^{113}Cd NMR Characterization of the Metallothionein MTC from *Callinectes sapidus*

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Metallothioneins (MTs) are low molecular weight proteins with a high content in cysteines and a high ability to bind metal ions[1]. MTs are found in all living organisms. They coordinate essential metal ions, copper(I) or zinc(II), participating in their homeostasis. But MTs also binds toxic metals such as cadmium(II), silver(I) or mercury(II), being directly implicated in their detoxification. MTs are also involved in essential functions of the cells, such as their metabolic control of their energy production. Likewise, they act as antioxidant and antiapoptotic agents.

The crustacean *Callinectes sapidus* have three MTs, being MTC a singular isoform that is phylogenetically separated from the other two[2]. Among several distinct features, the primary structure of MTC contains two triplets of cysteines (CysCysCys). MTC also shows a high affinity for copper(I). Our group is studying MTC from *C. sapidus* with the goal of determining the role of the cysteine triplets, the way of binding of MTC to different metal ions and its three dimensional structure. Within this frame, we have amply characterized this metallothionein by means of several techniques. Here, we present the characterization we are carrying out through ^{1}H and ^{113}Cd Nuclear Magnetic Resonance.

^{113}Cd direct detection and heteronuclear ^{1}H-^{113}Cd experiments were performed in several CdMTC samples. We have also characterized the synthetic peptide MP10, whose primary sequence is that of the first 31 aminoacids of MTC. Comparing the spectra of the different samples we have deduced the number of metal ions directly bound to MTC, as well as the existence of two metal-thiolate clusters (A and B). One of them (cluster B) clearly binds four Cd(II) ions, while the other one (A) binds three ions with high affinity. In this last domain, one or more binding sites with lower affinity towards Cd ions are also observed.

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