

of the preparation, a field showing abundant spores. Suppose there are five kinds of spores in the mount; assign a letter to each kind. Move the slide from left to right so that the spores appear to be slowly travelling along between the two lines. Call off to an assistant the proper letter for each spore passing by the vertical line within the parallel lines. Let the assistant record the letters on plotting paper conveniently divided off into 50 or 100 squares. This method will enable a record to be made almost as fast as one can talk. Of course, simply counting the letters will give the percentage of each type of spore. This procedure will obviously eliminate some of the experimental error common in "field counts," and is much faster since there is no pause to answer the question, "Have I counted that one before?" The writer is using the method in studying a species of *Fusarium*.

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#### CULTURE MEDIUM FOR THE CILIATE LACRYMARIA

LACRYMARIA has been observed by numerous biologists who noted its form and structures, "its phenomenal power of elongation, its wonderful elasticity and its great freedom of movement,"<sup>1</sup> but no one has as yet made a thorough study of this remarkable organism. This is no doubt due to its scarcity.

Mast, who has had wide experience in collecting protozoa, says ('11, p. 230): "Lacrymaria is relatively scarce in nature. It is occasionally found in cultures containing decaying aquatic plants but never in great numbers. One rarely finds more than two or three specimens in a drop of solution." And no one has heretofore succeeded in cultivating it in the laboratory. Professor Mast called my attention to this and suggested the following experiments:

Various concentrations of (1) timothy hay, (2) wheat, (2) beef extract and (4) malted milk in distilled water were prepared in two sets, one of which was boiled and the other not. All were seeded with *Lacrymaria* and examined from time to time for several weeks.

The *Lacrymaria* died out, without any apparent increase in numbers, in all the cultures except those containing malted milk, 1-5 mgr to 100 cc water. The best growth was obtained in cultures containing 3 mgr malted milk to 100 cc water. In some of these the *Lacrymaria* became very abundant and continued

to thrive for more than six weeks without adding anything to the cultures.

These cultures contained *Halteria* and another similar organism which was not identified and numerous bacteria. The *Lacrymaria* were observed to capture *Halteria*, but they appeared to feed mostly on the other organisms.

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#### SPECIAL ARTICLES

##### A PRELIMINARY REPORT ON THE CULTIVATION OF THE MICROBE OF OROYA FEVER

OROYA fever, or Carrion's disease, is a highly fatal infection endemic in certain regions of Peru. Its most striking clinical feature is a rapidly progressing severe anemia, associated with febrile reactions. In the red blood cells of patients suffering from the disease Barton found, in 1905, peculiar bacilliform elements, the specificity of which has been confirmed by Barton's subsequent observations and those of later investigators. Strong, Tyzzer, Sellards, Brues and Gastiarru concluded that these bodies are of protozoan nature and proposed for them the name of *Bartonella bacilliformis*. Their cultivation, however, had not been achieved, and the problem offered opportunity for the trial of procedures recently developed for the cultivation of certain spirochetes, flagellates and rickettsia-like microorganisms.

In the summer of 1925 one of us (B.) went to Lima and secured the material for study. Through the generous permission of Dr. Olacchia, of the Dos de Mayo Hospital, Lima, blood was withdrawn into citrate solution from a case of oroya fever and brought to the Rockefeller Institute, where the cultural and experimental work has been carried out.

Of the various media employed, including those which had been found suitable for the cultivation of anaerobic treponemata, as well as aerobic media used for the cultivation of the leptospiras, flagellates and rickettsia-like microorganisms, only the aerobic media, solid or semisolid, containing blood or serum yielded growth of *Bartonella bacilliformis*. The initial cultures were obtained both on leptospira medium and on blood agar slants (20 to 30 per cent. of defibrinated horse blood) containing certain carbohydrates. The organism grew in pure condition on the first attempt, and pure cultures have been repeatedly obtained from the original citrated blood. Growth occurred at 37° C. and also at 28° C. within 48 to 72 hours. Subcultures were readily obtained on similar media, and the strain has been maintained in the laboratory since the beginning of October, 1925.

<sup>1</sup> Mast, 1911, "Habits and Reactions of the Ciliate *Lacrymaria*," *Journ. Animal Behavior*, Vol. 1, pp. 229-243.