

Microbialites in chemosynthetic ecosystems

*Tomoyo Okumura¹

1. Japan Agency for Marine-Earth Science & Technology

Microbialites are macroscopically identifiable the earliest evidence of life, which are formed with association of benthic microbial communities and some environmental factors (1). Most microbialites consist of carbonate, but also consist of silica, phosphate, iron- and manganese-oxides, iron sulfides (2), and magnesium hydroxide (3). Previous investigations for microbialites in terrestrial (rivers, lakes, and hot springs) and shallow marine settings revealed that photosynthetic microbes such as cyanobacteria and algae play an important role for microbialite formation (2). However, microbialites are also occurred in some chemosynthetic ecosystems at subsurface and deep-sea settings. Due to the sample availabilities, reports and descriptions of such unusual microbialites were limited so far. Here, I summarize the studies on microbialites at the dark environments, and introduce characteristics on a microbialite that have newly found on 2013 at the Shinkai Seep Field, Southern Mariana.

Some carbonates at deep-sea methane seep sites (4, 5) and in pipeline at the landfill sites (6, 7) contained microbialite textures. Carbon isotope ratio of the carbonates and biomarkers indicated the association of methanogen and/or methane-oxidizer for making microbialite textures. Some iron sulfide deposits near deep-sea hydrothermal vents also exhibited microbialite textures, which have been interpreted as organosedimentary structures associated with various chemosynthetic microbes. A newly discovered chimney at the Shinkai Seep Field consisted of magnesium hydroxide and calcium carbonates, which were interpreted as a product of serpentinite-associated alkaline fluid.

Microbialite textures were locally found in the chimney and only consisted of magnesium hydroxide. Scales of microbialites at the chemosynthetic ecosystems were ranged from several mm to cm. In addition, some of them occurred with bivalves and tubeworms. On the other hand, microbialites at the photosynthetic ecosystems occur larger scales at a maximum of several km (2). Further studies for understanding microbialites at the chemosynthetic ecosystems will provide useful information for reconstruction on early life from ancient microbialites.

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Keywords: microbialite, chemosynthetic ecosystems

Decomposition of the plant in the ocean of the Late Cretaceous

-Focused on the Upper Cretaceous Yezo Group distributed in Nakagawa Town, Hokkaido, Japan-

*Yuma Yamamoto¹, Robert Jenkins²

1.Division of Environmental Science and Engineering, Graduate School of Natural Science and Technology, Kanazawa University, 2.School of Natural System, College of Science and Engineering, Kanazawa University

Land plants, mainly composed of cellulose and lignin, are an important carbon reservoir on the earth. However, those organic compounds are relatively difficult to decompose for most organisms. In modern marine environments, wood-boring bivalves, which have symbiotic microbes to decompose cellulose, can mechanically break up the wood by their shells and consume cellulose. Then, decomposed organic compounds are useful for benthic organisms which have no ability to decompose original wooden material. Based on such degraded organic matter and other chemical compounds produced during decay of woods, sunken-wood communities will be established. Thus, it is important to know role of wood-boring bivalves through the ages. The wood-boring bivalves have appeared in the Early Jurassic and diversified in late Cretaceous. However, degradation process of the wood by boring bivalves in the Cretaceous ocean hasn't been fully revealed. Thus, the current study aim to reveal the degradation process of the wood in the ocean using the fossils from the Cretaceous Yezo Group distributed in Nakagawa Town, Hokkaido, Japan.

Totally 67 carbonate concretions were collected from the localities where Cretaceous deposits were exposed. Surface of the observation, cut polished surface and thin section of the observation, X-ray CT imaging, such as the cleaning of containing invertebrate fossils were carried out in the laboratory. Within the sample, ca. 70 % of concretions contained wooden trunk fossils. Within the concretions contained wooden fossils, about 34 % were bored by wood-boring bivalves. Detailed observations of borings suggest that the all bore holes were made by xylophagous wood-boring bivalves which are known as deep-sea wood borers. Within the bore holes, aggregations of framboidal pyrites, indicating activity of sulfate reducing bacteria were frequently found, and pyrites accumulated within the wood and around the wooden fossils. Fecal pellets, sometimes contain wood fragments, were also accumulated around the woods. It indicates that it was revealed that at least about 30% of the wood is degraded by deep-sea wood-boring bivalves and sulfate reducing bacteria in the deep sea zone of Yezo basin of the Cretaceous.

