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Kinetic evaluation and reaction optimization of a two-step enzymatic cascade for sustainable biosynthesis of coniferol from lignin-derived ferulic acid

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PURPOSE OF THE ABSTRACT

Coniferol (CA) is one of promising precursors for varieties of flavor, fragrance, and pharmaceutical chemicals. A two-step enzymatic reaction to convert ferulic acid (FA) into CA via activities of a carboxylic acid reductase from *Norcadia iwensis* (NiCAR) and an aldo-keto reductase from *Coptotermes gestroi* (CgAKR-1) has been recently published in literature, mainly focusing on whole cell biocatalysis. FA is a phenolic compound which can be derived from hydrolysis of lignin-containing biomass. Thus, this study focused on evaluation of kinetics of NiCAR and CgAKR-1 and optimization of the enzymatic reaction to outline bioprocess design to improve productivity of CA.

Kinetics study of NiCAR towards FA and CgAKR-1 towards CALD pointed out inhibitory effects of both substrates against the enzymes. It suggested that maintaining low concentrations of the substrates might allow greater productivity of CA. Ratio of NiCAR to CgAKR-1 clearly affects productivity of CA which the optimum was found at 1U of NiCAR to 25 U of CgAKR-1 at a certain condition. NADPH regeneration via oxidation of glucose by the glucose dehydrogenase (GDH) was introduced into the system to overcome expensive supplementation of the cofactor. However, development of an effective ATP regeneration is still a challenge.

In term of bioprocess design, the enzymes were immobilized on nickel-nitrilotriacetic acid (Ni-NTA) agarose beads. The immobilized enzymes exhibited significantly improved activity and stability without kinetic parameters alteration. As batch, it provided volumetric productivity of CA at 57 mg·L⁻¹·h⁻¹. The productivity was clearly improved to 95 mg·L⁻¹·h⁻¹ when performed as fed batch. Moreover, the immobilized enzyme can convert FA obtained from an alkaline hydrolysis of sugar beet pulp as batch with productivity of 15 mg·L⁻¹·h⁻¹.

In summary, the study provided kinetics understandings of NiCAR and CgAKR-1 enzymes to convert FA into CA, contributing to further bioprocess design to intensify the productivity of CA. Moreover, it emphasizes potential of enzymatic cascade reaction, particularly CAR-AKR-GDH, for the synthesis of high value chemicals from lignocellulosic waste stream.

FIGURES

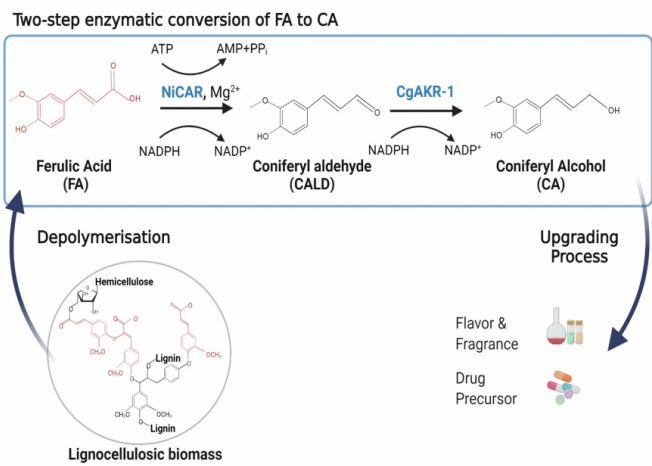


FIGURE 1
Figure 1 Illustration of two-step enzymatic conversion of FA into CA.
The figure shows potential of the two-step enzymatic reaction to convert FA derived from lignocellulosic biomass into CA, potentially serving as a precursor for fragrance and

FIGURE 2

KEYWORDS
Coniferyl alcohol | Lignocellulosic biomass | Carboxylic acid reductase (CAR) | Aldo-keto reductase (AKR)

BIBLIOGRAPHY