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Identification of biomineralization processes and bacterial species forming the speleothems of Iron Curtain Cave, Chilliwack, British Columbia, Canada.

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Caves are extreme, often oligotrophic, environments that house diverse groups of microorganisms. Many of these microbes are able to perform microbiologically induced carbonate precipitation (MICP) to form crystalline secondary cave structures known as speleothems. The urease family is a group of enzymes involved in MICP that catalyze the breakdown of urea, which is a source of energy, into ammonia and carbonate. Carbonate anions efflux to the extracellular surface of the bacterium where it then binds to extracellular calcium to form calcium carbonate which then continues to grow in crystal form. We studied bacterial communities from the Iron Curtain Cave to determine whether urease-positive (U +ve) bacteria were present in the cave, the species present, and the degree to which they contribute to speleothem growth. Sediment in the cave is high in iron, and there are limestone structures throughout the cave. Iron Curtain is a wet and humid cave, containing six pools of water. The temperature varies seasonally from 4–12 °C depending. Ninety-nine bacterial strains were isolated from popcorn (PCS) and soda straw (SSS) speleothems. These isolates were screened for urease enzyme activity and eleven candidates were found to be U +ve. The selected U +ve candidates were then grown for 62 days on a modified B4 agar medium in conditions to mimic the cave environment; humidity = ≥90 % and temperature = 7-9 °C. U +ve candidates were also cultured for 84 days into liquid B4 medium to further study the minerals precipitated and potential metabolic pathways. After the incubation, species-specific crystal morphologies were observed. The U +ve candidates were all identified to the genus level by 16S rRNA analysis. The two best U+ve candidates are identified as *Pseudoarthrobacter* sp. PCS056 and*Sphingobacterium* sp. SSS035. The PSC056 and SSS035 consistently produce crystals in both agar and broth media. The results from this study are consistent with the involvement of the U +ve bacteria isolated from the ICC in the formation of the cave's speleothems. Genome sequencing and mineral chemistry are in progress, along with analysis of Fe as a catalyst.