was mixed with a known volume of diluent and the total volume of the resulting suspension measured. The concentration of the cells present was computed from the difference in the two volume measurements. Formalized red cells possessed negligible buffering power while normal red cells are known to exert a buffering action in suspension.[8] Probably due to modification of protein amino groups among others by the formaldehyde, the constituents of the cells could no longer appreciably unite with hydrogen ions. In the absence of a buffered diluent it was found difficult to maintain the suspension of cells at a constant pH. Finally, while sedimented normal red cells can be resuspended rather easily, suspensions of formalized red cells, upon sedimenting, formed a mass of almost rubbery consistency and required vigorous stirring to resuspend the cells thoroughly. These disadvantages attending the use of formalized red cells are of minor importance and are greatly outweighed by the advantages of red cells stabilized by treatment with 10 to 50% formalin for several days.

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Practical Method for Determination of Serum Streptomycin Level Using *"Donovania granulomatis"* as Test Organism.*

ROBERT B. DIENST, CALVIN H. CHEN, AND ROBERT B. GREENBLATT.
(Introduced by G. Lombard Kelly.)
From the University of Georgia School of Medicine, Augusta, Ga.

Following the successful cultivation of *"Donovania granulomatis" in vitro* in fresh yolk medium by Dienst, Greenblatt and Chen,[1] the technique for propagating this organism has been made comparable in simplicity to those used for ordinary bacteria. With the demonstration of the high degree of susceptibility of *"Donovania granulomatis"* to streptomycin,[2,3] it was conceived that this heretofore hard-to-culture micro-organism might be used in streptomycin assay. As a result of a series of experiments the following procedures have been evolved, which we consider as being simple and accurate enough for any clinical or bacteriological laboratory to use. The principle of this method is based upon the comparison of the least amount of serum to a known quantity of streptomycin that has to be present in the culture medium in order to inhibit the growth of *"Donovania granulomatis."*

The fresh yolk medium is prepared in the following manner: Yolk is removed aseptically from fresh eggs and diluted 1:1 with sterile saline. A base medium containing 1% Bacto peptone, 0.3% Bacto tryptone, 0.3% dextrose, 0.2% sea salt, and 0.12% agar is prepared and adjusted to pH 7.3. The base medium is then sterilized and allowed to cool to 50-60°C before adding an equal portion of the diluted yolk. Four cc of this medium is placed in each of a series of 8 test tubes. The serum to be assayed is diluted with sterile distilled water to 1:1, 1:2, 1:4, 1:8, 1:16, 1:32, 1:64, and 1:128. To the first tube of yolk medium is added 1 cc of the 1:1 dilution of serum, to the second tube is added 1 cc of the 1:2 dilution of serum, and so on, so that the final volume of each

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tube is 5 cc. The serum and the medium are thoroughly mixed by vigorous shaking. Then 0.1 cc of a 4-day-old culture of *"Donovania granulomatis"* is added to each tube and again mixed well by shaking. These tubes are incubated at 37°C and on the fourth day smears are made from each and stained with Wright's stain and examined under oil immersion lens for the presence of the Donovan micro-organism. The tube containing the least amount of serum that inhibited the growth of the organism is regarded as the endpoint.

Calculation. The least amount of streptomycin that will inhibit the growth of the organism has been found by a similar procedure to be 0.075 γ/cc. Therefore, the serum streptomycin level is equal to 0.075 × Serum Dilution × Total Volume of Medium × 2. For example, if the endpoint of the assay is tube 5, which contains a serum diluted to 1:16, the serum streptomycin level is 0.075 × 16 × 5 × 2 = 12 γ/cc.

With this simple method, a serum level of 0.75 γ/cc to 48 γ/cc can be determined This range embraces the serum levels that are commonly encountered in streptomycin therapy. However, with slight modifications, such as using undiluted serum as the first tube and diluting the serum further, one is able to determine a streptomycin level from 0.375 γ/cc to any level higher than 48 γ/cc.

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Electron Microscopic Observations on *Pseudomonas aeruginosa* Bacteriophage.

E. W. SCHULTZ, P. R. THOMASSEN, AND L. MARTON.*

From the Department of Bacteriology and Experimental Pathology, School of Medicine, and the Division of Electron Optics, Stanford University.

Isolations of bacteriophages active for *Ps. aeruginosa (B. pyocyaneus)* have been reported on a few occasions only. For a time the iridescent erosions which may appear in pyocyaneus colonies were interpreted as manifestations of bacteriophage activity.[1-3] It has now been established, however, that these iridescent changes are unrelated to phage activity and that bacteriophagy among pyocyaneus bacilli is not unlike that observed in other species of bacteria.[4-11] Fastier[12] has recently reported what he erroneously believed to be the first isolation of a pyocyaneus bacteriophage. Between 1930 and 1934 Schultz and Sheets[13] isolated 4 pyocyaneus bacteriophages which are distinguishable from each other and from the one which is the subject of this report.

The present report is based on a strain isolated by one of us (E.W.S.) in 1943 from the exudate of an extensive third degree burn heavily infected with *Ps. aeruginosa*. It bears our number "Phage 238" and is distinguishable from our earlier strains (Phages 75, 139, 177 and 195)[13] by its higher lytic titer and

* Present address: National Bureau of Standards, Washington, D.C.

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