

SUMMARY

Multiple precipitin formation may be induced in the rabbit by the successive introduction of different antigens, and many precipitins may exist simultaneously in the blood for some time. Precipitins no longer demonstrable in the blood of a rabbit subjected to multiple successive immunization may reappear on the injection of only one of the antigens previously injected. Whether an unused antigen would have the same effect has not been determined.

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CATALYTIC REDUCTION AND DEACETYLATION OF THE METHYL ESTER OF HEX-ACETYL "β"-METHYLBALDOBIONIDE TO 6-GLUCOSIDO-β-METHYL-GALACTOSIDE

The methyl ester of "β"-methylaldobionide, from

gum arabic, was acetylated and the new crystalline hexa-acetate

(having m.p. = 140°, $[\alpha]_D^{25} = -54.3^\circ$ (in acetone) and the following composition:
Found: C 49.24, H 5.8, OCH₃ 10.01, COCH₃ 40.96
C₂₆H₃₆O₁₈ requires " 49.04, " 5.7, " 9.75, " 40.57)

has been reduced and deacetylated in methyl alcohol solution in an atmosphere of hydrogen, in the presence of copper chromite catalyst, at a temperature of 175° and pressure of 3,600 pounds per sq. in. during 5 hours. The product obtained was quite free from uronic acid (naphthoresorcinol test) and had the following composition:

Found: C 44.25, H 6.9, OCH₃ 9.84
C₁₈H₂₄O₁₁ requires " 43.80, " 6.8, " 8.71

It had $[\alpha]_D^{25} = -61.6^\circ$ (in water), thus indicating that the product is probably the β-methyl-glycoside of 6-glucosido-galactose.

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SCIENTIFIC APPARATUS AND LABORATORY METHODS

ON SECURING LARGE QUANTITIES OF DIATOMS FROM THE SEA FOR CHEMICAL ANALYSIS¹

A KNOWLEDGE of the chemical composition of marine diatoms is desirable because of the essential role these organisms play each year in the constructive part of the food cycle of the sea and because of the importance diatom substance may have had in the formation of petroleum deposits. Although countless billions of diatoms may exist in small areas of the ocean, their minute size makes it extremely difficult to obtain any considerable quantity. Chief among the difficulties encountered are the very large volumes of water which must be strained and the rapid clogging of the filtering surfaces by the diatom cells. During the past summer at the Woods Hole Oceanographic Institution methods of circumventing these difficulties were explored.

The use of a large silk net towed from a boat was found more effective than the pumping method,² provided that the proper size mesh was selected for the type of diatoms desired. Another method, consisting of culturing a pure strain of diatoms on a large scale, is on trial by Mr. Bostwick Ketchum at the Harvard

¹Contribution No. 164, Woods Hole Oceanographic Institution.

²However, pumps or sea-cocks have been found useful for collection of plankton in smaller quantities. Cf. L. D. Phifer, SCIENCE, 79: 298, 1934.

Biological Laboratories. However, exceedingly few species have been cultured successfully by any one. The advantages of procuring the diatoms directly from the sea are that any types occurring in great abundance (and hence important in the economy of the sea) can be obtained, that large quantities are procurable within a short time (at least six nets could be used simultaneously from a boat), and, since no danger exists of changes due to artificial conditions, the chemical constitution of the organisms secured will be as in nature.

To permit any useful interpretation of the chemical analysis of the material obtained, a certain degree of purity of the catch is required. If particles of detritus or protozoans of the same size as the diatoms are abundant in the water, it is obviously impossible to obtain a large quantity of uncontaminated diatom material by simple straining. Another time or place must be sought. In cases in which the detritus is very finely divided, it can be avoided by the use of the coarsest net which will retain the diatoms. However, copepods or other crustacea are almost sure to be encountered, and, because of their larger size, even small numbers of these animals would be a serious contamination quantitatively. Success in excluding organisms larger than diatoms was attained through the use of a cone of coarser silk placed over the mouth of the net and towed apex foremost. The mesh of this

cone was of such a size (No. 0 silk = 38 strands per inch) that the diatoms could pass freely through it, but copepods, etc., were stopped and swept off to one side.

Using one net (1 m diameter, 4 m long, No. 10 silk = 109 strands per inch) provided with a cone of this type, large quantities of diatoms were obtained on two occasions during September in Vineyard Sound. The net was towed at very slow speed, and at intervals of three minutes was hauled in and emptied. Each catch, which amounted to about six quarts of a thick suspension, was allowed to stand for two half-hour periods. At the end of each period the supernatant liquid, in which the diatoms remained suspended, was poured into a strainer, leaving in the bottom of each jar the heavier sand, detritus, and whatever zooplankton had accidentally entered. In the strainer (a bag of No. 20 silk) the material drained in the course of about half an hour to a sludge. For the first day's work, this sludge was spread out on towels to dry, and later transferred to a chemical hood heated to 40° C. On the second occasion the sludge was not dried but was shaken immediately with an equal volume of ether. As a result of a day's work we obtained on the first occasion about 200 grams of dry, flaky material, and on the second occasion about two quarts of sludge in ether.

A complete analysis of the plankton in a sample of sea water taken during the first operation, kindly carried out for me by Miss Lois Lillick, showed that the concentration of diatoms in this area was almost 200,000 cells per liter. This approaches the maximum richness observed for diatom flowerings. One species of diatom (*Rhizosolenia alata*) composed 85 per cent. of the count, another species (*Corethron hystrix*) formed 8 per cent., and seventeen other forms together amounted to less than 7 per cent. Examination of a sample of the sludge indicated that the detritus and the animal contaminants were as low as ever found in phytoplankton hauls, in this case being certainly less than 1 per cent. of the total volume. All the material has been turned over to Professor Hans T. Clarke, of the Department of Biological Chemistry at Columbia University, for chemical analysis.

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A COLOR TEST FOR THIAMIN (VITAMIN B₁)

A FEW milligrams of thiamin (crystalline, synthetic, Merck) and about five milligrams of p-dimethylaminobenzaldehyde are placed in a small crucible. 0.1 cc of glacial acetic acid is added and the mixture heated until all the acid is evaporated. After cooling, one drop of glacial acetic acid is added. An intense brick red color develops immediately.

The red compound is probably a Schiff's base, as most primary amines readily form colored condensation products with aldehydes.¹ Proteins and amino acids interfere.

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A METHOD FOR OBTAINING NEWLY HATCHED TADPOLES IN A CLEAN STATE

IN removing newly hatched tadpoles from their egg mass, especially when this has been procured outside the laboratory, some difficulty in obtaining them perfectly free from their "jelly" is often encountered. To overcome this the following procedure is suggested.

Place the material—the egg mass, if the larvae haven't hatched as yet, or the mixture of animals and debris—in as small a dish as convenient. Set this in a second container of sufficient size to enable water in it to more than cover the first vessel. Slowly fill the larger with clean water, being careful not to disturb the material in the smaller dish as the water overflows into it. Enough water to cover the dish by about three eighths of an inch should be added, and allowed to stand undisturbed. As soon as they are able, the tadpoles will rise and leave the smaller dish, entering the clean water surrounding it, from which they may be easily removed by means of a pipette.

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¹ O. Frehden and L. Goldschmidt, *Mikrochim. Acta*, 1: 338, 1937.

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