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Activity, stability and applications of aldoxime dehydratase biocatalysts immobilized on metal affinity resins

AUTHORS

Barbora KRISTKOVA / INSTITUTE OF MICROBIOLOGY OF THE CZECH ACADEMY OF SCIENCES, VIDENSKA 1083, PRAHA
Ludmila MARTINKOVA / INSTITUTE OF MICROBIOLOGY OF THE CZECH ACADEMY OF SCIENCES, VIDENSKA 1083, PRAHA
Robert RADISCH / INSTITUTE OF MICROBIOLOGY OF THE CZECH ACADEMY OF SCIENCES, VIDENSKA 1083, PRAHA
Lenka RUCKA / INSTITUTE OF MICROBIOLOGY OF THE CZECH ACADEMY OF SCIENCES, VIDENSKA 1083, PRAHA
Natalia KULIK / INSTITUTE OF MICROBIOLOGY OF THE CZECH ACADEMY OF SCIENCES, NOVOHRADSKA 237, TREBON
Margit WINKLER / ACIB GMBH AND GRAZ UNIVERSITY OF TECHNOLOGY, PETERSGASSE 14, GRAZ
Miroslav PATEK / INSTITUTE OF MICROBIOLOGY OF THE CZECH ACADEMY OF SCIENCES, VIDENSKA 1083, PRAHA

PURPOSE OF THE ABSTRACT

Aldoxime dehydratase (Oxd) is a valuable tool in organic synthesis, for producing nitriles from aldoximes in aqueous media at near-neutral pH and moderate temperatures, and, furthermore, with enantioselectivity to certain chiral aldoximes. However, the robustness of the Oxds needs to be increased to fully exploit their potential. One of the ways to achieve this goal is enzyme immobilization, but this has rarely been used for Oxds (e.g. in the form of whole cells immobilized in polyacrylic acid and used in organic solvents [1]).

In this study, we immobilized an Oxd on metal affinity resins (Talon, Ni-NTA) for the first time. Previously, this type of method was used, e.g., for the immobilization of a carboxylate reductase [2]. Here, we used OxdFv (an Oxd from Fusarium vanettenii), which we previously overproduced in Escherichia coli. This enzyme is of great interest, because it does not require anaerobic conditions. We also optimized the reaction to maximize the nitrile yield [3].

OxdFv was applied as a crude extract on Talon and Ni-NTA resins, and the unbound protein was washed out. The activities and stabilities of the immobilized Oxd catalysts were studied using a model reaction of (E,Z)-phenylacetaldoxime to phenylacetonitrile. The catalysts could be used for at least five cycles with over 40% conversion in each (Figure 1). The same approach can be used for preparative reactions, which increases the cost efficiency of the process in terms of catalyst productivity. Other arylacetonitriles, and alkylnitriles can be prepared in an analogous way.

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FIGURES

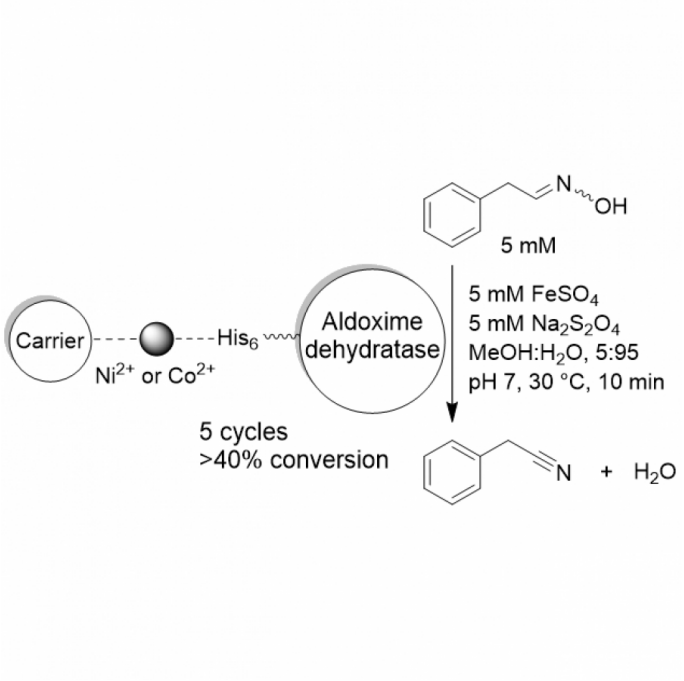

FIGURE 1

Figure 1

Conversion of (E,Z)-phenylacetaldoxime to phenylacetonitrile by aldoxime dehydratase immobilized on nickel or cobalt affinity resin.

FIGURE 2

KEYWORDS

Aldoxime dehydratase | (E,Z)-Phenylacetaldoxime | Phenylacetonitrile | Metal affinity resin

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