The Supposed Heat Stability of some Bacterial Enzymes: a Correction

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Oliver (1952) reported observations which he interpreted as indicating that decarboxylase systems of some lactic streptococci and some coliform organisms were relatively stable towards heat and chemical disinfectants. A recent check on the results has shown that the method used for estimating the carbon dioxide formed by a culture during a given period was faulty; the conclusions drawn as to the stability of the enzyme systems were therefore fallacious.

In the original experiments, cultures were grown in 100 ml. medium contained in a 150 ml. conical flask which was connected by a short length of 7 mm. diam. glass tubing to a second 150 ml. conical flask containing baryta. The apparatus, during incubation and during any heat treatment applied to the culture flask, was shaken only occasionally. In some experiments a vacuum was applied to the baryta flask in an effort to make sure that all the CO₂ formed in the culture was drawn into the baryta. In an examination of the method it became evident that under the conditions of Oliver's experiments carbon dioxide did not pass from the culture flask to the baryta flask as readily as had been assumed. A condition of supersaturation with respect to CO₂ often existed in the bacterial culture. Consequently the CO₂ formed by a living culture before the killing of the bacteria by heat or chemical disinfectants was only gradually evolved after the sterilizing procedure; hence the erroneous conclusion that the enzymes were heat stable.

The error of the method was made quite clear in a trial in which a solution of NaHCO₃ acidified with lactic acid to pH 3 was used in place of the bacterial culture in the apparatus. Under static conditions 25% of the theoretical amount of CO₂ passed over into the second flask in 24 hr. and was absorbed in the baryta; with constant gentle agitation 70% of the CO₂ was absorbed in 24 hr.; when a slow stream of CO₂-free air was bubbled through the solution, 100% of the CO₂ was absorbed in 1 hr.

Re-examination of the alleged stability of the decarboxylases using a technique in which the cultures were de-gassed by passage of CO₂-free air and the removed CO₂ absorbed in baryta in Pettenkofer tubes showed that the decarboxylase system of the lactic streptococci did not survive death of the bacteria. The enzyme systems of some coliform organisms did, however, show some evidence of heat stability, although much less than that claimed by Oliver.

REFERENCE

- OLIVER, W. H. (1952). The stability of some bacterial enzymes toward heat and chemical bactericides. *J. gen. Microbiol.* 7, 329.

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