snow albedo picks up and becomes nearly equal to the values at Mawson and Davis, both the models simulate almost equal upper ice temperature.

The seasonal variation of ice thickness is shown in Figure 2g-i. Semtner's model underestimates the ice thickness considerably at all the stations. This may be due to the fact that there is no provision for conversion of snow to ice or vice versa in Semtner's model, leading to inaccurate determination of snow thickness. The snow layer is less likely to affect the sensitivity of the Arctic sea ice to environmental changes but can affect the response of Antarctic sea ice cover to present or future climatic changes⁹. Also, the high oceanic heat flux in the region contributes to the melting of ice beneath the ice layer. Winton's model overestimates the ice thickness for Davis almost throughout the year, whereas it overestimates the ice thickness for Mawson only for first half of the year. For Casey, there is not much difference between the observed and simulated (from Winton's model) values. As the ice thickness reaches below 12.5 cm, Semtner's threelayer model switches itself to the zero-layer model using simple mass-budget equations. However, the zero-layer thermodynamic model does not allow ice to store heat like the three-layer model and hence it tends to exaggerate the seasonal variability in ice thickness⁶.

The monthly change in snow thickness is depicted in Figure 2j-l. Both the models overestimate snow thickness at Mawson, but underestimate it at Davis. The high snow albedo in the Mawson coast reflects back much of the incoming radiative flux, which prevents the snow layer from melting. At Casey, Semtner's model underestimates the snow thickness.

Both the Semtner and the Winton models are able to capture seasonal variability of Antarctic sea ice using only thermodynamics. However, thermodynamics as well as the dynamic processes play a significant role in determining the thickness of sea ice. Ice motion can lead to significant changes in sea ice thickness as well as transport of sea ice. Thus, there is need to investigate sea ice using ice–ocean coupled model that realistically simulates the feedback among the atmosphere, ice and ocean. Antarctic sea ice variability is likely to be controlled by both remote and local processes. Simulating the brine content in the upper ice layer, use of depth-dependent conductive coupling, and the provision of snow-to-ice conversion are advantages in Winton's model.

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Evidences for microbial involvement in the genesis of speleothem carbonates, Borra Caves, Visakhapatnam, India

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Speleothem carbonates are normally considered as inorganic precipitates, but recent work has demonstrated active biological influence in their formations. The present work focuses on the microfabric record and its relation to microbial involvement in the speleothems from Borra Caves, Visakhapatnam. Thin section petrography revealed the presence of lithified structures and micrite, occurring as laminated to clotted with chocolate-brown blebs and identical to microbialites observed in modern and ancient stromatolitic carbonates. SEM observations confirmed the presence of calcified bacteria, micro-rods, and needle calcite. Organic mats (yellow-orange in colour) comprise of mineralized filamentous bacteria, bacterial stalks, cells and sheaths. These microfabric evidences suggest that microorganisms have actively participated in the genesis of speleothem carbonates.

Keywords: Borra Caves, genesis, microbial involvement, speleothem carbonates.

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CAVES have always haunted the imagination of mankind since they host a wide spectrum of fascinating life forms, such as cave-dwelling spiders, leeches, mites, beetles, scorpions, fishes, snails and worms along with thick beds of bacteria and fungi that live on the rocks¹. Caves offer examples of possible past or present geomicrobiological interactions. A recent article in *Science* gave a new insight on cave biology¹. Several investigators are exploring the biogeochemistry, community characteristics and related metabolic activities of different novel microorganisms in caves and sulphur-fed systems^{2–4}.

Microorganisms are found to be associated with carbonates and speleothems⁵, and are able to precipitate carbonates in laboratory experiments⁶⁻¹⁰. Microbial carbonates are important in fluviatile, spring, cave and soil environments. The principal organisms involved are bacteria, particularly cyanobacteria, small algae and fungi, which form biofilms and/or microbial mats. Various metabolic processes, such as photosynthetic uptake of CO₂ and/or HCO₃ by cyanobacteria, and ammonification, denitrification and sulphate reduction by other bacteria, can increase alkalinity and stimulate carbonate precipitation. Extracellular polymeric substances widely produced by microbes are important in facilitating sediment trapping and binding¹¹, and providing nucleation sites for carbonate minerals. Significant research has been done on bacterially-mediated precipitation of carbonate minerals¹²⁻¹⁴, but little attention has been focused on microfabric features to show bacterial involvement in speleothem formation. The present study examines microfabric evidences of microbial influence in speleothem formations from Borra Caves, Visakhapatnam, Andhra Pradesh.

