

tion of a partially synthetic medium and Vischniac (J. Gen. Microbiol., **12**, 455, 1955) developed a completely synthetic substrate which supported the growth of *Labyrinthula*. Whereas these latter media are invaluable for studying the nutritional requirements of this genus, optimal growth of the slime mold is obtained on serum-sea water agar.

The serum-sea water medium supplemented with antibiotics has proved equally effective for the isolation and cultivation of a holocarpic, marine phycomycete belonging to the genus *Thraustochytrium*, described by Sparrow (Biol. Bull., **70**, 236, 1936), from mixed cultures. This diminutive member of the Saprolegniales appeared consistently on plates which were inoculated with leaves of *Zostera marina* collected

from both the Pacific and the Atlantic shores of the United States in 1951 and 1952. After implantation on this medium a few drops of sterile sea water were added to the border of the eelgrass. Motile zoospores developed in this liquid environment, and the isolated colonies of *Thraustochytrium* which arose from them were transferred to fresh media. Pure cultures were obtained after two or three transfers.

Application of these techniques readily permits the isolation and cultivation of these singular organisms, which heretofore have been available for study only in crude enrichment cultures. The characteristics of two or more species of *Labyrinthula* are now being investigated by one of us (S. W. W.) and will be reported subsequently.

RAPID METHOD FOR DETERMINING THE ACTIVITY OF MICROORGANISMS ON NUCLEIC ACIDS

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The ability of microorganisms to hydrolyze nucleic acids has been observed by a number of investigators. Some have been concerned with the effect on the phosphorous content of the soil (Koch and Oelsner, Biochem. Z., **134**, 76, 1923), although the majority have been investigating the bacteriolytic systems of the soil actinomycetes (Muggleton and Webb, Biochim. et Biophys. Acta, **9**, 343, 1952; Born, J. Gen. Microbiol., **6**, 344, 1952) and *Streptomyces albus* (Salton, J. Gen. Microbiol., **12**, 25, 1955; McCarty, J. Exptl. Med., **96**, 555, 1952). Still others have been concerned with the ability of the organism to depolymerize nucleic acids, both ribonucleic acid (RNA) and deoxyribonucleic acid (DNA) (McCarty, J. Exptl. Med., **88**, 181, 1948; Brown, J. Bacteriol., **60**, 673, 1950; MacFayden, J. Biol. Chem., **107**, 297, 1934; Woodward, J. Biol. Chem., **156**, 143, 1944).

The procedure described here is simple and gives positive reactions in certain cases in which other techniques are unsatisfactory (*Serratia marcescens*).

A semi-synthetic medium modified from that

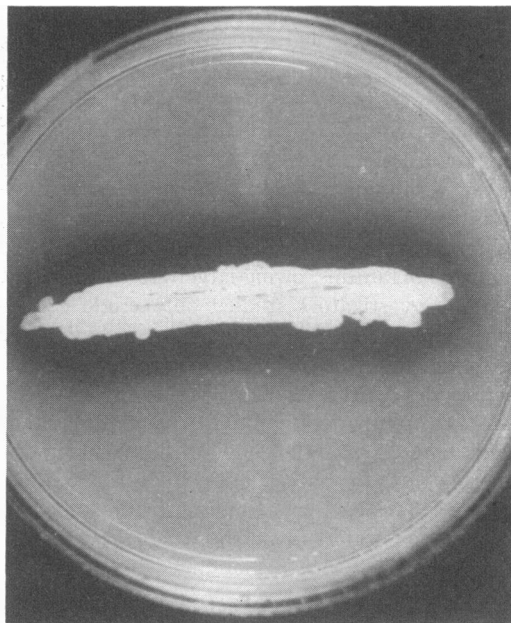


Figure 1. Activity of *Streptomyces albus* on nucleic acid medium.

TABLE 1

The effect of varying pH and incubation temperatures upon the ability of bacteria to break down nucleic acid as determined by the agar plate method*

	Zone Size in Cm from Colony Edge								
	37 C			30 C			25 C		
	pH								
	6.0	7.0	8.0	6.0	7.0	8.0	6.0	7.0	8.0
<i>Bacillus megaterium</i>	0.2	0.2	0.2	0.3	0.5	0.4	0.4	0.9	0.8
<i>Streptomyces rimosus</i>	0.6	1.4	1.2	0.7	1.3	1.2	0.1	0.6	0.4
<i>Streptomyces albus</i>	0.5	1.0	0.9	0.5	0.9	0.5	±	0.3	±
<i>Serratia marcescens</i>	—	0.8	0.6	±	0.9	0.9	±	0.8	±
<i>Serratia rubidaea</i>	—	0.6	0.2	—	0.7	0.2	—	0.5	0.2
<i>Proteus vulgaris</i>	—	±	±	±	0.1	0.1	—	±	—
<i>Bacterium zenkerii</i>	—	±	±	±	0.1	0.2	±	0.1	0.2

* These data refer to a ribonucleic acid substrate only.

described by McCarty (J. Exptl. Med., **96**, 555, 1952) gives satisfactory results with *Streptomyces* sp. and other non-exacting organisms (glucose, 5 g; casamino acids vitamin free (Difco), 5 g; K₂HPO₄, 5 g; NaCl, 2 g; FeSO₄, 0.05 g; MgSO₄, 0.5 g; agar, 15 g). The complex media available commercially have also given satisfactory results.

The solutions of nucleic acid are prepared to give a concentration of 2 mg per ml of medium. The RNA is dissolved by careful addition of 1 N NaOH, the pH not being allowed to exceed 5.0. The DNA readily dissolves in distilled water. The nucleic acid solutions are added to the liquefied agar just prior to autoclaving and the plates are poured as soon as the medium has cooled to 50 C.

Inoculation of agar plates can be made in several ways, but the method used throughout this examination was band inoculation. The inoculated media may be incubated at several temperatures in order to determine the maximal enzyme production, as this is sometimes at a temperature other than that providing optimal

growth (*Bacillus megaterium* is a case in point). The incubation time has been 36 hr.

The enzymatic activity is then assayed by flooding the plate with 1 N HCl. The acid reacts with the nucleate salts in the medium, yielding free nucleic acid, and consequently a cloudy precipitate. A positive reaction is indicated by a clear zone around the colony (figure 1). The width of this clear zone is related to the amount of the exocellular enzyme produced.

Some typical results obtained by this method are presented in table 1. These results suggest that the organisms are more exacting as to pH for production of the enzyme than they are to the temperature.

Among the genera which have been found to give positive results are *Bacillus*, *Streptomyces*, *Serratia*, *Penicillium*, *Aspergillus*, *Streptococcus* and *Achromobacter*. Of the few species of the genera *Flavobacterium*, *Alkaligenes*, and *Micrococcus* which have been surveyed none has exhibited any action.

AN OVAL TUBE ADAPTER FOR THE QUEBEC COLONY COUNTER

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In the course of some of our bacteriological studies, Fisher agar slant tubes (modified form)

have proved to be of greater value than petri dishes for counting bacterial colonies. Applica-