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Live (Rose-bengal stained) foraminifera from deep-sea anoxic salt brine in the Eastern Mediterranean: toward understanding limit of life for single-celled eukaryotes (foraminifera)

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What is a limit of life for the eukaryotes? Eukaryotes are thought to adapt and evolve under oxic environmental conditions. Recently, there are many exceptions for this hypothesis, as many eukaryotes including metazoan groups are found in anoxic environmental conditions.
We found many rose-bengal stained foraminifera from a deep-hypersaline anoxic basin (DHAB) in the eastern Mediterranean. During KH06-04 cruise, we conducted oceanographic research at Medée Lake, the largest DHAB, that is located 100km southwest of Crete Island in the eastern Mediterranean. The lake situates at 2920m in water depth. Depth of saline water is 120m in maximum. Both water and sediment samplings were carried out both with Niskin bottles and multiple corer attached to camera watching sampling system at three sites, inside of the lake (CS), the edge of the lake (OMS) and the normal deep-sea floor (RS). Temperature, salinity, and dissolved oxygen concentrations at central saline lake are 15.27 oC, 328PSU, and 0.0 ml/L, respectively. Strong smell of hydrogen sulfide was detected from the lake sediment.
Subsamples were conducted for multiple core samples using 3 subcores(φ 2.9cm) from each core tube (φ 8.2cm). Sediment samples were fixed with 4% formalin Rose Bengal solution on board. In laboratory, samples were washed with 32μm sieve. Rose Bengal stained specimens were picked under binocular stereomicroscope (Zeiss Stemi SV11) for surface 0.5cm layer, and identified with inverted microscope (Nikon ECLIPSE TE300).
In total, 26 species belonging to 9 genera were identified from three sites. Six species belonging to two genera were identified in the center of the salt brine. Only a few species are common among three sites, even though the numbers of common species were 10 between OMS and RS sites. In DHAB, spherical organic-walled species, such as allogromiid and psammosphaerid, are dominant. In contrast, tube-like chitinous foraminifera, such as Resigella, Conicotheca and Nodellum, are common at oxic deep-sea floor site (RS). Calcareous species, Eilohedra cf. tumidula, were found both inside and outside of DHAB with almost same frequencies.
Recently, some deep-infaunal foraminifers are known to take part in nitrogen cycles under dysoxic environments. Virgulinella fragilis is the species that is adapted to euxinic environments with occasional oxygen supply. In contrast to these foraminifers, foraminifers in salt brines are thought to be obligatory anoxic foraminifera. Both TEM observations and DNA analyses will be held for understanding adaptive strategy of foraminifera that are dwelled in anoxic environments.