

## Plastid NAD-dependent Malate Dehydrogenase: Cysteine oxidation as a regulatory switch of a multitasking protein?

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Most plant cell compartments possess oxidoreductases that catalyze the interconversion between malate and oxaloacetate (OAA) in a reversible reaction. These malate dehydrogenases (MDHs) are either NAD- or NADP-specific.

Together with malate/OAA translocators, MDHs enable the indirect exchange of reducing equivalents across subcellular membranes. The exported malate can then be used as a substrate for mitochondrial ATP production or to supply NADH to the cytosol.

Chloroplasts contain two MDH isoenzymes that differ in their cofactor specificity and function: the redox-regulated NADP-dependent MDH restores NADP upon illumination to prevent acceptor site limitation at Photosystem I and is only active in the light, whereas the plastid NAD-dependent MDH (pNAD-MDH) has been described to be constitutively active and to restore NAD for plastidial glycolysis. In addition, it has been proposed that pNAD-MDH has a moonlighting function in plastid biogenesis and is involved in starch degradation, both of which are independent of its metabolic activity.

Since pNAD-MDH appears to have multiple functions in central metabolism, redox homeostasis and plastid biogenesis, we were interested to find out if and how pNAD-MDH is regulated *in vitro* and found that pNAD-MDH is a target of redox regulation. Oxidation of recombinant pNAD-MDH inhibits its NAD-MDH activity and induces a conformational change of the protein visualized by non-reducing SDS-PAGE. Both redox effects were reversible upon re-reduction and prevented by its cofactors NAD and NADH. These results raise the possibility that pNAD-MDH is susceptible to redox regulation *in vivo* and provide an evidence for how pNAD-MDH might switch between different localizations and functions.

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