It is probable that the accuracy and clinical value of our formulae may be enhanced by altering its constants, which may be advisable after recalculation upon a larger series.

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Bacteriology of Leprosy. II. Growth and Staining Reactions of Organisms Inoculated into Minced Chick Embryo Medium.

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In a previous communication[1] a description was given of an acid-fast organism isolated from human and rat leprous lesions by the tissue culture technic.

Some of the organisms were acid-fast and some non-acid-fast in fresh tissue cultures and in minced embryo medium, but entirely acid-sensitive on the usual laboratory media.

The best growth of acid-fast organisms occurred in from 24-48 hours. There were usually as many acid-fast as acid-sensitive organisms present. After this period of incubation the acid-fast rods gradually disappeared and on about the tenth day the organisms were non-acid-fast or almost so. On transferring to fresh tissue cultures or minced embryo medium, the above picture was repeated.

The tinctorial characteristics varied depending upon the living condition of the tissues. In vigorous, actively growing tissues the organisms were strongly acid-fast. As the tissues became less vigorous the acid-fast property was less pronounced and, finally as the tissues died and autolyzed, only non-acid-fast diphtheroids were seen.

The organisms isolated from human and rat lesions revealed the same morphological and physiological characteristics. It was, therefore, concluded that human and rat leprosy are caused by the same organism.

In order to determine whether or not the acid-fast phase is characteristic of bacteria in general, when inoculated into minced embryo medium, the following organisms were tested: *Escherichia coli, Eberthella typhi, Eberthella dysenteriae, Eberthella paradysen-*

[1] Salle, A. J., *J. Infect. Dis.*, in press.

teriae (Bacillus dysenteriae Flexner), *Bacillus subtilis, Bacillus subtilis* (dissociating strain), *Bacillus anthracis, Corynebacterium diphtheriae* (American No. 8), *Corynebacterium xerosis, Corynebacterium pseudodiphthericum (hoffmanni), Staphylococcus aureus, Sarcina lutea, Rhodococcus roseus, Clostridium botulinum* (Type A), *Clostridium welchii* (Type 1), *Clostridium tetani, Streptococcus pyogenes* (beta or hemolytic strep.), *Streptococcus mitior* (alpha or viridans strep.), *Hemophilus influenzae, Neisseria intracellularis* (meningococcus), *Diplococcus pneumoniae, Saccharomyces cerevisiae* (baker's yeast), *Sporotrichum schencki, Monilia krusei, Epidermophyton cruris, Acladium castellani, Blastomyces hominis, Actinomyces violaceum, Actinomyces* isolated from air.

The minced chick embryo medium was prepared in the following manner:

Minced chick embryos (9-12 days)	1 part
Tyrode solution	5 parts
	6 parts

The embryos were decapitated, after which they were minced in a tissue grinder and mixed with Tyrode solution in the above proportion. The heads were removed[2] because the pigment granules contained in the eyes often led to confusion when smears were made to determine the sterility of the medium or to note presence or absence of growth of the inoculated organisms. About 3 cc. amounts of the medium were measured into test tubes, after which it was ready for use.

Smears were stained for 30 minutes in cold carbol-fuchsin, decolorized with 10% sulfuric acid and then counterstained with Loeffler's methylene blue.

The results showed that, with the exception of the yeast *Saccharomyces cerevisiae* and the mold-like organism *Actinomyces violaceus,* none of the organisms retained the fuchsin stain when treated with 10% sulfuric acid. The true bacteria were acid sensitive when stained by the acid-fast technic. Therefore, it may be stated that the ability of an organism to retain the acid-fast stain in minced chick embryo medium is not a phenomenon characteristic of the true bacteria (Eubacteriales), but restricted so far to organisms falling into other orders (Actinomycetales, etc.).

[2] Rivers, T. M., *J. Exp. Med.*, 1931, **54**, 453.