currence of oxidation in case of air saturated suspension, but it is clear that cavitation of dissolved gases are essential in bringing about killing and dissolution of bacteria.

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Photometric Study of Bacteriophage Action.

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Krueger and Northrop[1] were the first to undertake the study of bacteriophage action from a quantitative point of view. Based on extremely elaborate technical procedures, involving frequent and separate enumeration of bacteria and of bacteriophage throughout each experiment, they concluded that production of phage was proportional to a power of bacterial growth and that lysis set in almost explosively at the moment when phage/bacteria ratio attained a definite critical value. It would appear that according to their result the time required for lysis of a given bacterial concentration is proportional to the dilution of phage. Other conditions being constant, the strength of any 2 phages can be compared by noting the relative length of time necessary for reduction of a constant concentration of bacteria to an arbitrary end point.

The present paper is not intended to add anything new to the mechanism of bacteriophage action, but rather to present an accurate though considerably simpler device for attacking the problem from the same viewpoint. The apparatus used is a Pulfrich photometer which works on the principle of the Tyndall phenomenon, so that for a given bacterium, the number of organisms per cc. can be read off directly and quickly and is expressed in terms of percentage of a given standard. Very minute particles, namely, phage or protein particles, liberated during the process of dissolution of bacteria present a very weak Tyndall phenomenon and do not, therefore, affect the readings. Provided the bacterial suspension is not so thick as to interfere with penetration of light, the accuracy of readings obtained approached $\pm 0.5\%$ of the given standard, disregarding slight variations in the size and thickness of the tubes used.

[1] Krueger, A. P., and Northrop, J. H., *J. Gen. Physiol.*, 1931, **14**, 223, 493.

The phage used throughout the experiment is from d'Herelle's laboratory and has been in our stock since 1927. Sixteen to 18 hours' agar culture of a single strain of *B. dysenteriae* Shiga was suspended in meat-infusion broth, pH 7.6, and distributed in 9 cc. lots to a series of test tubes, to each of which 1 cc. of various dilutions of phage in saline was later added. With a view of securing uniform distribution of bacteria and phage, the suspensions were incubated in a mechanical shaker in a water bath at 32°C. Readings were taken every few minutes till the completion of lysis.

The results of a large number of experiments conducted with similar or modified techniques such as checking bacterial growth by limiting the source of nutritional supply, by substituting saline for broth, or by employing a thick initial bacterial suspension, or omitting constant shaking, all bore out the same conclusions which may be conveniently represented by the 3 accompanying graphs for (1) various dilutions of phage against a constant bacterial concentration, (2) constant dilution of phage against various bacterial concentra-

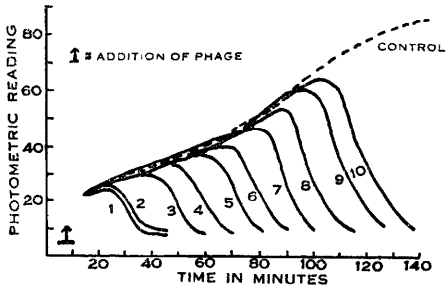

FIG. 1.
Various dilutions of phage (10^{-1}-10^{-10}) against constant bacterial concentration.

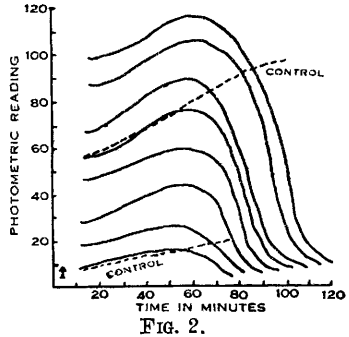

FIG. 2.
Constant dilution of phage (10^{-4}) against various concentrations of bacteria.

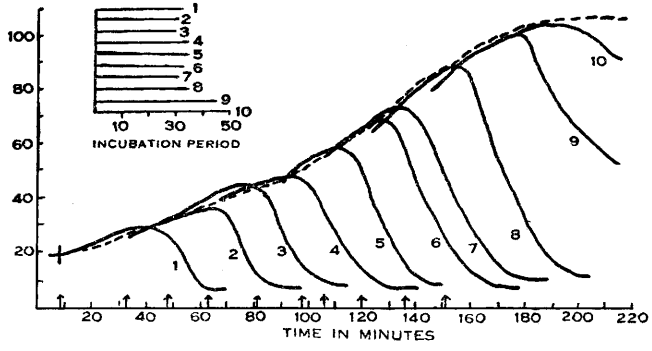

FIG. 3.
Constant dilution of phage (10^{-4}) added to constant initial bacterial concentration at various intervals of growth period.

tions and (3) constant dilution of phage added to constant initial bacterial concentration at different intervals of growth period.

Fig. 1 brings out the fact that with constant bacterial concentration, the incubation period (period between addition of phage and onset of lysis) is a function of dilution of phage, so that by plotting concentrations of phage as abscissa and incubation period in minutes as ordinate, a straight line connecting all points is obtained, except for very high concentration of phage. The latter is due to the fact that no matter how concentrated the phage is, there is always bound to be a minimal incubation period. When the experiment was repeated, a different slope was met with, but a similar curve conforming to a straight line is always obtained. It is interesting to note that within wide limits, the difference in the initial bacterial concentration does not influence the incubation period. (Fig. 2). This is to be expected, considering that production of phage is dependent on the growth of bacteria and that since a thicker suspension contains more bacteria in the process of active division, the increase of bacteriophage should also be proportionately greater. On the other hand as may be seen from the curve, the greater the concentration of bacteria at the end of incubation period, the longer the time required for clearing up of the culture. Here again the relation is that of a direct proportionality.

That the rate of production of phage depends on the multiplication of bacteria is also brought out in Fig. 3. Here, one may note that as growth of bacteria begins to slow down, the incubation period is also lengthened. Moreover, lysis of bacteria ushered in at that period is usually prolonged and frequently incomplete. It is well known that bacteria which have passed the stage of active growth become more resistant to the action of phage or perhaps a certain number of bacteria have died off; in the latter case they are not only refractory to the lytic action of phage but are able to remove the greater number of phage particles from the surrounding broth than would living, susceptible bacteria.

The above observations indicate that it is possible to apply the Pulfrich photometer to the study of bacteriophage action with the same precision as that obtained through the use of more intricate methods hitherto described by others.