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Factors Affecting the Survival of *Treponema pallidum in vitro.**

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(Introduced by Thomas B. Turner.)

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Preliminary quantitative experiments have been undertaken in an attempt to define various factors which influence the survival of *Treponema pallidum in vitro.*

Utilizing virulent *T. pallidum* (Nichols strain) obtained by Waring Blendor emulsification of rabbit testes in the early stage of acute syphilitic orchitis, it has been possible to compare the *in vitro* survival time of the organisms at a constant pH 1) in various gaseous atmospheres and 2) at varying temperatures.

Brewer's Fluid Thioglycollate Medium, used by one of us in previous work on the Reiter spirochete,[1] was arbitrarily chosen as a basal medium. This was supplemented by isotonic phosphate buffer and crystalline bovine albumin (Armour Laboratories) prepared as a 7% solution in isotonic saline pH adjusted to 7.0 with 1 *M* NaOH. Viability has been determined by counting 50 organisms at random under darkfield examination and recording the percent motile. To date, all specimens exhibting motility have proven virulent as demonstrated by intracutaneous rabbit inoculation. There has been no increase in the total number of organisms on prolonged incubation.

Since survival has repeatedly been limited to less than 24 hours at 37° C incubation, lower temperatures, 32° C and 27° C, have been utilized throughout this series of experiments.

It has been noted that survival of the organism is consistently prolonged when a 5% carbon dioxide, 95% nitrogen atmosphere is present over the culture tubes in the Brewer anaerobe jar. Half-life (*i.e.*, 50% of original number of motile organisms remaining motile) has been prolonged to 96 hours in specimens exposed to 5% CO_2 as compared to less than 24 hours of controls in 100% nitrogen or hydrogen. Specimens freely exposed to air had an even shorter half-life, *i.e.*, less than 12 hours.

Summary. 1. Preliminary observations on the survival of the virulent Nichols strain of *Treponema pallidum in vitro* indicate that under a given set of experimental conditions exposure to 5% CO_2 had a definite prolonging effect on survival time. 2. Survival was improved when temperatures below 37° C were used. 3. No indication of multiplication of the organisms has been noted. 4. Further quantitative experiments are in progress in an attempt to clarify the role of CO_2 and to define other physical and chemical factors which may affect this organism's survival *in vitro.*

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1 Eagle, H., and Steinman, H. G., *J. Bact.,* in press.