amounts of alveolar air caught in the test chamber. The 3 long groups are samples taken automatically. It can be seen that such sampling was necessary at this respiration rate. During the sampling, the heart rate and electrocorticogram do not change, indicating that there were no appreciable disturbances by vagus reflexes. The next example shows a slower respiration of about 60/minute, and here it is obvious that the sampling was unnecessary. The CO_2 concentration does not become any higher during prolongation of expiration. The third example demonstrates conditions under severe hypoxia, which was induced shortly after example 2. The alveolar CO_2 has dropped considerably, and it remains low throughout the gasping period which follows until oxygen is resupplied. The immediate increase to a very high CO_2 concentration is shown in the last strip, which was recorded during post-anoxic convulsions. Under the conditions of the last two examples, automatic sampling was proven unnecessary and even undesirable because of increased vagus excitability. When such a high excitability is combined with a high respiration rate, the described sampling method is not feasible.

Summary. An infra-red CO_2 analyzer having a measurement rate of seven analyses per second is described for continuous CO_2 tension determination over the expiration time (or volume). The value recorded from the last portion of expired air is regarded as "the alveolar CO_2 tension" if a plateau has developed during expiration. Respiration in small animals is often so rapid (relative to the measurement rate) that forced prolongation of a single expiration is necessary in order to draw undiluted alveolar air into the analyzer. A simple method is described for accomplishing this by means of a valve whose closing is electronically initiated and timed.

Received March 28, 1949. P.S.E.B.M., 1949, **71.**

17112 P. Multiplication of the T3 Bacteriophage Against *E. coli.*

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Steps in the development of new bacteriophage particles from diseased bacteria are particularly well seen in electron micrographs of young and frequently transferred colon bacilli infected with the T2 strain of their bacteriophage.[1] Examination of such photographs has suggested that this bacteriophage is self-reproducing with a mode of multiplication that in some respects resembles that of coccoid bacteria. An unforeseen result of these observations has been the rapidity with which lysis, understood as rupture of the bacterial membrane, appears after infection with bacteriophage. It has usually been taken for granted that when a bacteriophage particle is adsorbed to a susceptible bacterium, it proliferates either at the surface or after penetrating the organism until, at the conclusion of the multiplication process, the cell bursts and frees the new particles that have been forming. Evidence from the electron micrographs demonstrates that the membranes of young colon bacilli in a growing culture rupture soon after mixture of the cells with enough T2 particles to disease them. On this basis the "burst interval" observed for these bacteriophages is determined by the dispersal of newly formed bacteriophage from the disappearing protoplasmic mass in which it has been developing.

Further light upon this and other aspects of the multiplication of bacteriophage has now been obtained by investigating the lysis of young *E. coli* by the two different bacteriophages, T1 and T3.

When a one-hour culture of the B strain of *E. coli* obtained by two or three transplants

[1] Wyckoff, Ralph, W. G., *Nature*, 1948, **162**, 649.

