A NOTE ON THE AEROBIC CULTURE OF ANAEROBES AT HIGHER TEMPERATURES

A POSSIBLE inverse relation between the temperature and oxygen tension requirements of bacteria has been indicated by Rabinowitch[1] who showed that the minimum temperature requirement of certain supposedly ortho-thermophilic organisms refusing to grow aerobically below 50° C. could be reduced at least to 37° C. by anaerobic culture. Her results account for the finding of thermophilic bacteria as parasites in the human intestine by MacFadyen and Blaxall.[2]

We have had no occasion to confirm the work of Rabinowitch as to the anaerobic growth of thermophilic aerobes as lower temperatures but we have taken advantage of the opportunity afforded in a collection of obligative anaerobes to test the converse possibility, that is the aerobic growth of anaerobes at a higher temperature. A successful result would perhaps have provided a simple means of surface culture for purposes of isolation in certain cases but the results were clearly in the negative.

It does not matter for this purpose that some of these cultures are as yet incompletely identified. The list, showing sources and the identity of the known forms, is to be published shortly in the *Journal of Bacteriology* in a paper describing our work on the inhibitory action of gentian violet and its application in preventing spurious presumptive tests due to these organisms in the bacteriological examination of water. Cultures of *B. botulinus, B. tetanus, B. chauvei, B. œdematis maligni* and the Ghon Sachs bacillus, were included among the twenty-one. All were free from ærobic contamination, as shown by tests on agar slants at 37° C. although we can by no means be certain that some of the unidentified cultures do not consist of more than one species of anaerobic microorganism.

Media containing 1 per cent. glucose, 1 per cent. peptone and 3 per cent. agar were used both for the anaerobic controls inoculated as shake cultures for incubation at 37° C., and the ærobic tests slanted for streak inoculation and incubation at 54° C., in a constant temperature acetone bath. Three per cent. agar was necessary to withstand the latter temperature for the period of the test, fifteen days. Three separate trials were made as follows: In the first, subcultures were made from stock cultures several days old in deep sterilized beef brain. These could not be considered certainly negative due to the resemblance of the transferred brain to surface growth. In the second trial, subcultures were made from 24-hour glucose broth cultures in the constricted tube and marble device for anaerobiosis.[3] Nothing developed on the surface of the slanted agar incubated at 54° C. which could be considered a bacterial growth. This test was repeated with identical results.

In a fourth test, 24-hour glucose broth cultures in constricted tubes were transferred in quantities of 1 c.c. per tube to melted glucose 2 per cent. agar at 42° C. and hardened in the upright position as shake cultures. It was thought that if the premise of this study were true, the greatest growth should occur nearer the surface in the test at 54° C. than in the control incubated at 37° C. But no growth occurred aerobically or anaerobically at 54° C. This test was duplicated in method and results.

The controls at 37° C. gave vigorous growth within 48 hours in every case as evidenced by the distinct and characteristic colonies or opacity and all but four produced abundant gas. The freedom of the control tests from ærobic contamination was also proven by failure of growth on plain agar subplants at 37° C.

Lillian Jordan Ellefson,
Ivan C. Hall

Department of Pathology and Bacteriology,
University of California

[1] Rabinowitch, "Ueber die thermophilen Bakterien," *Zeitschr. f. Hyg.*, 1895, XX., 154.

[2] MacFadyen and Blaxall, "Thermophilic Bacteria," *Jour. Pathology and Bacteriology*, 1896, III., 87.

[3] Hall, "A New Aerobic-Anaerobic Culture Tube," Univ. of Calif. Pub. in Pathology, 1915, II., 147.