with the enzyme were partially or totally refractory to the pressor action of renin.

The material (liver extract) was then injected intravenously in daily doses into five dogs made hypertensive by the Goldblatt[6] technique. The blood pressure of all fell to normal levels within three to five days. The blood pressure of five normal dogs was similarly affected, but less. Associated with the fall was a reduction in the amount of urea nitrogen in the blood. A return to previous levels occurred a few days after the injections were stopped; when it was resumed the changes were repeated. In two hypertensive dogs the blood pressure and urea nitrogen both fell, but giving, subsequently, larger doses resulted in a further fall in blood pressure, a rapid rise in urea nitrogen, and death from uremia. Subcutaneous injections of this substance were followed by similar changes, but abscesses occurred. It was for that reason impossible to estimate the nature of the effect on hypertension. Larger quantities of the material given by mouth had little or no effect in three dogs and one monkey. The changes observed did not result from the action of non-specific proteins; aliquot amounts of horse serum and ten preparations of inactivated enzyme, given daily, did not affect blood pressure.

The preparation appeared to be somewhat toxic to anesthetized rats, but did not adversely affect unanesthetized dogs. The material was insoluble and the enzyme unstable. It did not oxidize phenols. As many impurities were present it can not be concluded that the results were due to the action of amine oxidase, but this interpretation is possible. If so, amines are concerned in this type of hypertension.

Note: The author is indebted to Merck and Co., for supplying the preparation of amine oxidase.

HENRY A. SCHROEDER
HOSPITAL OF THE ROCKEFELLER INSTITUTE FOR MEDICAL RESEARCH, NEW YORK

SCIENTIFIC APPARATUS AND LABORATORY METHODS

THE USE OF BROMOFORM IN THE SEPARATION OF NONCALCAREOUS MICROFOSSILS

POLLEN grains, plant spores, diatoms, sponge spicules and other microfossils are much more commonly present in unconsolidated sediments of Cenozoic age than has ordinarily been assumed. If properly identified, the microfauna and microflora of such deposits may prove to have great value in correlation and in studies dealing with the climatic and other conditions at the time and place of deposition. The separation of foraminifera and other calcareous microfossils from sediments by means of heavy liquids such as bromoform has become standard practice in many petrographical studies.[1] The value of such methods for the separation of the non-calcareous microfossils has not, however, been generally recognized.

During the last few years the writer[2] has been using a bromoform-acetone solution of a standard specific gravity to separate such fossils from various types of unconsolidated sediments ranging in age from Cretaceous to Recent. The technique that has been developed has led to the discovery of fossils in sediments that have formerly been considered to be completely barren. The method of separation and concentration is simple, rapid and complete. The microfossils of various types are concentrated together, thus making possible quantitative studies of the different elements of the fauna and flora. The method is applicable to most types of microfossils found in sediments, including foraminifera, ostracods (with the two valves in place), radiolaria, silicoflagellates, siliceous sponge spicules, diatoms, pollen and spores of plants, and the light remains of plants and animals, which are often of diagnostic value. The method is especially valuable when dealing with sediments containing relatively few individuals.

The skeletal remains of most diatoms, radiolaria and silicisponges are composed of colloidal silica in the form of opal with a specific gravity of 1.9–2.3. Since quartz and the common clay minerals of which most sediments are composed have a somewhat higher density (2.5–2.7), it is possible to separate these siliceous fossils from unconsolidated sediments by means of a heavy liquid with a specific gravity of 2.3. Although the writer has found no previous reference to the use of heavy liquids in separating sponge spicules, radiolaria and silicoflagellates from sediments, he has found that the method has been occasionally used for the concentration of diatoms. Such a method is described by F. Hustedt.[3] In this case Thoulet solution with a specific gravity of 2.3 was employed. This solution, however, is extremely poisonous and because of its corrosive properties and the difficulties involved in its use, it is not as suitable as bromoform.

Sediments also frequently contain small numbers of

[6] H. Goldblatt, Harvey Lectures, 33: 237, 1937–1938.

[1] Marcus A. Hanna, *Econ. Geol.*, 22: 1, 14–17, 1927.

[2] Acknowledgment is gratefully expressed for financial aid received from Mr. Robert W. Sayles and the associates in science of Harvard University.

[3] F. Hustedt, "Die Kieselalgen Deutschlands, Österreichs und der Schweiz" in Rabenhorst, L., *Kryptgamen-Flora von Deutschland, Österreich und der Schweiz.* Vol. vii, pp. 189–190, 1927.

pollen grains and spores. These are customarily concentrated by using hydrofluoric acid, which dissolves out the silica and leaves the plant residue in a great enough concentration to facilitate pollen counts.[4] This concentration can be accomplished more efficiently and with less danger by using bromoform and acetone with a gravity of 2.3. Although pollen grains may be separated by means of a liquid of a lower density, experience has shown that with a high gravity solution there is less chance of loss, because there is less rapid settling of the sediment. Recently V. P. Grichuk[5] described a similar method for recovering pollen from loess deposits. The liquid used was "Toulé" (Thoulet?) solution with a specific gravity of 2.2.

The following is a description of the method that has been employed by the writer in examining various types of sand, silt, clay and till for microfossils. The method can be easily modified and can be used in conjunction with other methods commonly employed, when a greater concentration or a thorough cleaning of the microfossils to be studied is desired.

The unconsolidated sediment must first be completely broken up into its constituent parts and thoroughly deflocculated so that the fossils may be as free as possible from adhering material. With the coarser sediments this can usually be done by shaking the dried sediments in the bromoform to be used for the final separation of the microfossils, until the particles are entirely separated. With finer sediments, such as silt and clay, other methods are usually necessary. Ordinarily these sediments can be easily broken up by soaking small pieces of the material in water or in acetone followed by repeated agitation. If the sediments can not be broken up in this way, it becomes necessary to use alkalies or acids, although such means should not be used unless absolutely necessary because of the danger of destruction of some of the microfossils.

After the material is broken up and washed in acetone, it is thoroughly dried and then placed in a mixture of bromoform and acetone with a specific gravity of 2.3. After thorough mixing with the heavy liquid it is centrifuged. The light portion containing the fossils, which is found floating on the surface of the liquid, is poured off, filtered, washed thoroughly in acetone and dried over a hot plate. In most cases it is desirable to centrifuge the sediments several times in order to get a complete separation, especially when quantitative studies are being made. For preliminary work, however, it is generally necessary to centrifuge them only once. The dried material is then examined for fossils, and if present they are mounted directly or further concentrated and cleaned by methods commonly employed in the study of the different types of microfossils.

The bromoform may be recovered from the acetone washings and used again. This is done by mixing the washings in water and separating the bromoform from the water and acetone by means of a separatory funnel. It should be pointed out that bromoform is somewhat poisonous, but if it is used in a well-ventilated room or under a ventilating hood, there is no danger of unfortunate effects.

The method described above should be of considerable value to those engaged in ecological studies of modern lake sediments and deep-sea deposits as well as in many geological investigations involving the use of microfossils.

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GEOLOGICAL MUSEUM,
HARVARD UNIVERSITY

[4] G. Assarsson and E. Granlund, *Geol. Fören. in Stockholm Förhandl.* Bd. 46 H 1–2, pp. 76–82, 1924.

[5] V. P. Grichuk (Gritchouk), *Problems Phys. Geog. Acad. Sci.*, U.S.S.R., Vol. viii, pp. 53–58 (Russian; French summary), 1940.

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