Visakhapatnam is located on the east coast of India (Figure 1)¹⁵. The city covers approximately 110 sq. km and sweeps towards the west with the seashore in the east. The Simhachalam Hills and Dolphin Nose (Yarada Hills) flank it on the north and south respectively.

The geology of the region is represented by the Khondalite suite of rocks (garnetiferrous sillimanite gneisses, quartzo-feldsphatic garnet gneisses) of Archaen age in the Eastern Ghats mobile belt. Quaternary deposits consist of red bed sediments, laterites, pediment fans, colluvium, alluvium and coastal sands^{16,17}. All these rocks lie within calc-silicate granulites and garnet-sillimanite gneisses (khondalites) which have been repeatedly folded and metamorphosed at high grades and form a part of the Eastern Ghats granulite belt.

The Borra Caves are situated in the Araku Valley, approximately 90–95 km from Visakhapatnam (18°15'N; 83°3'E). The word 'Borra' means something that has bored into the ground. These are the second largest caves in the Indian subcontinent. The Borra Caves lie in the reserved forest area and consist of 14 villages inhabited by tribals in the Ananthagiri Mandal of Visakhapatnam district. The entire area around Borra Caves is thickly forested. It is also one of the important hill regions of the Eastern

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Ghats and is known not only for the diversity of its flora and fauna, but also for the richness of its minerals. These caves were discovered by William King George of the Geological Survey of India in 1807, and the caves have a religious and historical importance. The annual temperature of Araku Hills is approximately 25°C and annual rainfall is 950 mm (mostly coming from the northeast monsoon). The caves have an archaeological importance due to the discovery of some Palaeolithic implements. These caves host a variety of speleothems ranging from very small to big and irregularly shaped stalactites and stalagmites. The carbonate rock at Borra is pure white, and coarsely crystalline. At Borra, deformed and banded marbles extend over a triangular area of 2 sq. km. They are surrounded by diopside-scapolite-feldspar calc-granulites. The pyroxenite outcrops at Borra are dark and massive and include discontinuous calc-silicate bands, some of brown mica and others with calcite¹⁵. Between the solidified stalactites and stalagmites is the Gosthani River which flows out of the caves. Inside the cave there is a twilight zone with limited light penetration. The deep cave is totally aphotic. The stalactites ranged in lengths from approximately 0.1 to 3.5 m. The stalagmites were around 1.2 m long, and columns were 6 m long and 0.75 m wide (Figure 2a-c). The length of the cave is approximately 2 km; it is 12 m high and 1300 m asl. The bedrock is mainly limestone. The average temperature of the inner cave wall was approximately 16°C. Deep inside



Figure 1. Geological map of the Borra area. Blank areas are quartzo-feldspathic gneisses. RS, Railway station.



Figure 2. a, Stalactite; b, Stalagmite formation; c, Stalactites; d, Orange organic mat on spring.

are sulphidic springs discharged into the cave passages causing corrosion of limestone. Mucus-like biofilms which are thick orange microbial mats (2.5-3 cm thick) with patches of yellow biofilms extending 3 m from the aphotic deep cave orifice were seen floating on the spring waters (Figure 2 *d*). The fauna in the caves were predominantly bats and bat guano deposits were observed. The flora consisted of mosses and brown-to-green algae.

Stalactite, stalagmite and column samples were collected in sterile ziplock bags. Thin sections were prepared for petrological observations. Sample powders for XRD were scanned between 4 and 64° 2θ at 1° 2θ /min on X'Pert Pan X-ray diffractometer. The minerals were identified using the JCPDS database¹⁸. Scanning electron microscope (SEM) studies were conducted on freshly broken surfaces of stalactite and stalagmite samples using a Philips PSEM 515. Representative chip samples were mounted on stubs and coated with gold in an evaporative coater and then transferred to the sample chamber of the instrument prior to imaging.

