

Alternative NADH:ubiquinone oxidoreductase (NDH2) of *Yarrowia lipolytica*

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Alternative NADH:ubiquinone oxidoreductases (NDH2) are found in the respiratory chain of plants, fungi and many bacteria. In contrast to complex I NDH2 does not contribute to the generation of a proton gradient across the membrane. In the obligate yeast *Yarrowia lipolytica* as an eukaryotic model system NDH2 is a small, single subunit enzyme of about 50-60 kDa. It contains one molecule non-covalently bound FAD and its active site is located at the outer face of the inner mitochondrial membrane. Redirection of NDH2 to the matrix side of the membrane by means of a N-terminally attached mitochondrial targeting sequence rescues the lethality of complex I deletion mutants and enables the cells to grow in the presence of the complex I inhibitor 2-decyl-4-quinazolinyl amine.

Kinetic measurements on mitochondrial membranes show, that NDH2 employs a ping-pong reaction mechanism.

1-hydroxy-2-dodecyl-4(1H)chinolon was found to be an effective inhibitor for NDH2, which causes a 50% inhibition at a concentration of 200 nM.

In the NDH2 sequences highly conserved regions can be found, e.g. two dinucleotide binding motifs as well as two apolar/aromatic regions. Site directed mutagenesis showed, that these domains may be involved in the binding of NADH and FAD and ubiquinone respectively.

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Key Words: Alternative NADH dehydrogenase; *Yarrowia lipolytica*; site directed mutagenesis