Keywords: Wood-boring bivalves, Sulfate reducing bacteria, Cretaceous

Influence of cold seep methane on the forming of echinoderm skeletons

*Moe Kato¹, Tatsuo Oji², Kotaro Shirai³, Tatsuya Unuma⁴

1. Graduate School of Environmental Studies, Nagoya University, 2. Nagoya University Museum, 3. Atmosphere and Ocean Research Institute, The University of Tokyo, 4. Hokkaido National Fisheries Research Institute, Fisheries Research Agency

Recently, a few modern species of echinoderms have been reported as a member of the chemosynthetic community organisms, and at least two fossil species of echinoderms have been found from cold seep deposits (Pawson and Vance, 2004; Gaillard *et al.*, 2011; Landman *et al.*, 2012). In order to understand how these echinoderms from cold seep environment take carbon to form their skeletons, two analyses were done; 1) stable carbon isotope analysis of the fossil skeletons, and 2) stable carbon isotope analysis of a living sea urchins in aquaria under environments of controlled stable carbon isotope ratios ($\delta^{13}\text{C}$).

The fossil echinoderms have been collected and studied from two cold seep areas; the Upper Cretaceous (Campanian) in South Dakota, USA and the coeval formation in Hokkaido, Japan. The both crinoids from South Dakota and Hokkaido have clearly lower $\delta^{13}\text{C}$ values than normal echinoderms, suggesting their skeletons were influenced by cold seep methane which has extremely low $\delta^{13}\text{C}$ values (Kato and Oji, 2015). However, it was still unknown how these crinoids formed their skeletons with such low $\delta^{13}\text{C}$ values.

A lot of marine organisms such as many mollusks, are considered to precipitate their skeletons in equilibrium with the ambient sea water in regards to their carbon isotope (Epstein *et al.*, 1951). For example, bivalves shells from a chemosynthetic community show $\pm 5\%$ $\delta^{13}\text{C}$ values (e.g. Mae *et al.*, 2007). However, echinoderm skeletons, even from a normal (non-seep) environment, are known to be formed not in isotopic equilibrium, and the $\delta^{13}\text{C}$ values of the echinoderms skeletons are different depending on the classes or species (e.g. Weber, 1968). Thus, in echinoderm biomineralization, an isotope fractionation due to vital effect or some carbon sources other than from sea water should be possible.

In order to clarify the cause of inequilibrium of the $\delta^{13}\text{C}$ value in the skeletons of echinoderms and to estimate degree of isotope fractionation in forming skeletons, specimens of modern echinoid (*Strongylocentrotus intermedius*) have been raised in two experiments; 1) the echinoids has been raised under three types of sea water with differently controlled $\delta^{13}\text{C}$ value of DIC (dissolved inorganic carbon), and 2) with two different types diets (a kelp (*Saccharina longissima*) and a land plant (*Fallopia sachalinensis*)) with different $\delta^{13}\text{C}$ value.

Compared with the controlled case of culture with normal sea water with seaweed diet, $\delta^{13}\text{C}$ value of the echinoid test changed in both cases (in waters with high $\delta^{13}\text{C}$ value, and with different diets). Thus, it suggests that skeletons $\delta^{13}\text{C}$ of this echinoid, are influenced by isotope ratio of both sea water and foods. This result clearly shows that the process of isotope fractionation and forming skeletons of echinoderms should be different from those of most of mollusks. It is thought that the echinoderms in cold seep environment depend on bacteria mat for their diet, or detritus that have low $\delta^{13}\text{C}$.

