as that of acetylcholine.

(2) Mendel and Rudney³ observed that purified preparations derived from serum and certain tissues exhibited enzymatic hydrolysis of acetylcholine, tributyrin and methyl butyrate, while those obtained from brain tissue and the red blood cells of some mammals possessed demonstrable activity only on the acetylcholine. These workers concluded that a non-specific enzyme, for which they proposed the name "pseudo-cholinesterase," was present in the former case and a specific cholinesterase in the latter.

(3) In a later communication Mendel, Mundell and Rudney⁴ reported that acetyl- β -methylcholine was

³ C. L. Hamner and H. B. Tukey, *Bot. Gaz.*, 106: 232-245, 1944.

⁴ P. C. Marth and J. W. Mitchell, *Bot. Gaz.*, 106: 224-232, 1944.

¹ G. A. Alles and R. C. Hawes, *Jour. Biol. Chem.*, 133: 375, 1940.

² R. C. Hawes and G. A. Alles, *Jour. Lab. Clin. Med.*, 26: 845, 1941.

³ B. Mendel and H. Rudney, *Biochem. Jour.*, 37: 59, 1943.

⁴ B. Mendel and D. B. Mundell and H. Rudney, *Biochem. Jour.*, 37: 473, 1943.