TraceSpec2009: Detection of Ni(II) and Co(II) in fibroblasts using CLSM

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Metals have many different functions in human cells. As essential metals they act as cofactors in enzymes, as functional factors in different molecules like for example Fe in haemoglobin. Humans take up these metals with their food. If this food contains unnatural high concentrations of specific metals or specific metals and species, the metabolism can not bear with, toxic effects and deceases can occur.

To get information about the species and their localization in cells especially living cells is difficult using standard analytical techniques and requires optical methods with high lateral resolution like the confocal laser scanning microscopy. This technique in combination with standard fluorescent dyes for metal labelling and organelle labelling is suitable to display single living cells, their structures and the localization of specific metals. Newport Green DCF binds Ni(II) and Co(II) resulting in fluorescent enhancement. As test organism human fibroblasts from gingiva were used. The fibroblasts were incubated with different concentrations (100 µM, 300 µM and 500 µM) of the metal components $NiCl_2$, $NiSO_4$, Ni_3S_2 and $CoCl_2$ for different periods of time between 16h and 48h. The cells were then incubated with Newport Green DCF and analyzed with the CLSM with a magnification of 40.

The analysis showed that incubation with all four metal components tested result in fluorescence in the cells especially in structures around the nucleus. The fluorescence in cells incubated with Ni(II) components showed brighter fluorescence then cells treated with $CoCl_2$. Prolonging the incubation time with Ni(II) components and enhancing their concentration still result in fluorescence around the nucleus. Fluorescence is also detected inside the nucleus and the cytoplasm. In contrast to that, cells incubated with $CoCl_2$ showed no fluorescence inside the nucleus even after 48h of incubation with the highest tested concentration of 500 µM and additionally the fluorescence in $CoCl_2$ treated cells seems to be more diffuse. Parallel detection of Ni(II) or Co(II) with Newport Green DCF, the endoplasmic reticulum with ER Tracker and the golgi apparatus with Bodipy TR gave hints, that the fluorescence around the nucleus correlates with the endoplasmic reticulum.