Keywords: cold seep, echinoderms, chemosynthetic community, biomineralization

Mass occurrence of the enigmatic gastropod *Elmira* in the Late Cretaceous Sada Limestone seep deposit in southwestern Shikoku, Japa

*Takami Nobuhara¹, Daigaku Onda¹, Takuya Sato¹, Hidemi Aosawa¹, Toyoho Ishimura², Akira Ijiri³, Urumu Tsunogai⁴, Naoki Kikuchi⁵, Yasuo Kondo⁶, Kiel Steffen⁷

1.Science Education (Geology), Faculty of Education, Shizuoka University, 2.Department of Chemistry and Material Engineering, National Institute of Technology, Ibaraki College, 3.Kochi Institute for Cores Sample Research, JAMSTEC, 4.Graduate School of Environmental Studies, Nagoya University, 5.Museum of Nature and Human Activities, Hyogo, 6.Sciences Unit, Natural Sciences Cluster, Kochi University, 7.Department of Palaeobiology, Naturhistoriska riksmuseet, Stockholm

Elmira is a medium- to large-sized gastropod genus, which has so far been recorded only from a presumably Eocene methane-seep deposit in Cuba, and its systematic affinity and paleoecology are unknown. We report a mass occurrence of *Elmira* sp. from a Late Cretaceous seep deposit in Shikoku, Japan, called Sada Limestone, with its mode of fossil occurrence, carbonate petrology, and stable carbon isotope analyses. Sada Limestone is characterized by the dominant occurrence of a large-sized thyasirid bivalve "*Thyasira*" *hataii* and serpulid worm tubes. The mass occurrence of *Elmira* sp. occurs as a lens-shaped carbonate body, 6.5 m in length and less than 2 m in thickness, intercalated in the thyasirids-rich limestone. The *Elmira*-rich lens body has a flat top and a concave base, and consists of multiple shell accumulation layers, which were formed by winnowing and filling of a depression in slope mud. The rare occurrence of *Elmira* sp. elsewhere in the Sada Limestone suggests that it lived in local aggregations in the vicinity of the depression. The matrix of the *Elmira* mass occurrence is rich in dolomite and ankerite and is less depleted in ¹³C (δ¹³C values of calcite: -5.3 to -2.4‰; of dolomite: -8.3‰) than the calcitic matrix of the surrounding limestones. This suggests that the *Elmira* mass occurrence was cemented below the sulfate reduction zone and thus with little influence of anaerobic methane oxidation. It is, therefore, difficult to consider that *Elmira* sp. harbor chemosymbiotic bacteria. As some trochiform gastropods do in seep sites, *Elmira* sp. was maybe a bacteria grazer gregarious on bacteria mats and/or hard bottoms.

Keywords: chemosynthesis, cold seep, Gastropods, Cretaceous, Shikoku

Meiofaunal community compositions around hydrothermal vents in three seamounts in NW Pacific -Vent copepods want to stay with vent polychaetes?

*Motohiro Shimanaga^{1,2}, Reina Senokuchi¹, Yuki Uejima¹, Hidetaka Nomaki², Hiromi Kayama WATANABE², Tomo Kitahashi²

