eous activation confirms previous studies of this relationship. The activation by protamine is definite, though in most instances not as great as is that by streptokinase (Fig. 3). This is at variance with the previous impression that the fibrinolytic activity of protamine was due to inactivation of antifibrinolysin.[10] In the present study protamine sulphate (up to 1.2 mg/tube) failed to diminish antiproteolytic activity tested by the method previously reported.[8]

Dog sera differ from human in that their spontaneous activation is of the same order as the protamine and streptokinase activation (Fig. 3).

Summary. A method for preparation of plasminogen from small quantities of blood has been described. Activation by streptokinase, protamine sulphate, and spontaneous activity have been observed. Studies of other activators and of species differences are in progress.

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10 Scroggie, A. E., Jaques, L. B., Rocha e Silva, M., PROC. SOC. EXP. BIOL. AND MED., 1947, **66**, 326.

17111. Automatic Measurement of Alveolar CO_2 in Small Animals.

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The following paper will describe the application of an infra-red analyzer to alveolar carbon dioxide studies in small animals such as the rabbit, where rapid respiration and small respiratory volumes pose severe problems. As reported elsewhere,[1] this analyzer has been used successfully in measuring the alveolar CO_2 tension in humans during hyperventilation as it is performed in routine electroencephalography. The analyzer was developed for other purposes by Luft[2] and applied to CO_2 measurement by Schmeiser.[3] It measures the infrared absorption of one gas in a mixture by using a fixed quantity of the same kind of gas as a thermomechanical metering device. This method allows an analysis which is rapid enough to fully record a change in CO_2 tension in one-seventh of a second. Thus, in human studies, alveolar air sampling devices are unnecessary, and a portion of the expired air is drawn continuously through the analyzer. The latency of measurement then depends solely upon the speed of transport of this air through the test chamber. The only additional equipment required is a long expiration tube to prevent dilution of the continuous sample by outside air. The measured CO_2 value after a certain expired volume can be safely regarded as the conventional "alveolar CO_2 tension."

Fig. I illustrates the CO_2 analyzer in simplified form. A hot-wire infra-red source (S) projects a beam of infra-red through a transparent (mica) test cell (T) containing the air for CO_2 analysis. The remaining infra-red is absorbed in a following chamber (A), one wall of which is a thin metal membrane. The temperature increase due to the absorption displaces the membrane by increasing the pressure, thus measuring the amount of infra-red reaching the chamber. If the chamber is filled with CO_2, it becomes sensitive to only those wave lengths which the CO_2 in the test cell absorbs. Thus, it is insensitive to any other gases appearing in the test cell. To balance the system, an identical arrangement (S', C, A') is attached to the other side of the membrane, and the two sides "buck" each other. Displacements of the membrane are measured by utilizing it as one plate of a condenser and

1 Blinn, K. A., and Noell, W. K., *EEG Clin. Neurophysiol.*, 1949, in press.

2 Luft, K., *Z. f. Tech. Phys.*, 1943, **24**, 97.

3 Schmeiser, K., unpublished work.

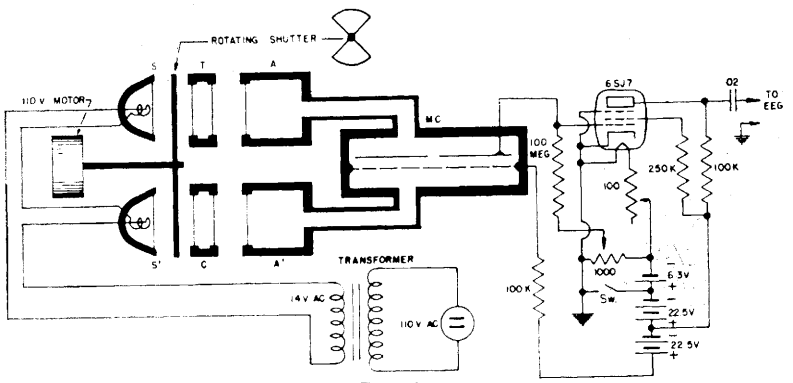

FIG. 1.

Diagram of the CO_2 analyzer and its associated preamplifier. S,S'—hot wire infra-red sources; T,C—test and control chambers; A,A'—absorption chambers of membrane-condenser, MC.

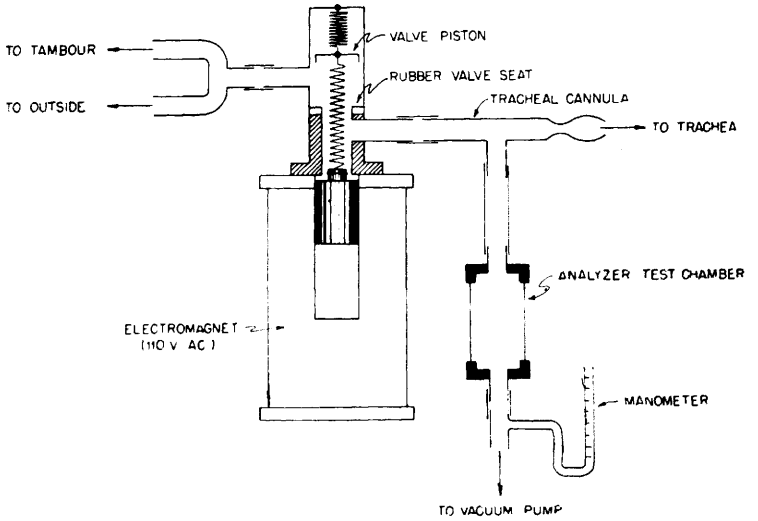

FIG. 2.

