

difficult, but in the four animals that we have injected in this manner and which have died or been killed in from one to five days, definite consolidation of the lungs was evident in all and a sero-fibrino-purulent pleurisy occurred in all but the 24-hour case.

A histology study of the lungs in these cases apparently shows the characteristics described by MacCallum in his interstitial broncho-pneumonia, namely, plugging of aveoli with polymorphonuclear leucocytes, red blood corpuscles, serum and fibrin in definite relation to bronchi which are also filled with a purulent exudate. There is a definite infiltration of polymorphonuclear leucocytes and lymphocytes about the bronchi and blood vessels and marked desquamation of the bronchial epithelium. A further study will show in what respect, if any, this experimental pneumonia in rabbits differs from that produced by the pneumococcus.

22 (1604)

**The bactericidal action of rabbit bile on certain strains  
of streptococci.**

By RUTH L. STONE (by invitation).

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The phenomenon here described was noted during the course of a series of experiments on rabbits designed to test the pathogenicity of a certain strain of hemolytic streptococcus. It was found that, although at autopsy the various organs of the peritoneal cavity were filled with living streptococci, the bile was always sterile. This led to the testing, *in vitro*, of bile from other rabbits as well as from various other animals, to find out, whether they possessed bactericidal action on this strain of streptococcus. All samples of rabbit bile proved to be bactericidal, whereas the bile of the ox, sheep, cat, dog, pig, guinea pig, and human exerted no deleterious effect on the streptococci.

The strain of streptococcus used (Strain "H")<sup>1</sup> in these preliminary experiments was, according to Holman's classification, *Streptococcus pyogenes*—a hemolytic, non-mannite fermenting strep-

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<sup>1</sup> Gay and Stone, *J. Infec. Dis.*, 1920, xxvi, 265.

tococcus. Our next step was to test various strains of streptococci from human and animal sources, these strains having been classified according to their hemolytic and sugar fermenting properties.

The results may be briefly summarized, by dividing the organisms into three groups, at least two of which are apparently clear cut.

I. All those hemolytic non-mannite fermenting *Streptococci* which fall, by Holman's classification, into the *Streptococcus pyogenes* group, were killed by rabbit bile, 1/50 of 1 c.c. of bile, or less being sufficient to kill 0.1 c.c. of a 24-hr. serum broth culture. About twenty cultures of this type were tested.

II. All non-hemolytic *Streptococci*, whether of human or bovine origin, were unaffected by rabbit bile.

III. Hemolytic, mannite fermenting streptococci are *almost* always unaffected by rabbit bile. In a group of thirty or more of such strains tested, only two were killed by bile.

Since this bactericidal power of rabbit bile is undiminished by sterilization, attempts were made, by fractioning the bile, to determine, if possible, what constituent of rabbit bile is responsible for this highly selective bactericidal action.

Bile was dried with sand, to give greater surface for extraction, and the resulting mixture ground and treated with absolute alcohol, thus precipitating the proteins. The resulting filtrate was evaporated to dryness and then resuspended in broth to the original volume of the bile. This was sterilized and tested for its bactericidal power, which was found to be undiminished. Next, a portion of this alcoholic extract was treated with absolute ether, causing a further precipitate. Both filtrate and precipitate were dried and resuspended in broth, and tested as before. It was found that only the precipitate contained this bactericidal substance. It may be of interest here to note that Neufeld found that the pneumococcus dissolving substance of bile was also located in this fraction. However he found this to be true of various types of bile, whereas the phenomenon here described only occurs with rabbit bile, and is a bactericidal and not a lytic process, since the bacterial bodies are visibly intact even after 48 hours.

On treating the alcohol soluble fraction with acetone, both

filtrate and precipitate were found to be slightly bactericidal, but neither equal to the original power of the alcoholic extract.

It is evident, therefore, that this bactericidal substance in rabbit bile for certain strains of streptococci, is present with or identical with a bile salt, being precipitated by ether, and alcohol soluble. However, since other types of bile do not give these reactions which seem to be peculiar to rabbit bile, one must conclude that rabbit bile either has some substance in its composition that is not found in other types of bile, or that its chemical construction is different, thereby giving it this peculiar property.

23 (1605)

**The viability of *B. typhosus* in alkaline bile in vivo.**

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In as much as Nichols suggests the use of alkaline therapy for the purpose of eradicating *B. typhosus* within the gall bladder of human carriers of the disease, the following observations are pertinent.

While carrying out a series of tests with experimental rabbit carriers of typhoid, it was noted in a certain instance that the hydrogen ion concentration of the bile was different from that supposed to characterize the normal animal. This indication was followed with other animals as opportunity presented itself.  $P_H$  determinations were made on the bile of uninfected animals as materials appeared. The method followed was that of Clark<sup>1</sup> and Lubs with the comparator block introduced into the system. Readings were made as soon after the death of the animal by exsanguination as possible, generally within three quarters of an hour. In order that contact with the air and consequent loss of dissolved gases might be reduced to a minimum, the bile was kept either within the closed syringe with which it had been aspirated or was placed within a small bore agglutination tube. All animals

<sup>1</sup> Clark, W. M. and Lubs, H. A., *Jour. Bact.*, 1917, ii, 1-34, 109-136, 191-236.