

MICROBIAL BISCUITS OF VATERITE IN LAKE ISSYK-KUL (REPUBLIC OF KYRGYZSTAN)—DISCUSSION

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Giralt et al. (2001) reported the formation of microbial biscuits of vaterite in Lake Issyk-Kul (Republic of Kyrgyzstan). Although they state that the trigger mechanisms for the natural precipitation of vaterite remain unclear, they propose an explanation of the origin for the vaterite biscuits involving bacterial activity. Our recent research seems to support their hypothesis, although the way we obtain vaterite precipitation is not strictly related to lacustrine or natural aqueous environments.

We have used laboratory cultures to demonstrate the ability of some soil bacteria to transform an oxalate carbon source into carbonate, a hypothesis supported by observations (Cromack et al. 1977; Verrecchia et al. 1993) and biogeochemical studies (Verrecchia and Dumont 1996). The bacteria used were isolated by Tamer and Aragno (1980) and identified as *Xanthobacter autotrophicus* (DSM 432, ATCC 35674, syn.: *Corynebacterium autotrophicum*), previously described by Wiegel et al. (1978). The bacteria were grown in a B4 media (Boquet et al. 1973) to which calcium acetate or calcium citrate was added. In this experiment, the only way to produce carbonate ions involves bacterial metabolism using the wide range of carbon sources contained in the yeast extract, including acetate and citrate (Fig. 1). Moreover, *X. autotrophicus* is able to produce a lot of exopolysaccharide and polyglutamine (Wiegel 1991).

In our experiments, bacterial metabolism lead to the production of calcium carbonate (CaCO_3) crystals inside the medium, associated with the bacteria colonies. X-ray diffraction of the samples shows that the carbonate mineral forming these crystals is vaterite and not calcite. The shape of the crystals (Fig. 2A) is similar to those described by Giralt et al. (2001). A blank experiment was conducted with the same medium exposed to the same conditions of humidity and temperature, but the medium was not inoculated. No crystals were formed under these conditions. This emphasizes the significant role of bacteria in the precipitation of crystals. The lack of crystals in the blank experiments means the crystals associated with the bacterial colonies cannot be artifacts due to the desiccation of the medium.

In order to verify that the *X. autotrophicus* strain is related to vaterite production, another bacterium, *Alcaligenes eutrophus* (syn.: *Ralstonia eutropha*), was chosen to produce calcium carbonate under the same experimental conditions. *Alcaligenes eutrophus* does not produce any polyglutamine and much less polysaccharide than *X. autotrophicus* as observed in the culture. Using exactly the same media inoculated with *A. eutrophus*, calcite is precipitated instead of vaterite (Fig. 2B). The change of media (acetate-rich instead of citrate-rich media) did not affect the result: *X. au-*

trophicus and *A. eutrophus* produce crystals of vaterite and calcite respectively.

Giralt et al. (2001) explain the precipitation of vaterite by "local supersaturation in the microenvironment in which nucleation and growth of calcium carbonate occur, . . . the presence of polyaspartates in the mucilaginous mat of bacteria [favoring] the epitaxial precipitation of oriented vaterite fibers" (p. 433). Our experiments suggest a crucial link between the presence or absence of exopolysaccharides produced by the bacteria and the nature of the mineral precipitated, i.e., vaterite instead of calcite. Moreover, we have cultured bacteria in a mineral liquid medium containing oxalate in order to control the pH during bacterial oxidation of low-molecular-weight organic sources. The initial pH of the medium was $\text{pH} \approx 7$. After seven days and total consumption of the carbon source, the final pH was $\text{pH} \approx 9.5$ indicating stable conditions for calcium carbonate precipitation and preservation.

Although the general chemical conditions, crystal mineralogy, and morphology obtained in our laboratory experiments are in total agreement with Giralt et al.'s (2001) observations, it is still possible that the Lake Issyk-Kul vaterites might be a post-sampling artifact. Giralt et al.'s samples from dredged sediments of Lake Issyk-Kul were "stored in plastic bags for study" (p. 430). It is possible that the micro-scale vaterite crystals Giralt et al. described could have been produced by bacteria during storage. This is indirectly suggested by the authors when they noted that the microbial biscuits "could form very recently" (Giralt et al., p. 434). In addition, microbial biscuits are present only in surficial sediments. This can be explained by the fact that appropriate bacteria able to transform carbon sources into vaterite are present only in these favorable environments (aerobic strains).

In conclusion, although it is always possible to get artifacts with microbiological material, Giralt et al. (2001) throw light on a new possible source of secondary calcium carbonate, one involving transformation of vaterite into calcite. They bring new observations and arguments for a likely microbial mediation in the precipitation process of this CaCO_3 , indirectly emphasizing two main consequences: (1) many secondary calcite continental deposits could result from a vaterite source, a possibility that is widely underestimated because of the difficulty of detecting primary vaterite, which transforms into calcite very rapidly; and (2) the possibility for microorganisms to precipitate a mineral phase such as vaterite or calcite from organic carbon. This latter process undoubtedly constitutes a new potential carbon sink.

