**PO 4.15**

# Quantitative determination of Cd-Metallothioneins in prawn samples by HPLC-ICPMS

G. Raber, K.A. Francesconi, R. Raml, W. Goessler ,

Karl-Franzens University Graz, Institute of Chemistry, Analytical Chemistry,
Universitätsplatz 1, 8010 Graz, Austria,
georg.raber@uni-graz.at

Metallothioneins (MT) are ubiquitous, cysteine-rich proteins that have been ascribed various biological roles including involvement in metal detoxification processes. Earlier aquarium experiments with crabs exposed to cadmium showed that they produce a particular and unusual isoform at high exposure, and in a follow-up field study, this same MT isoform was identified as one of at least five MTs in the muscle of female coral prawns collected from a site naturally high in cadmium. These data suggested that the MT sub-isoforms in coral prawn may be formed in response to high Cd exposure, and hence it could be a selective biomarker of excessive, and toxic, Cd levels. The data on the prawn abdominal muscle so far show that several Cd-MT isoforms are present, and that the pattern of these MTs is related to the total concentration of cadmium.

We report studies on the development of an HPLC-ICPMS method for the quantitative determination of the various Cd-MTs in coral prawn. Effort has been directed to developing a simple extraction procedure to give intact Cd-MTs in an extract which is compatible with direct injection onto a reversed-phase HPLC column followed by selective detection of Cd and other metals with ICPMS. Factors investigated include extraction efficiency under various conditions, the stability of the MTs at each stage of sample preparation, and HPLC column recoveries of Cd. Besides parameters influencing the chromatographic separation, special attention has been paid to ICPMS instrumental factors influencing quantitative results.

We established a simple method for the quantitative determination of cadmium in sub-isoforms of metallothioneins separated with reversed phase chromatography using a methanol gradient. A novel approach to level the dependence of the response in ICPMS on the carbon content in the plasma was studied.