Thin-section observations show that carbonates occur as alternate white and dark laminated bands and voids at different places (Figure 3 a-c), as well as the presence of lithified structures, internal fabrics similar to microbialites with organic inclusions (Figure 3 a-d) and an enormously large number of micro-rod calcites (Figure 4a and b). Microcrystalline calcites are distinct in white bands. Dark bands seem to contain mucus material. Elsewhere, submicron laminations are visible (Figure 3c). There is a fine scale alternation of micrite and spar calcite similar to the lamination observed in stromatolites (Figure 3 c)^{11,12}. The micrite varies as laminated to clotted with chocolatebrown blebs (Figure 3d). Dense dark micritic layers (Figure 3 *a*) may have formed during microbial activity and the spar layers formed when there was no microbial activity³. Clotted peloidal fabrics (Figure 3 d) were observed and these are also common in stromatolites, thrombolites, travertines, and in reefs. The presence of enormously large number of micro-rod calcites, organic inclusions and lithified structures that resemble stromatolitic microbial mats may indicate that they are produced by microbial activity³. Micritic laminae are often considered as fossil microbial mats¹⁹. Micrite, although previously considered as inorganic cement, is now interpreted as calci-



Figure 3. *a*, Alternating layers of micrite and spar; *b*, Micrite deposition in chains; *c*, Micrite depositions as seen in stromatolites; *d*, Micrite seen as clotted chocolate-brown blebs.

fied bacterial aggregate^{11,20}. The observed micritic fabric is similar to that of bacterial origin reported from marine environments²¹.

SEM studies indicate that calcite with a rhombic morphology was dominant (Figure 4 c). However, minor amounts of dolomite and aragonite crystals were also observed in X-ray diffractograms. Various mineralized organic filaments were observed. Calcified filaments, micro-rods (Figure 4 a and b) and needle calcite (Figure 4 a, d and e) were enormous and these may have been precipitated by bacterial metabolic activities.

Micro-rod calcite is classified based on its morphology as given below: the filaments (in length) ranging up to 10 μ m (Figure 4 *a*) as micro-rods, 10–50 μ m (Figure 4 *a* and *b*) as calcified filamentous bacteria and needle calcite from 200 to 300 μ m (Figure 4 *a* and *e*)^{22,23}. Its origin is attributed to rapid precipitation at high super-saturation states during evaporation in soils or to calcification of bacilliform bacteria²³. Also observed were curved, filamentous structures (Figure 4 *b*). Organic mats covered the calcite crystals and mats were found intertwined with crystals suggestive of microbial involvement (Figure 4 *f*) and *g*). The microfabrics and fossil bacteria, similar to those observed in marine environment and cave speleothems elsewhere, suggest possible microbial involvement in speleothem precipitations. Also, the occurrence of needle and fibrous calcite suggests microbial involvement^{2,21,24}.

Thick orange, filamentous microbial mats (approximately 2.5–3 cm thick; Figure 2 d) with patches of yellow biofilms extending 3 m from the aphotic deep cave orifice were floating on the waters in the stream coming from spring outlets (Figure 2 d). SEM examination of the mats showed the occurrence of filamentous bacteria, bacterial stalks, cells and sheaths. These microbial mats appeared to have a distinct microbial community (Figure 4 f and g).

Microbial carbonate microfabrics are heterogeneous. Grain-trapping is locally important, but the key process is precipitation, producing accumulation of calcified microbes and enhancing mat accretion. It is important to determine the conditions under which speleothems form by studying the fabrics preserved in the cave precipitates/deposits. The criteria for recognizing microbes and microbial activity include documentation and recognition of mineralized microbes, recognition of stromatolites that are laminated



Figure 4. a, Calcified filaments, micro-rods; b, Curved calcified filaments; c, Rhombic crystal calcite; d, Needle-shaped calcite, filaments; e, Micro-rods needle-shaped calcite; f, Orange organic mat with bacterial filaments, sheaths, stalks; g, Bacterial filaments, cells, sheaths, stalks.

structures formed by microbes that trap and bind detrital grains to a substrate or act as nucleation sites for mineral precipitation and identification of fabrics/textures that are known to be indicative of microbial activity²⁵.

The observed stromatolitic structures and mineralization of these structures is facilitated by the metabolic processes of microorganisms. The filament surface acted as a substrate for calcite mineralization. These results indicate that microbes and calcified microbes in the structures/ textures preserved in the speleothems are indicative of microbial activity. The microbial influence cannot be ruled out in the precipitation process in the Borra Caves. Further *in situ* laboratory-based culture experiments are ongoing and will confirm the extent of microbial involvement in the speleothem formations.

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Are nitrate concentrations in leafy vegetables within safe limits?

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Leafy vegetables are an important source of nutrition in the human diet. Estimation of nitrate concentration in samples of leafy vegetables collected from the local markets of Delhi has revealed that a significant number of spinach and chenopodium samples contained nitrate in concentrations higher than the Acceptable Daily Intake (ADI) limit for an average 60 kg person (if consumed @ 100 g/day). However, nitrate concentration in fenugreek, coriander and sowa samples was well

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