The alveolar gas sampling apparatus. The connections between the electromagnetic valve, the tracheal cannula, and the analyzer test chamber are made as short as possible.

measuring changes in electrical charge on the condenser. To produce an alternating voltage for amplification, the two infra-red beams are simultaneously interrupted 7 times per second by a rotating shutter. After amplification the output of the analyzer can be recorded on any instrument, for example, an electroencephalograph. The output appears as a train of 7c/s waves, the amplitude of which is proportional to the CO_2 concentration within limits determined by the thickness of the test cell.

Respiration in humans, even during hyperventilation, is conveniently slow; but in small animals, such as the rabbit, it may be so rapid that the seven-per-second shutter frequency is proportionally too low, and the respiratory volume exchange becomes a critical factor. Unfortunately, the shutter frequency can be increased only at the expense of a major part of the sensitivity. In these animals, the respiratory rate may exceed 200/minute. Therefore, in order to obtain an undiluted alveolar sample in the test chamber, it was necessary to artificially lengthen a single expiration whenever such a rate prevailed. Fig. 2 shows how this was managed. A tracheal cannula

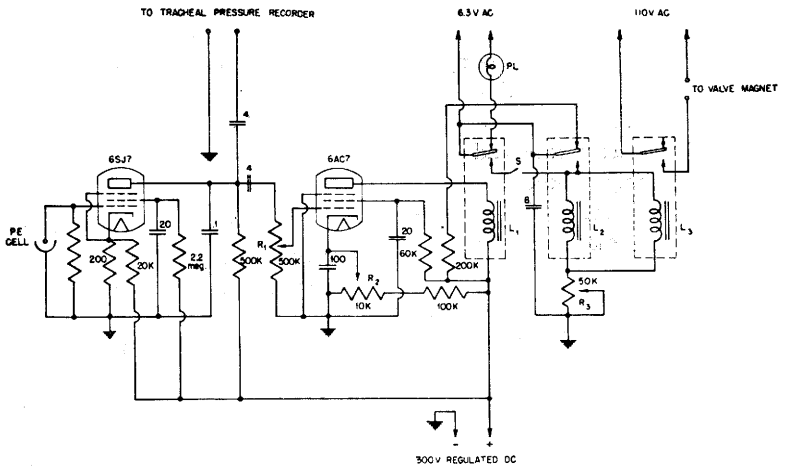

FIG. 3.

The electronic circuit for activating the valve. Bias is adjusted beyond cut-off with R_2; R_1 is then advanced until the pilot light PL begins to extinguish at the peak of expiration. When the switch S is closed, the valve magnet is activated for a single sampling of predetermined duration. R_3 varies the sampling duration from 1/3 sec. to 2 sec. L_1, L_2, and L_3 are 10,000 ohm relays.

was used, with a side arm leading to the test chamber and suction pump. To obviate disturbances of the respiratory rhythm and vagus effects, an automatic system was devised for briefly closing the cannula at the peak of the expiratory pressure curve. An electromagnetic valve closes the distal end of the tracheal cannula so that an alveolar sample is drawn through the analyzing chamber by the pump. The onset and duration of the closing are controlled electronically in the following manner: tracheal pressure fluctuations are transformed into voltage fluctuations by means of a tambour-operated shutter in front of a photoelectric cell. The electrical fluctuations operate a series of relays (Fig. 3) in such a manner that when a sampling switch is closed, the electromagnetic valve closes at the peak of the next expiration and remains closed only for a preset length of time.

Fig. 4 illustrates the result. The CO_2 is recorded on the electroencephalograph tape simultaneously with cortical potentials, the EKG, and the tracheal pressure. Each strip is about 12 seconds long. In the top example, the respiration rate is about 180/minute, and the short groups of waves represent the CO_2 concentrations over single expirations. The highest amplitude varies from one group to another because of the varying

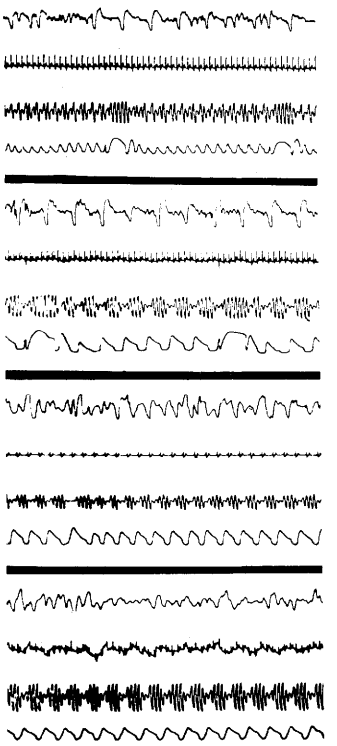

FIG. 4.

Ink-writer recordings illustrating alveolar CO_2 measurements in a rabbit. From top to bottom, the tracings in each strip are: electrocorticogram (area striata), EKG, CO_2 tension, and tracheal pressure. Description in the text. The cortical activity is being driven by one-per-second photic stimulation.

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.

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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.