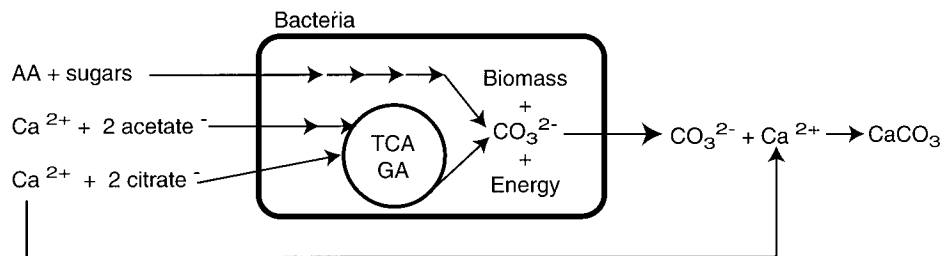


FIG. 1.—Possible metabolic pathways leading to carbonate precipitation in bacteria. AA, amino-acids; TCA, tricarboxylic acid cycle; GA, glyoxylic acid cycle.

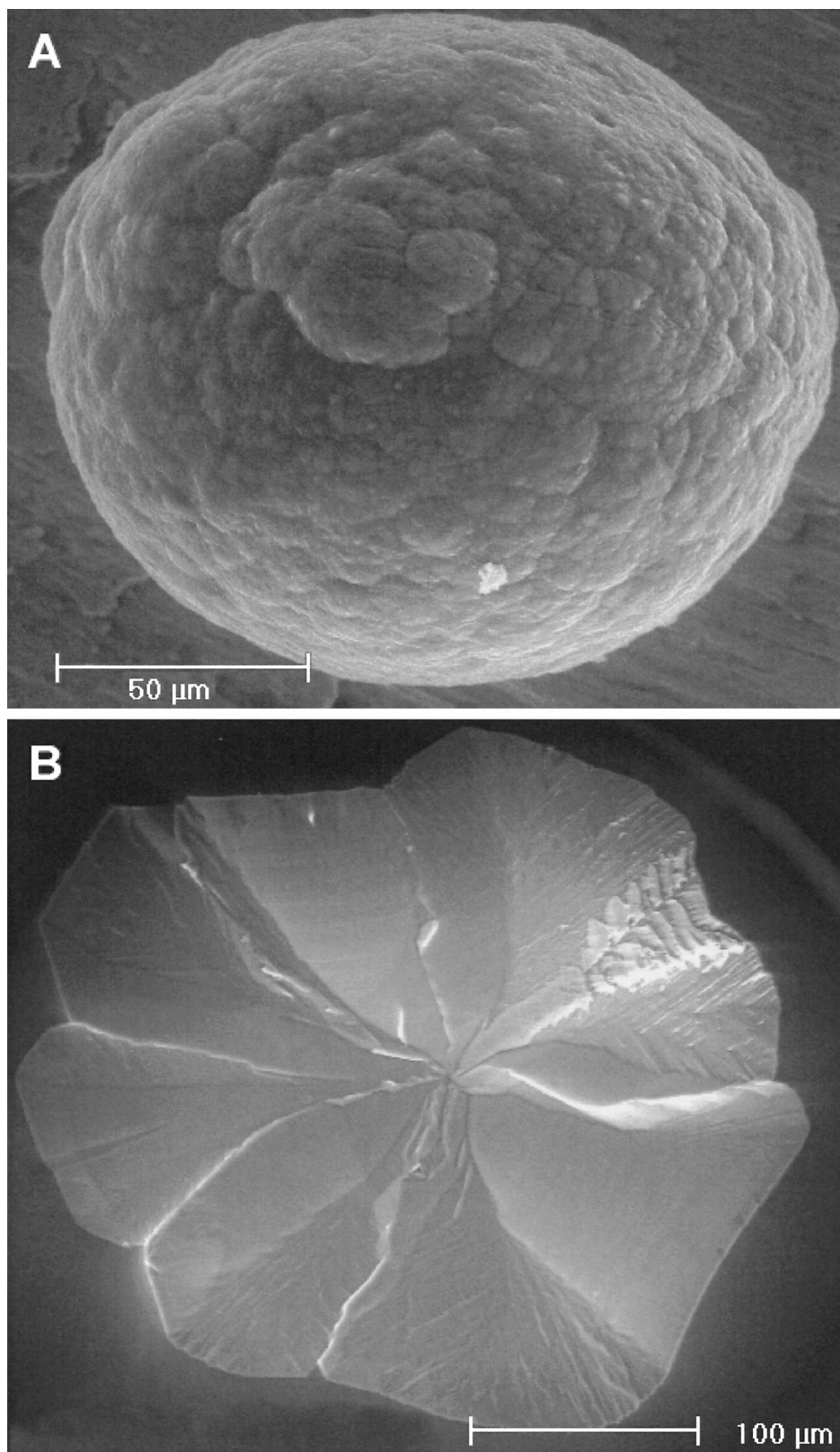


FIG. 2.—Scanning electron microscope photographs of **A**) vaterite crystal obtained from the *Xanthobacter autotrophicus* culture, and **B**) calcite crystal obtained from the *Alcaligenes eutrophus* culture.

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