

formly dark blue color. I have come into possession of a set of serial sections of this species found among the effects of Dr. Walton,⁹ and these yield some additional information but unfortunately the specimen is sexually immature.

I have now to report the finding in woods of the Appalachian Mountains of a new endemic rhynchodemid. To date three specimens have come to hand,¹⁰ collected in Maryland, Virginia, and West Virginia, respectively. All three are at or near sexual maturity, and study of the copulatory apparatus in serial section has shown that the animal belongs to the genus *Diporodemus* Hyman, 1938.¹¹ This genus differs from *Rhynchodemus* in that in addition to the usual common genital pore opening in the midventral line there is a female or vaginal pore to one side of the median line by which the copulatory bursa opens to the exterior. I established this genus for a species from Yucatan, Mexico. Since then a second species of the genus has been found on Barro Colorado Island, Canal Zone,¹² and now the Appalachian form is the third member. All three species are practically identical in external appearance. They are black or brownish black, plump, cylindroid and about 15 mm in length; the Panamanian and Appalachian species are further alike in that the eyes are degenerate, detectable only in sections, and the creeping sole continues onto the ventral surface of the head as a glandular cleft, probably acting as an adhesive organ in food capture. The Yucatan and Appalachian species have also in common a pair of longitudinal sensory tracts on the cephalic ventral surface.

A complete account of these endemic and introduced land planarians will be published elsewhere.

L. H. HYMAN

AMERICAN MUSEUM OF NATURAL HISTORY,
NEW YORK CITY

CONCERNING PURE CULTURES OF SPIRILLUM

ALTHOUGH bacteriologists have long been interested in spiral bacteria, very few workers have reported success in the isolation of pure cultures. Esmarch,¹ Beijerinck,² Kutscher,³ Bonhoff,⁴ Dimitroff⁵ and

⁹ Dr. H. I. Strohecker, of Kenyon College, Gambier, Ohio, kindly forwarded to me a box of slides of land planarians made by Dr. Walton.

¹⁰ Sent for identification by the U. S. National Museum and collected by Dr. J. P. E. Morrison of that institution.

¹¹ "Fauna of the Caves of Yucatan," Carnegie Inst. Wash., Publ. No. 491.

¹² A collection of land planarians from Barro Colorado Island was kindly sent to me for identification by E. C. Williams, Jr., of Northwestern University; description in press.

¹ E. von Esmarch, *Centr. f. Bakt.*, 1: 225, 1887.

² M. W. Beijerinck, *Centr. f. Bakt.*, 14: 827, 1893.

³ Kutscher, *Zeitschr. f. Hygiene*, 20: 46, 1895.

⁴ Bonhoff, *Archiv f. Hygiene*, 26: 162, 1896.

⁵ V. T. Dimitroff, *Jour. Bact.*, 12: 19, 1926.

Giesberger⁶ have described methods by which they were able to isolate several species from water and feces.

Several months ago, the writer wished to procure some species of this genus for cytological study, but after considerable correspondence, found that such cultures are not available. It now seems probable that all or most of the pure cultures isolated by earlier investigators have been lost. *Rhodospirillum rubrum* (Esmarch) Molich and *Spirillum virginianum* Dimitroff were obtained from the American Type Culture Collection. The cells of these species are rather small, however, and not well suited for cytological study. Since pure cultures could not be obtained from culture collections, isolation of the desired species from original sources was accomplished.

This note is written to correct a general impression that the isolation of pure cultures of *Spirillum* is a difficult matter. My experience has been that several species are readily isolated from raw cultures by simple routine methods employed for other groups of bacteria. Suitable raw cultures must be obtained by enrichment methods, but this is not difficult since most surface waters appear to contain a variety of species of this genus.

Several culture media were tested for enrichment, but none was found more satisfactory than hay or other plant infusions. Decaying cultures of fresh water algae usually contain many spiral bacteria. Spiral cells are rarely seen in the early stages of decomposition, but as the culture ages they become more and more abundant until, at times, the surface pellicle becomes a swarming mass in which they predominate. This peak period may be reached within a few days but generally requires from one to three weeks. Raw cultures are generally rather short lived.

During the swarming period, pure cultures are readily isolated by streaking out on the surface of beef extract peptone agar. The colonies appear promptly and are readily recognized by microscopic examination *in situ*. By this method the following species were isolated from creek and pond waters in the vicinity of Austin, Texas: *Sp. serpens* (Müller) Winter, *Sp. undula* (Müller) Ehrenberg, *Sp. tenue* (Müller) Ehrenberg, *Sp. itersonii* Giesberger. The most common species in this vicinity proved to be *Sp. serpens*.

Although rich raw cultures of *Sp. volutans* Ehb. became available on two occasions, it was not possible to obtain pure cultures since no colonies developed on the streaked plates or in poured plates with semi-solid agar as the plating medium. The giant spiral cells of this species could be seen lying on the agar surface,

⁶ G. Giesberger, Delft thesis, W. D. Meinema, pp. 1-136, 1936.

but there were never any indications of growth or cell division.

