GROWTH OF SPORE-FORMING ANAEROBES AT 50°C.

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The following observations were made during the course of some studies on anaerobes in sewages and sewage effluents. While the work was in progress a paper by Damon and Feirer (1925) on thermophilic anaerobes was published. These authors described four species which were isolated from horse manure and grew at 50° and 55°C. The authors were apparently unaware of a paper by Veillon (1922) in which he described three thermophilic anaerobes which developed both at 37° and 50°C. However, the organisms isolated by Damon and Feirer do not correspond with the descriptions of those studied by Veillon. None of these anaerobes appears to belong to the commonly known spore-forming types.

I have shown that the anaerobes most commonly found in sewages and effluents are *Cl. welchii* and *Cl. sporogenes* (Greer, 1925). On the sulphite-glucose-iron medium of Wilson and Blair (1924) these organisms produced black colonies when grown at 37°C. When the plates were incubated at 50°C. these same anaerobes appeared. The colonies were somewhat larger and developed more rapidly at the higher temperature.

Single cell strains of a number of spore-forming anaerobes were then studied. The organisms used were *Cl. aerofetidus*, *Cl. bifermentans*, *Cl. botulinum* A, *Cl. botulinum* B, *Cl. chauvei*, *Cl. edematiens*, *Cl. fallax*, *Cl. histolyticum*, *Cl. putrificum*, *Vibrion septique*, *Cl. sporogenes*, *Cl. tetani*, *Cl. tertium* and *Cl. welchii*. The cultures were made in beef heart medium, sealed with vaseline and incubated at 50°C. Observations of growth were made after four, eight and twelve hours and one, two, three, four and five days. All the above anaerobes except *Cl. histolyticum*

and *Cl. tetani* showed growth, at least by the end of the fifth day. These two organisms had not developed at the end of ten days. Duplicate cultures incubated at 37°C. all showed growth in five days. In many cases growth was evident at 50°C. as soon as at 37°C. *Cl. sporogenes, Cl. welchii, Cl. fallax* and *Cl. tertium* produced gas earlier at 50°C. than at 37°C.

Cultures of *Cl. welchii* and *Cl. sporogenes* were made in milk and coagulated egg medium, respectively, to determine whether or not these characteristic reactions occurred at the higher temperature. Both the stormy fermentation of milk by *Cl. welchii* and the liquefaction of coagulated egg white by *Cl. sporogenes* took place as rapidly at 50°C. as at 37°C.

An incubation temperature of 55°C. was also tried, using beef heart medium under vaseline seal. *Cl. botulinum* A, *Cl. botulinum* B, *Cl. bifermentans, Cl. chauvei, Cl. edematiens, Vibrion septique* and *Cl. sporogenes* developed at this temperature, but not as profusely as at 50°C. *Cl. welchii, Cl. tetani, Cl. tertium, Cl. putrificum* and *Cl. histolyticum* gave no visible growth in five days.

CONCLUSION

The anaerobes investigated with the exception of *Cl. histolyticum* and *Cl. tetani* appear to have a wide temperature range of growth, very little difference being observed between 37° and 50°C. In many cases growth was more rapid at the higher temperature. Some anaerobes will develop at 55°C., but the growth is somewhat retarded.

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