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The cultivation of *Bact. abortus* Bang.

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Many methods have been described for the cultivation of *Bact. abortus* Bang. Following the original work with this organism by Bang and Stribolt in 1897, several investigators have described methods for the cultivation of this germ. Those which are most frequently mentioned, and used are those of Nowak, Holth, Priez, and Fabyan. Recently (1921) Stafseth and Huddleson have described culture media and methods of growing *Bact. abortus* which differ from those already in use. The former recommends a media prepared from liver and spleen. He states that "strains of the abortion bacillus have been isolated more easily by the aid of these media." A glass jar from which the air was partially exhausted by a suction pump, was used in which to grow the cultures. Huddleson emphasizes the importance of an increased carbon dioxide-tension for growing *Bact. abortus* Bang. His conclusions are as follows:

"There is sufficient proof that:

- "(1) The growth of *Bact. abortus* is not due to a reduced oxygen tension.
- "(2) A carbon-dioxide tension greater than that of the air governs and greatly facilitates the primary growth of *Bact. abortus*.
- "(3) An atmosphere containing (by volume) 10 per cent. of CO₂ gas appears to produce the earliest and most luxuriant growth of *Bact. abortus*."

Huddleson recommends the use of a generator containing calcium carbonate to which hydrochloride acid is added as a source of the CO₂.

We have been working with *Bact. abortus* for many years and have experienced the same difficulty of isolating the organism as described by other authors. All the mentioned methods have been used with more or less success. The abortion germ usually grows with great difficulty in the cultures made from the original

infected material as the stomach contents of an aborted fetus. After two or three transfers, the bacilli grow quite readily even under ordinary aërobic conditions.

We have been using the method described by Huddleson, but have used in place of a CO₂ generator described by him, commercial CO₂ from a tank. This is because too much chlorine was given off with the gas from the Huddleson generator. We also used a serum agar media, the agar being made from lean beef and when used 10 per cent. of naturally sterile horse serum was added to the melted agar, cooled to 50° C., and the tubes are allowed to solidify in a slanting position. The reaction of the agar is adjusted to a P_H of 6.8 to 7.2, which is slightly more alkaline than heretofore recommended.

These culture tubes are heavily seeded with the material to be cultured, and are placed in a Whitall Tatum museum jar to which 10 per cent. CO₂ is added. The jar is then sealed and placed in an incubator at 37½° C. After 24 hours' incubation, small pin-point colonies will be observed, and after 48 hours' incubation, well-developed colonies of *Bact. abortus* will be noticed.

Huddleson makes the statement that CO₂ accelerates and favors the growth of *Bact. abortus*. We have been able to obtain the same results, using 10 per cent. hydrogen. It would therefore seem that the diminished oxygen tension rather than any specific effect of the CO₂, is involved. This last was suggested by Edwards in his review of the article by Huddleson.

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The effect of phenolsulphonephthalein upon the glomerular circulation in the frog.

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Further studies upon the function of the frog's glomeruli observed by the method of A. N. Richards¹ have shown that phenolsulphonephthalein is also excreted through the glomeruli. After

¹ Richards, A. N., *Am. J. Med. Sc.*, 1922, clxiii, 1.