Geophysical Research Abstracts,
Vol. 10, EGU2008-A-08839, 2008
SRef-ID: 1607-7962/gra/EGU2008-A-08839
EGU General Assembly 2008

The isopod cuticle: a model to study formation and function of amorphous calcium carbonate in calcified tissues

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The mineralized exoskeleton (cuticle) of crustaceans is an excellent model to study mineralization processes of calcified tissues. The cuticles of all isopods have a similar structural principle: An organic matrix composed of chitin-protein fibers associated with various amounts of crystalline and amorphous calcium carbonate. The cuticle is subjected to frequent molting in which it is periodically decalcified and shed. A new larger cuticle, synthesized before shedding, is mineralized after every molt. These processes cause spatial and temporal changes of the mineral distribution. The occurrence of ACC within the cuticle is of particular interest. ACC is the least stable form of the known phases of calcium carbonate. It is thought to be a precursor phase for crystalline modifications and, because of its high solubility, serves as transient calcium carbonate reservoir. In order to better understand the formation and function of ACC, changes in the distribution and content of mineral phases within the cuticle of the land living crustacean *Porcellio scaber* (Isopoda) were monitored during different stages of the moulting cycle and after in-vitro decalcification. Micro-Raman spectroscopy shows that calcite is restricted to the outer area of the cuticle, whereas amorphous calcium carbonate (ACC) is localized in the middle, having only little overlap with the calcite layer. During moulting the protective outer calcite layer is shed away during each molt, whereas ACC is recycled to quickly re-establish the protective calcite layer in the new cuticle. Additionally detected magnesium and phosphate derivates suggests

that they may stabilize the ACC.

Supported by the DFG within the research program “Principles of Biomineralisation” (SPP 1117)