

Haematoxylin crystals	4 gms.
Alcohol 95 per cent.	25 cc
Sat. sol. of ammonia alum	400 cc

This solution was placed in an open dish, at a distance of 15 cms from a Cooper-Hewitt burner, operating at 140 volts and 3.3 amperes, for one hour. The solution was then filtered and to the filtrate was added:

Methyl alcohol	100 cc
Glycerine	100 cc

This solution was placed under the Cooper-Hewitt burner at the same distance for two hours. The solution was then filtered and used for staining purposes.

No appreciable difference was noticed between the staining quality of the Haematoxylin prepared in this manner and that left for sixty days to ripen.

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SOAP AS A MOSQUITO LARVICIDE

DURING some experiments with mosquito larvicides the writer has observed that the addition of soap has brought about a much larger increase in toxicity to mosquito larvae than that which could be ascribed to improved penetration. This suggested that soap may possess direct toxicity to mosquito larvae. To verify this point tests were carried out with a liquid soap, consisting of a mixture of potassium oleate and coconut oil soap containing about 40 per cent. actual soap.

For this purpose larvae and pupae, taken from a partially polluted ditch breeding primarily *C. pipiens*, were transferred to large porcelain dishes containing about 500 cc of a mixture of tap water and ditch water. Various concentrations of soap were then mixed in. After 24 hours the number of dead and living insects were counted.

The results, given in the accompanying table, clearly show that concentrations of 0.2 per cent. soap or higher gave 100 per cent. kill of larvae and pupae.

The value of soap as a larvicide can perhaps be utilized in treating clear standing water, fire barrels, etc., where application of oil or larvicides containing toxic or inflammable chemicals are objectionable.

TOXICITY OF SOAP TO MOSQUITO LARVAE AND PUPAE

Per cent. soap concentration	Number of larvae	Per cent. dead after 24 hours	Number of pupae	Per cent. dead after 24 hours
0.05	80	20	40	5
0.10	100	65	60	55
0.20	80	100	60	100
0.50	60	100	40	100
1.00	80	100	80	100
Check	100	0	60	0

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FIXING THE PRINT OF CARBON COPIES

THE tendency of the print to become smudgy in use in bound copies of dissertations, etc., may be almost entirely eliminated by a simple treatment, that is, of melting the colored wax of the print by heat into the fibers of the paper. This may be accomplished by passing a tall Bunsen flame rapidly over the surface of the sheet. The paper should be lying flat on a smooth, good-conducting surface while flaming. After such a heat treatment the sheets are somewhat warped but may be readily flattened out in a good binder's press.

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SPECIAL ARTICLES

DEUTERIUM OXIDE AND ASPERGILLUS

RECENT investigations seem to indicate that deuterium oxide in high concentrations exerts a toxic or inhibitory effect on living organisms, both plant and animal. However, as a result of his work with *Spirogyra* sp., it was suggested by Barnes¹ that very unusual and interesting effects might be noted when the deuterium was used in less concentrated solutions.

¹ T. C. Barnes, "A Possible Physiological Effect of the Heavy Isotope of H in Water," *Jour. Am. Chem. Soc.*, 55: 4332-4333, 1933; "Further Observations on the Physiological Effect of the Heavy Hydrogen Isotope on *Spirogyra*," *Am. Jour. Bot.*, 20: 681-682, 1933.

This suggestion has received confirmation and support by Richards,² whose observations on the growth of *Saccharomyces cerevisiae* indicate that, in dilute concentrations, deuterium may have a decided effect in accelerating growth and development as opposed to its pronounced lethal effect in high concentrations. Macht and Davis,³ using a solution of one part deuterium to two thousand parts of protium, expressed

² O. W. Richards, "The Growth of Yeast in Water Containing Deuterium," *Am. Jour. Bot.*, 20: 679-680, 1933.

³ D. I. Macht and M. E. Davis, "Some Pharmacological Experiments with Deuterium," *Jour. Am. Chem. Soc.*, 56: 246, 1934.

doubt that any very remarkable effects would be forthcoming from the use of dilute solutions.

In this connection, the writer felt that it might be interesting to determine the effect of a dilute solution of deuterium oxide upon the growth of a fungus, *Aspergillus sp.* This organism has been found to be extraordinarily sensitive to differences in the culture medium (Mann⁴) and so might be expected to reflect in its growth and fruiting any changes brought about by the presence of the isotope of hydrogen.

