prevented by giving oxygen instead of air to breathe, at pressures of 380 and 190 mm Hg; and, second, the phenomenon occurs when the rats are given, at 760 mm Hg pressure, a mixture of 10½ per cent. O_2 and 89½ per cent. N_2 to breathe. These two experiments also show that pressure changes *per se* are not responsible for the negative water balances. Furthermore, it is probable that anoxia causes the phenomenon because of the hyperventilation which it induces. When rats were made to hyperventilate by giving them gas mixtures high in CO_2, keeping the O_2 constant at 21 per cent., a negative water balance of considerable magnitude is induced. Thus, with 5 per cent. CO_2, it was 5.1 cc per 100 gms of rat; with 10 per cent. CO_2, 4.7 cc and with 15 per cent. CO_2, 5.2 cc. These figures approach, but do not quite reach, the negative water balance of exposure to 380 mm Hg pressure (5.9 cc; see Table 1). Hyperventilation in effect passes more air over the evaporating surface of the lung and there results a greater water loss.

We feel that this negative water balance may be of importance in the etiology of pilot fatigue. The latter is postulated by Armstrong[1] to be closely related to adrenocortical insufficiency. In our opinion, the chain of events leading to this insufficiency is as follows: first, a water loss, as demonstrated here, and, second, a renal salt loss, due to the previous water loss[2] and to the relative alkalosis of acapnia.[3,4] Such a salt loss has been shown to occur in mountain climbers[5,6] and during exposures to low oxygen tensions.[7] It appears to be in some way mediated by the adrenal cortex.[7] The combined salt and water loss, unaccompanied by thirst, puts a considerable strain on the adrenals; if repetitive, it would tend to produce a subacute adrenocortical insufficiency. McCance[8] has described a somewhat similar situation in which great loss of body salt and water was unaccompanied by thirst. This resulted, in his experiment, in a condition simulating adrenal insufficiency and in a train of symptoms startlingly like those of pilot fatigue.

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SCIENTIFIC APPARATUS AND LABORATORY METHODS

DETERGENTS AND STAINING OF BACTERIA

IN order to obtain a satisfactory microscopic preparation of bacteria, in which cells are uniformly distributed, it is necessary to have slides which are thoroughly cleansed by chemical methods, flaming, or both. Otherwise, the high surface tension caused by the presence of fat-like substances on the surface of the slides produces an uneven and massed distribution of the bacteria.

In course of the routine laboratory work it has been found that satisfactory preparations could be made on slides which were cleaned mechanically from the dust particles with a piece of cloth, if to the suspension of the bacteria a small amount of a detergent was added. The following procedure gave satisfactory results.

Water solutions of "Aerosol OT" or a saline solution of "Aerosol MA"[1] 1:500 were kept at hand. One loopful of one of those dilutions was placed with a loop on the slide. Bacteria were added from the liquid or solid media to this drop of detergent and the suspension was spread uniformly over the desired area with the loop. Preparations were air dried without heating, fixed with methyl alcohol or heat, and stained in the usual way. No detrimental effect of the presence of the detergent on the quality of the staining was noticed.

It was also found that the preparations made for the staining of the flagella gave the same results on the slides which were cleansed mechanically and the bacteria were suspended in distilled water containing "Aerosol OT" in the dilution 1:1000, as on the slides which were cleansed chemically and flamed with the bacteria suspended in distilled water alone. The quality of the flagella preparations was still better, however, when washed and flamed slides were used and the bacteria were suspended in the "Aerosol OT" solution. In such preparations the distribution of bacteria on the slides and the arrangement of the flagella were found most satisfactory.

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[1] Samples were kindly supplied by the American Cyanamid and Chemical Corporation.

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