

demonstrate that molecular hydrogen but not molecular nitrogen inhibits the splitting of  $H_2$  from pyruvate by *Clostridium pasteurianum*. The effect of  $H_2$  is similar to that observed by Kubowitz (Biochem. Z., **274**, 285, 1934) with *Clostridium butyricum* and probably merely reflects a shift in the equilibrium of the reaction. The findings were not affected by substituting argon for the helium. From the point of view of comparative biochemistry, it is of interest that *Rhodospirillum rubrum* differs from *Clostridium pasteurianum* not only in the absence of a "Kamen-Gest reaction" in the latter but also in the absence of an effect of  $H_2$  on the photoevolution of hydrogen by the photosynthetic anaerobe. The organisms are similar in that  $H_2$  does not appear to inhibit nitrogen fixation by either.

#### APPARENT FUSION OF THE CHROMATINIC BODIES IN SPECIES OF BACILLUS

WILLIAM A. CASSEL

Laboratory of Microbiology, Department of Botany, University of Pennsylvania, Philadelphia, Pennsylvania

Received for publication July 5, 1951

Kleineberger-Nobel (J. Hyg., **44**, 99, 1945) observed that when relatively young cultures of various species of the genus *Clostridium* were exposed to air, the chromatinic bodies soon changed from a dispersed to a "fused" pattern. It occurred to the author that this reaction might result from the formation of hydrogen peroxide, in these catalase-lacking organisms, on exposure to oxygen.

A study of this phenomenon has been made using *Bacillus cereus*, *Bacillus pumilus*, *Bacillus mesentericus*, and two strictly anaerobic strains of *Clostridium perfringens*. These organisms were cultivated at 37 C on brain heart infusion agar, the *Clostridium* cultures being incubated for 4 to 7 hours, and the *Bacillus* cultures for 3 $\frac{1}{4}$  hours.

Agar blocks, cut from the incubated plates, were treated with hydrogen peroxide in various concentrations. Following such treatment, the organisms on blocks of agar were fixed for 2 minutes in osmium tetroxide vapors. Smears of the fixed cells were placed in normal hydrochloric acid at 60 C for 10 minutes, rinsed, then stained 10 seconds with 0.1 per cent basic fuchsin. After rinsing and drying they were examined microscopically.

It was found that apparent fusion of the chromatinic bodies could be affected in all species of *Bacillus* studied by exposure for 10 minutes to hydrogen peroxide at very low concentrations. For example, a 3 $\frac{1}{4}$ -hour culture of *B. cereus* showed apparent fusion at a concentration of 0.0009 per cent hydrogen peroxide (see figure 1). Apparent fusion had occurred in the majority of cells on the slide, while control smears from the same culture showed the usual dispersed pattern found in young cultures.

Catalase, applied to the *Clostridium* cultures in various ways, was ineffective

in preventing apparent fusion in these organisms on exposure to air. This possibly results because the relatively large catalase molecule cannot reach the intracellular site of hydrogen peroxide formation.

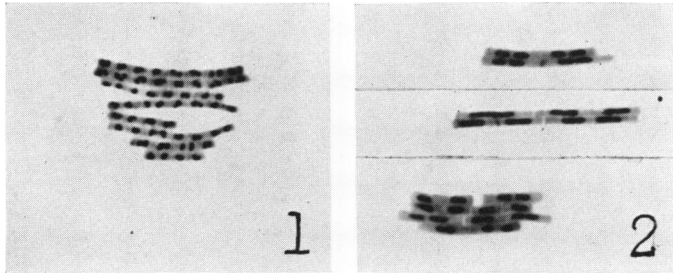


Figure 1. *Bacillus cereus*  $\times 1,400$ . Growth  $3\frac{1}{4}$  hours. Preparations stained as described in the text. No. 1. Cells not treated with hydrogen peroxide. No. 2. Cells from the same culture which have been exposed to 0.0009 per cent hydrogen peroxide for 10 minutes.

It is suggested that apparent fusion of the chromatinic bodies in species of *Clostridium*, on exposure to air, is a process induced by the formation of hydrogen peroxide.

## A STUDY OF THE WIDE SPREAD DISTRIBUTION OF CHROMOBACTERIUM SPECIES IN SOIL BY A SIMPLE TECHNIQUE

WILLIAM A. CORPE

*Department of Biology, Western Kentucky State College, Bowling Green, Kentucky*

Received for publication July 11, 1951

Purple pigmented bacteria tentatively identified as strains of *Chromobacterium* sp. were isolated from samples of soil by a simple technique.

Five gram samples of soil were placed in sterile petri dishes and soaked with sterile distilled water. Sterile polished or precooked rice grains were sprinkled over the surface of the soil and the plates incubated at room temperature (23 to 25 C) for 5 days. At the end of the incubation period, the rice grains were partially covered with a purple, membranous growth indicating the presence of *Chromobacterium* sp.

Sixty-eight out of 75 samples of soil collected in the vicinity of Bowling Green, Kentucky, central Michigan, northeastern Indiana, and northern Illinois showed the presence of *Chromobacterium* sp. by the described method.

Ordinary agar plating methods failed to reveal purple pigmented colonies, possibly because they are overgrown or inhibited by other soil microorganisms which occur in larger numbers.

An estimation of the numbers of *Chromobacterium* sp. in the soil samples was