

N°1117 / PC

TOPIC(s) : Biocatalytic cascade reactions / Enzyme discovery and engineering

## Exploiting ene-reductases promiscuous bioreduction of oximes to access tetrasubstituted pyrazines and aminoalcohols

### AUTHORS

Francesco MASCIA / AUSTRIAN CENTRE OF INDUSTRIAL BIOTECHNOLOGY, KRENNGASSE 37, GRAZ

Willem BREUKELAAR / UNIVERSITY OF GRAZ, HEINRICHSTRASSE 28/II, GRAZ

Stefan VELIKOGNE / UNIVERSITY OF GRAZ, HEINRICHSTRASSE 28/II, GRAZ

Nakia POLIDORI / UNIVERSITY OF GRAZ, HUMBOLDTSTRASSE 50/III, GRAZ

Amit SINGH / UNIVERSITY OF GRAZ, HUMBOLDTSTRASSE 50/III, GRAZ

Karl GRUBER / UNIVERSITY OF GRAZ, HUMBOLDTSTRASSE 50/III, GRAZ

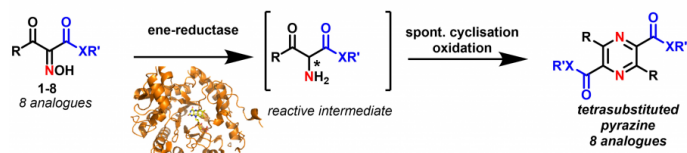
Silvia GLUECK / AUSTRIAN CENTRE OF INDUSTRIAL BIOTECHNOLOGY, KRENNGASSE 37, GRAZ

Corresponding author : Wolfgang KROUTIL / wolfgang.kroutil@uni-graz.at

### PURPOSE OF THE ABSTRACT

Promiscuous enzymatic activities recently gained much attention for contributing to the expansion of the repertoire of biocatalytic transformations, including new-to-nature reactions [1]. Ene-reductases (EREDs) from old yellow enzyme family are well known for their capacity to reduce C=C bonds [2]. Recently, we identified a promiscuous activity of ene-reductases showing that these enzymes reduce oximes like  $\alpha$ -oximo  $\beta$ -keto esters to their corresponding amines. In this reaction,  $\alpha$ -oximo  $\beta$ -keto esters were reduced to the corresponding  $\alpha$ -amino intermediate, which then spontaneously dimerized and oxidized, eventually leading to pyrazine products [3]. In this work, a library of eight  $\alpha$ -oximo  $\beta$ -keto esters was tested with six different ene-reductases, showing that all the substrates could be converted to the corresponding amines, with good product formation and isolated yields. Furthermore, to prove the presence of a reactive amine intermediate, a cascade reaction was set up, where an ADH acting on the intermediate but not on the substrate was selected, which led to the formation of chiral amino-alcohols [4].

## FIGURES



### FIGURE 1

#### Enzymatic oxime reduction

Reaction scheme of the ene-reductase catalysed oxime reduction to an amine intermediate, followed by the spontaneous oxidation and cyclisation leading to the tetrasubstituted pyrazines.

### FIGURE 2

## KEYWORDS

ene-reductases | promiscuous activity | oxime reduction | pyrazines

## BIBLIOGRAPHY

- [1] S. Athavale et al., J. Am. Chem. Soc. 2022, 144(41), 19097-19105
- [2] C. K. Winkler et al., Curr. Opin. Chem. Biol. 2018, 43, 97-105
- [3] S. Velikogne et al., ACS Catal. 2020, 10(22), 13377-13382
- [4] W. Breukelaar et al., ACS Catal. 2023, 13(4), 2610-2618