1. Kumamoto University, 2. Japan Agency for Marine-Earth Science and Technology

In contrast to specific large benthic invertebrates in chemosynthetic ecosystems around deep-sea hydrothermal vents, meiofaunal communities in such habitats have only recently been investigated. This is especially true in the Northwest Pacific Ocean, even though there are many seamounts with active hydrothermal vents in their calderas. We studied the variations in meiofaunal composition around hydrothermal vents on chimney structures (vent chimneys) and in the adjacent non-vent fields in the calderas of three neighboring seamounts (Bayonnaise Knoll, Myojin Knoll, and Myojin-sho Caldera), in Izu-Ogasawara Arc, NW Pacific. A typical meiofaunal composition (nematodes as the most abundant taxon; harpacticoid copepods as the second) was observed in the sediments on sea-floor in the non-vent (control) fields, and even in the sediment at the base of chimneys. On the surfaces of vent chimneys, on the other hand, copepods and their nauplii were the most abundant. There were significant differences in copepod compositions on vent chimneys between the three seamounts, and even between different substrates on the same chimneys. *Stygiopontius* (Dirivultidae, Siphonostomatoida), a typical vent copepod group, was abundant and often predominant on the dense mats of tubes of *Paralvinella* polychaetes living in the closest proximity to vent fluids among vent metazoan macrofauna. These copepods, however, showed the lower density on the beds of *Neoverruca* barnacles although their beds were adjacent to *Paralvinella* mats at cm scales. No significant genetic difference was observed among local populations of *Stygiopontius*, suggesting they are the same species. Furthermore, stable carbon and nitrogen stable isotopic ratios and radiocarbon abundances in meiofaunal soft tissues revealed the dirivultids utilized mainly chemolithoautotrophic microbes at vents. These results show that *Stygiopontius* copepods prefer *Paralvinella* mats as their habitats or have similar environmental preferences (such as higher water temperature or existence of chemolithoautotrophic microbes) to *Paralvinella*.

Keywords: hydrothermal vent, meiofauna, community compositions

Nutrition sources of meiofauna revealed by stable carbon and nitrogen isotope ratios and natural radiocarbon abundances explain meiofaunal distribution patterns at hydrothermal vent fields

*Hidetaka Nomaki¹, Yuki Uejima², Nanako O Ogawa¹, Masako Yamane¹, Hiromi Kayama WATANABE¹, Reina Senokuchi², Tomo Kitahashi¹, Yusuke Yokoyama^{3,1}, Naohiko Ohkouchi¹, Motohiro Shimanaga^{2,1}

1.Japan Agency for Marine-Earth Science and Technology, 2.Kumamoto University, 3.The University of Tokyo

Deep-sea hydrothermal vents host unique marine ecosystems which mainly rely on organic matters produced by chemolithoautotrophic microbes. Although there are abundant meiofauna at the hydrothermal vent field, studies on their distributional patterns and nutritional sources are still limited due to their small body sizes. In this study, we investigated dietary sources of meiofauna at hydrothermal vent fields in the western Pacific using stable carbon and nitrogen isotope ratios ($\delta^{13}\text{C}$, $\delta^{15}\text{N}$) and natural radiocarbon abundances (D^{14}C). Based on these data, we evaluated how much do the hydrothermal vent meiofauna gain their nutrition from the chemolithoautotrophic microbes and how does it related to their distributional patterns around hydrothermal vent fields. Bacterial mats of the hydrothermal vent chimney typically exhibited heavy $\delta^{13}\text{C}$ values (up to -10‰) and depleted D^{14}C values (~600‰). The $\delta^{13}\text{C}$ and D^{14}C values of Dirivultidae, an endemic copepod family inhabiting hydrothermal vent chimney, exhibited similar values to the bacterial mat but distinct from those of sediments at surrounding area or water column plankton, suggesting that they exclusively rely on bacterial mat at the vent chimney. Other copepods at the vent chimneys may also rely on bacterial mat to some extent. To the contrary, $\delta^{13}\text{C}$ values of nematodes at vent chimneys were -26.6 and 23.2‰, which were similar ranges to those at non-vent sites, suggesting vent nematodes did not gain their nutrition from the chemolithoautotrophic microbes. Those nutritional facts obtained from isotopic compositions explain well about distributional patterns of these meiofauna; while dirivultid copepods exclusively distribute at vent chimneys and other copepods are more abundant at vent chimneys than in non-vent sediments, nematodes showed similar abundances between vent chimney and non-vent sediments.

Keywords: Hydrothermal vent ecosystem, meiofauna, feeding habit, stable isotope ratios, natural radiocarbon abundance