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# Metabolic studies of lead toxicity and tolerance in green microalgae

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In an attempt to understand the metal detoxification mechanism of algae, we investigated the metabolic response of green microalgae exposed to various concentration of Pb. We determined the uptake and accumulation of Pb in *Chlorella vulgaris* by inductively coupled plasma mass spectroscopy (ICP-MS), and used the data to gain an understanding of effect of metals on the metabolic profiles and the Pb-detoxification mechanism, Pb-induced oxidative damage and antioxidative defense in *C. vulgaris* by nuclear magnetic resonance (NMR)-based metabolomics. Elemental analysis of algal medium and biomass showed that *C. vulgaris* displayed high Pb-removal efficiency (> 70%), and concentration-dependent Pb-accumulation capability (BCF > 1000). The high bioconcentration factor (BCF) can be interpreted as Pb being favourably taken up by *C. vulgaris* from the growth medium. This observation coincided with current knowledge of Pb-detoxification mechanism by phytochelatins, where *C. vulgaris* response to Pb exposure through the cheation of metals ions by this key class of chelators, and thus improved the Pb-accumulation capability in *C. vulgaris*. NMR-based metabolomics of *C. vulgaris* exposed to various concentration of Pb revealed the net changes in biochemical response between sample groups. Based on principal component analysis (PCA) of 1H spectra from *C. vulgaris* biomass, the Pb-dosed and control groups were significantly differentiated. The changes that influence the discrimination between the sample groups were in the concentration of lipids, surcose, betaine and several amino acids such as glutamate, lysine and arginine, which were substantially reduced in the Pb-dosed group as compared to the control group. We infer from the metabolic changes that excess Pb in *C. vulgaris* led to the formation of reactive oxygen species (ROS), which initiated lipid peroxidation and altered cell biochemical activities. In response to the oxidative stress, antioxidants (glutathione) and phytochelatins were upregulated to remove ROS and metal ions in the plants cells respectively. Significant reduction of glutathione and phytochelatin precursor (glutamate) demonstrated the Pb-detoxification and antioxidative defense mechanism of *C. vulgaris* in the presence of Pb.

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