A sample of deuterium oxide containing one part deuterium to 213 parts protium, approximately a .47 molecular per cent. solution, was obtained from the Ohio Chemical and Manufacturing Company at Cleveland. This water was distilled in order to remove any impurities present. After distillation, the relative density of the heavy water referred to ordinary water, both at 20° C., was 1.0019.

The nutrient solution used in these experiments was Pfeffer's three-salt solution, with sucrose as the source of carbon. Two groups of nutrient solutions were prepared, one with double-distilled H¹H¹O; the other with distilled H²H²O. The nutrient medium was placed in 150 cc Erlenmeyer flasks, 50 cc to the flask. The solutions were sterilized by streaming steam for a period of twenty minutes on each of three successive days. Inoculations were made from a pure, bacteria-free culture of *Aspergillus sp.* on bread by means of a platinum loop. The fungus was grown in an incubator for five days at a temperature of 37° C. The mycelial felts were then removed from the flasks, placed on weighed filter paper and dried for three days at 65° C. The filter paper used had been previously dried at the same temperature and placed in a desiccator. At the end of the three-day period, the felts were removed from the drying oven, placed in a desiccator and then weighed. From the total weight of the felt and the filter paper was subtracted the weight of the filter paper alone. Thus it was possible to determine and compare the weights of the felts grown in the deuterium oxide medium with those grown in that of protium oxide.

The fungus grown in the H¹H¹O Pfeffer's medium was in the form of a flat and evenly fruited felt. The average weight of the felts from four flasks of this solution was .0481 grams. The felts grown in the heavy water medium exhibited every indication of stimulation. They were markedly convoluted and cratered below, resembling a brain-coral; the fruiting was greatly diminished, and the distribution of spores on the surface of the felts was irregular and occurred in localized regions. When the dry weights of these heavy water felts were taken, it was found that the

average weight from the four flasks was .7719 (see Table I), or approximately sixteen times that of the felts grown in the ordinary distilled water medium.

TABLE I*

Series	Weight of filter paper	Weight of filter paper and felt	Weight of felt
1 a	1.3544 gr.	1.3963 gr.	.0419 gr.
1 b	1.3490 gr.	1.3994 gr.	.0504 gr.
1 c	1.3511 gr.	1.3983 gr.	.0472 gr.
1 d	1.3593 gr.	1.4123 gr.	.0530 gr.
Average weight of felt = .0481 grams.			
2 a	1.3592 gr.	2.1400 gr.	.7808 gr.
2 b	1.3722 gr.	2.1940 gr.	.8218 gr.
2 c	1.3681 gr.	2.0724 gr.	.7043 gr.
2 d	1.3442 gr.	2.1250 gr.	.7808 gr.
Average weight of felt = .7719 grams.			

*Series 1—Pfeffer's Three-Salt Solution + Distilled H¹H¹O.
Series 2—Pfeffer's Three-Salt Solution + Distilled H²H²O.

The writer, therefore, is of the same opinion as Barnes and Richards, that deuterium, when used in dilute concentrations, may have a decided effect in stimulating vegetative growth and development. Further experiments in this connection are planned and a more detailed consideration of the methods used and the results obtained will be published later.

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THE EFFECT OF FERRIC CHLORIDE INJECTIONS IN EXPERIMENTAL TUBERCULOSIS¹

SEVERAL years ago the writer showed that repeated intravenous injections of a 0.25 per cent. ferric chloride solution were followed by an accumulation of iron in caseous areas of the lungs of tuberculous rabbits.^{2,3} Concomitantly with this accumulation of iron, the life span of infected animals was found considerably increased over that of control tuberculous rabbits that had received no injections of the ferric salt solution. This was found to be the case in two entirely independent series of experiments comprising 36 rabbits.^{4,5} Furthermore by comparing tuberculous lesions of both experimental and control animals sacrificed at various intervals of time, it was shown that following the intravenous injections of ferric

¹ From the Department of Pathology, Harvard Medical School, Boston, Mass. Aided by a grant from the DeLamar Mobile Research Fund.

² V. Menkin, *Proc. Soc. Exp. Biol. and Med.*, 27: 1020, 1930.

³ V. Menkin and M. F. Menkin, *Jour. Exp. Med.*, 53: 919, 1931.

⁴ V. Menkin, *Jour. Exp. Med.*, 55: 101, 1932.

⁵ V. Menkin, *Am. Jour. Med. Sci.*, 185: 40, 1933.

⁴ M. L. Mann, "Calcium and Magnesium Requirements of *Aspergillus niger*," *Bull. Torrey Club*, 59: 443-490, 1932.