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Phage Display Antibody Against Gill Proteins of the Tropical Clam

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Introduction: Codakia orbicularis (Bivalvia, Lucinidae) is a tropical clam, which lives in areas of high sulfur concentrations and which contains endosymbiotic chemoautotrophic bacteria in its gills. Gill proteins are thought to be involved in sulfur metabolism and in symbiont/host interactions. We used the Recombinant Phage display Antibody System (RPAS) to raise Single chain Fragment variable (ScFv) antibodies against these proteins. Material and Methods: Griffin.1 library of phage display antibodies was used. Three rounds of selection of phage panning were performed on immunotubes coated with total extract of native gill proteins. The bound M13 phages were isolated and tested by ELISA and the positive clones were selected for ScFv production. The presence of ScFv DNA inserts was assessed by PCR. These chimeric antibodies were purified by ammonium sulfate precipitation or IMAC, and used in Dot-blot and Western-blot assays as well as in plate antigen capture combined with SDS-PAGE. Results and Discussion: Five out of 60 isolated clones were used for ScFv production. Purification on IMAC gave ScFv antibodies of about 30 000 Daltons at a higher yield compared to ammonium sulfate precipitation. Antigen capture showed a strong positive reaction to a 15 000 Dalton protein, a major band in the electrophoretic profile of gill proteins. These recombinant antibodies are being tested by immunohistochemistry on gill sections. Total gill extract has been used for the first time to raise phage antibodies against clam proteins, which antibodies might be useful in the isolation and study of the properties of the said proteins.