There has been but a single claim for isolation of this species. Kutscher identified one of his pure cultures as *Sp. volutans* Ehb., but his identification was questioned by Migula.⁷ According to Migula, the organism isolated by Kutscher should be designated as *Sp. giganteum* (Kutscher) Migula. This has caused considerable confusion since many workers employed Kutscher's organism in their studies on the cytology and physiology of bacteria, and referred to the organism as *Sp. volutans*. The illustrations by these workers bear little or no resemblance to the true *Sp. volutans* Ehb. There seems, therefore, no reason to believe that this species has yet been isolated.

A cytological study of the several species has been completed and will be published elsewhere. The pure cultures have been deposited in the American Type Culture Collection.

I. M. LEWIS

THE UNIVERSITY OF TEXAS

OXIDATION OF SULFANILIC AND ARSANILIC COMPOUNDS BY NASCENT HYDROGEN PEROXIDE

OTTENBERG and Fox¹ have reported that colored products appeared following ultra-violet irradiation of dilute solutions of sulfanilamide. The blue-violet products seemed of particular interest. Fox, Cline and Ottenberg² as well as Rimington and Hemmings³ emphasized that the formation of the blue derivative involves an oxidation of sulfanilamide, since the presence of oxygen is necessary for the formation of the blue-colored irradiation product.

Oxidation of sulfanilamide by chemical means to a blue product had not been accomplished, when Fox, Cline and Ottenberg published their article.² In the meantime I⁴ have reported that, under certain experimental conditions, solutions of sulfanilamide treated with oxygen form hydrogen peroxide and a blue-violet

compound which is reversibly reducible and oxidizable. The hypothesis was advanced that the formation of this blue-colored substance may be due to the influence of nascent H₂O₂. In further experiments being published in detail elsewhere it was found that nascent hydrogen peroxide as formed on autoxidation of hydrazine solutions (Gilbert,⁵ Schales⁶), in presence of cupric ions oxidizes sulfanilamide promptly to blue-violet derivatives. These substances are reversibly reducible and oxidizable, and, when freshly formed, extractable with amyl and butyl alcohols and other organic solvents. They are comparatively stable in those solvents but unstable in water, losing extractability and changing color.

Among related compounds studied arsanilic acid behaves in a manner comparable with sulfanilamide. The blue-violet butyl alcoholic extracts obtained from sulfanilamide and arsanilate showed an absorption spectrum practically identical in shape (maximum absorption at about 590 m μ). The identity of the two blue compounds is probable. It would be expected that if these compounds had retained their characteristic side-chains there would be a greater difference in the absorption spectra. If the oxidation products are identical the side-chains must have been lost. It is suggested that therapeutically or toxically active derivatives formed *in vivo* also may lack the characteristic side-chains of the original substance.

Rosenthal and Bauer⁷ recently published in this journal the extremely interesting observations that on oxidation of sulfanilamide by means of ultra-violet irradiation or ferric chloride and hydrogen peroxide the sulfonamide group is split off. My studies of the spectroscopical behavior of the blue oxidation products obtained from sulfanilamide and from atoxyl lead to a similar conclusion.

GEORG BARKAN

EVANS MEMORIAL, MASSACHUSETTS MEMORIAL
HOSPITALS AND BOSTON UNIVERSITY
SCHOOL OF MEDICINE

SCIENTIFIC BOOKS

THE CALCULUS

Introduction to the Calculus. By ARNOLD DRESDEN.
xii + 428 pp. New York: Henry Holt and Company. 1940. \$3.40.

THIS book, as its title indicates, is designed as an

⁷ W. Migula, "System der Bakterien." Jena, 1900.

¹ R. Ottenberg and Ch. L. Fox, Jr., *Proc. Soc. Exp. Biol. and Med.*, 38: 479-481, 1938.

² Ch. L. Fox, Jr., J. E. Cline and R. Ottenberg, *Jour. Pharmacol. and Exp. Therap.*, 66: 99-106, 1939.

³ Cl. Rimington and A. W. Hemmings, *Biochem. Jour.*, 33: 960-977, 1939.

introduction to the calculus, presumably for students of sophomore age. It is particularly noteworthy, inasmuch as it is the first serious attempt among American text-books to introduce the subject in a rigorous and logical manner. The first two chapters, about sixty pages, of the book, are devoted to the essentials of the

⁴ G. Barkan, *Proc. Soc. Exp. Biol. and Med.*, 41: 535-537, 1939.

⁵ E. C. Gilbert, *Jour. Am. Chem. Soc.*, 51: 2744-2751, 1929.

⁶ O. Schales, *Ber. Dtsch. Chem. Ges.*, 71: 447-460, 1938.

⁷ S. M. Rosenthal and H. Bauer, *SCIENCE*, 91: 2369, 509, May 24, 1940.