A contribution for the identification of azo reductase activity in intact yeast cells

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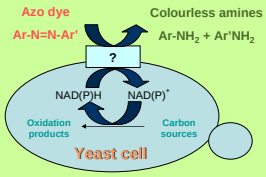
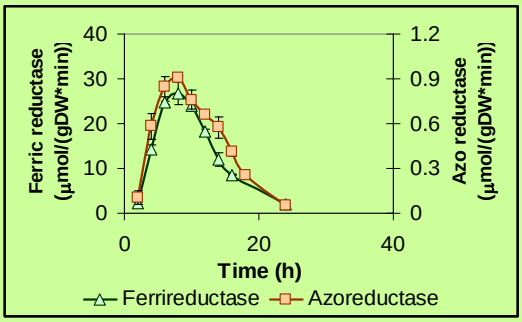
FACTS ABOUT AZO DYE DECOLOURISATION BY INTACT YEAST CELLS

- Reduction of the azo dye
- Unspecific and non-inducible activity
- Impermeant substrates
- Extracellular reduction



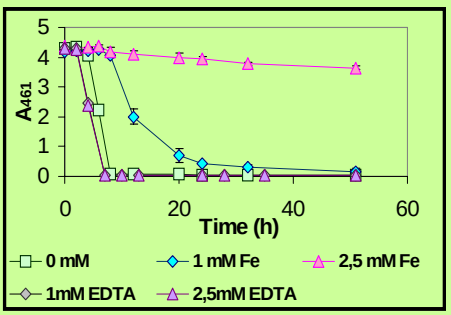
IS FERRIC REDUCTASE INVOLVED?



A

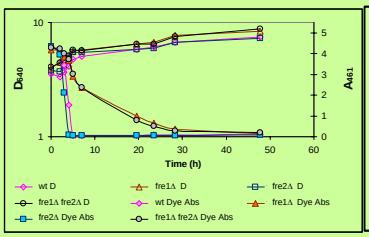
Ferric reductase may be involved in azo dye reduction because:

- Ferric and azo reductases have parallel activity curves with maxima in the late exponential growth phase (**A**)
- The addition of iron to the medium inhibits ferric reductase and delays decolourisation (**B**)

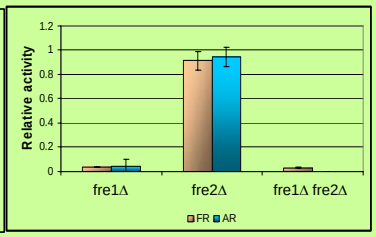


B

DEFECTIVE *fre1Δ* AND *fre2Δ* STRAINS



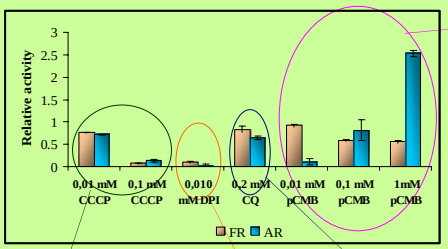
Reduction of decolourising capacity by delection of *FRE1* and *FRE2*



Effect of the deletion of *FRE1* and *FRE2* in the reductases activities

Fre1p is responsible for the major part of the azo reductase activity of intact yeast cells

INHIBITION



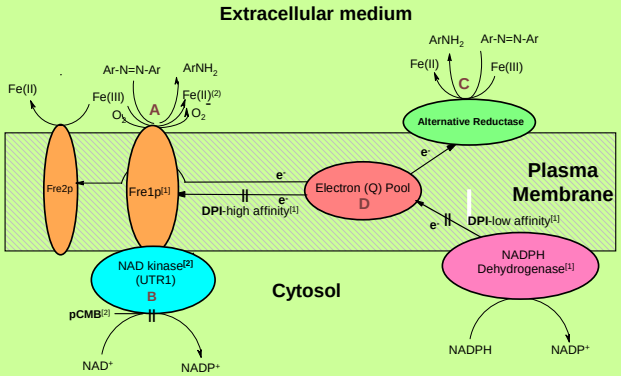
The stimulation of the azoreductase activity points to the existence of two alternative paths and inhibition of the most favourable one.

Both activities depend on a proton gradient

Both are NADPH dependent

Apparently CoQ_x is more important in the azo reductase activity

Proposed model



- **The major fraction of azo reductase activity depends on Fre1p [1; this work] and on a NADPH dehydrogenase [1] A**
- **Activity of Fre1p depends on a cytosolic NAD kinase [2] B**
- **Azo dye reduction (an presumably ferric iron reduction) must occur at an alternative site [this work]; also consistent with the properties of the rezasurin reductase [1] C**
- **pCMB stimulates azo reductase activity at higher concentrations [this work]**
- **These observations are consistent with the existence of membrane transporters capable of switching electrons between two external reduction sites (electron or Q pool) D**

[1] E Lesuisse, M Casteras-Simon and P Labbe 1996 JBC 271, 13578-13583

[2] S Kawai *et al.* 2001 FEMS Microbiol Lett 200, 181-184