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Biological response on change of hydrothermal fluid flow: quantification of bacterial endosymbionts in *Bathymodiolus puteoserpentis* using 3D image analyses

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Bacterial endosymbionts of Mid-Atlantic Ridge hydrothermal vent mussels live in dual symbiosis with sulphide- and methane oxidizing bacteria. A decrease of electron donor availability in the hydrothermal fluids can lead to significantly reduced bacterial turnover and to declining symbiont abundances. The flexibility of a symbiosis towards changes of the geochemical environment is critical for the production, growth and even survival of the symbiotic host populations.

Quantitative analyses of symbiont abundances can give information on the symbiont activity patterns. Vice versa, comparative quantification of symbionts in hosts collected at different locations can give information on the specific hydrothermal activity. In an initial approach, we investigated how flexible the *B. puteoserpentis* symbiosis is towards sudden cessation of electron donor supply by analyzing bacterial abundances in host specimens that had been displaced a few days earlier from active diffuse fluid flow to a hydrothermally uninfluenced site. For the quantification of the symbionts in the host gills, we combined fluorescence in-situ hybridization (FISH) with confocal laser scanning microscopy (CLSM). The resultant 3D images were analyzed with digital image analyses and deconvolution software for three dimensional distribution patterns and abundances of the symbiotic cells. Initial results showed a significant decrease after 10 days duration of the experiment. By using specific FISH probes, we are able to determine specific responses for the different symbionts. We aim to develop this method for comparisons among hydrothermal habitats. The quantitative data will

be verified in the future by other molecular tools such as quantitative PCR.