A novel twist on molecular interactions between thioredoxin and nicotinamide adenine dinucleotide phosphate-dependent thioredoxin reductase - DTU Orbit (04/02/2019)

A novel twist on molecular interactions between thioredoxin and nicotinamide adenine dinucleotide phosphate-dependent thioredoxin reductase

The ubiquitous disulfide reductase thioredoxin (Trx) regulates several important biological processes such as seed germination in plants. Oxidized cytosolic Trx is regenerated by nicotinamide adenine dinucleotide phosphate (NADPH)-dependent thioredoxin reductase (NTR) in a multistep transfer of reducing equivalents from NADPH to Trx via a tightly NTR-bound flavin. Here, interactions between NTR and Trx are predicted by molecular modelling of the barley NTR:Trx complex (HvNTR2:HvTrxh2) and probed by site directed mutagenesis. Enzyme kinetics analysis reveals mutants in a loop of the flavin adenine dinucleotide (FAD)-binding domain of HvNTR2 to strongly affect the interaction with Trx. In particular, Trp42 and Met43 play key roles for recognition of the endogenous HvTrxh2. Trx from Arabidopsis thaliana is also efficiently recycled by HvNTR2 but turnover in this case appears to be less dependent on these two residues, suggesting a distinct mode for NTR:Trx recognition. Comparison between the HvNTR2:HvTrxh2 model and the crystal structure of the Escherichia coli NTR:Trx complex reveals major differences in interactions involving the FAD- and NADPH-binding domains as supported by our experiments. Overall, the findings suggest that NTR:Trx interactions in different biological systems are fine-tuned by multiple intermolecular contacts. Proteins 2014; 82:607-619. (c) 2013 Wiley Periodicals, Inc.

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