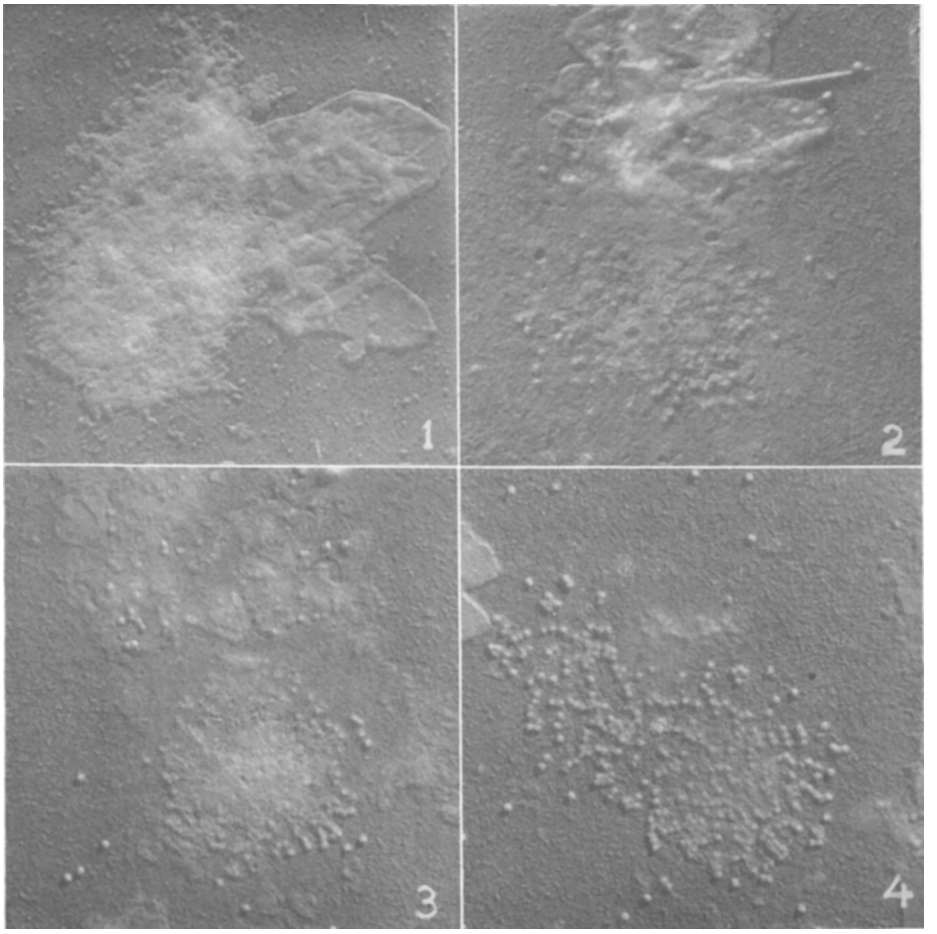

FIG. 1. A colon bacillus lysed through the action of T3 bacteriophage. From a culture after 5 minutes incubation. The cell membrane is at the right, the extruded granular protoplasmic contents at the left. Palladium shadowing on this and the following three preparations. Magnification, 16,667 ×.

FIG. 2. Particles of T3 phage developing within the protoplasm of a lysed colon bacillus. The empty cell membrane is at the top of the picture. Magnification, 16,667 ×.

FIG. 3. A somewhat more advanced stage in the development of a microcolony of T3 within protoplasm from a lysed bacterium. Magnification, 16,667 ×.

FIG. 4. A still later stage of phage production in which most of the cellular protoplasm has been consumed and the new particles are beginning to disperse. Magnification, 16,667 ×.

at hourly intervals has been mixed with enough T3 bacteriophage to infect at once all the bacteria present, electron micrographs have been obtained that agree with the earlier ones using T2 in showing prompt lysis; after 5 minutes incubation followed by immediate icing very few intact organisms have been found in centrifuged, formalinized and washed preparations. Such rupturing has not been observed in control preparations from which the bacteriophage has been omitted. When T1 bacteriophage of the same titre has been substituted for T3 many, but far from all the young cells in a culture have been quickly lysed. As Fig. 1 indicates, the bacterial protoplasm liberated by these bacteriophages is the same mass of spherical macromolecular particles seen after lysis with T2.

Preparations made after incubating for intervals up to half an hour following infection

with T3 have shown various steps in the development of new bacteriophage particles in the protoplasm of infected and lysed cells. After 15 minutes incubation fields like those of Fig. 2, 3 and 4 are common. The lysed cell of Fig. 2 resembles that of Fig. 1 in showing its protoplasm extruded from the broken membrane. Much of this protoplasm has, however, already been converted into new particles of T3. The conversion is still more complete in Fig. 3 where the cluster of coccoid T3 apparently is developing from one or more central particles. This cluster recalls a bacterial microcolony resulting from several divisions of a single cell. A still later stage wherein the newly formed particles have commenced to disperse is shown in Fig. 4. After a few more minutes incubation the formation of bacteriophage has progressed so far at the expense of the lysed protoplasm that it is hard to find either the protoplasmic masses or the clumps of bacteriophage evident in these earlier photographs. Many electron micrographs like the foregoing have been obtained; they exhibit T3 particles in small groups and chains that suggest they have arisen by the same kind of self-reproduction which is often seen in bacterial growths and which appears to underlie the multiplication of the T2 coli bacteriophage.

Summary. Cultures of young, rapidly growing susceptible colon bacilli obtained after hourly transplants to fresh broth appear quickly lysed by the addition of T3 bacteriophage. Electron micrographs show that the bacterial membranes rupture very soon after adsorption of bacteriophage and that colonies of new phage particles rapidly appear in the protoplasmic masses. This protoplasm from young cells is the same assemblage of spherical macromolecules seen after lysis with T2 phage. As was the case with this other phage the observed phenomena are those to be expected of particles behaving as if they were minute microorganisms multiplying by division at the expense of the bacterial protoplasm.

Received March 14, 1949. P.S.E.B.M., 1949, **71**.

17113. Effect of Chemical Irritation of the Peritoneum on Transmural Migration of Intestinal Organisms.

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For reasons already given elsewhere[1] it is believed that the peritonitis which develops during peritoneal irrigation for the treatment of acute renal failure is due to the transmural migration of bacteria from the intestine. While there is no evidence that this can occur in the absence of severe damage to the intestinal wall, it might be possible as a result of the combined effect of the uremic state and the irritating properties of the dialyzing fluid. In the absence of clinical evidence for or against such a hypothesis, animal experiments were performed to see if transmural migration of intestinal bacteria can be induced (1) in normal animals subjected to chemical irritation of the peritoneum and (2) in normal and uremic animals exposed to the intraperitoneal injection of the fluids used for peritoneal irrigation in acute renal failure. The results of the first part of these studies are herewith reported.

Method. Healthy rabbits and dogs received sterile physiologic saline solution or the fluid used for peritoneal dialysis of uremic patients, in a volume sufficient to grossly distend the peritoneal cavity twice daily up to 2 weeks. Peritoneal fluid was aspirated as completely as possible just before each infusion. The as-

[1] Frank, H., Seligman, A., Fine, J., *Annals Surg.*, 1948, **128**, 3.