

1 Nomenclature

EC number

4.1.3.41

Systematic name

3-hydroxy-D-aspartate glyoxylate-lyase (glycine-forming)

Recommended name

3-hydroxy-D-aspartate aldolase

Synonyms

D-3-hydroxyaspartate aldolase <1> [1]

D-HAA <1> [1]

2 Source Organism

<1> *Paracoccus denitrificans* (UNIPROT accession number: Q8GRC8) [1]

3 Reaction and Specificity

Catalyzed reaction

D-erythro-3-hydroxyaspartate = glycine + glyoxylate

threo-3-hydroxy-D-aspartate = glycine + glyoxylate

Substrates and products

S 2-amino-3-hydroxybutanedioic acid <1> (Reversibility: r) [1]

P aminoacetic acid + oxoacetic acid

S D-3-3,4-dihydroxyphenylserine <1> (Reversibility: ?) [1]

P ?

S D-erythro-3-3,4-methylenedioxyphenylserine <1> (Reversibility: ?) [1]

P ?

S D-erythro-3-hydroxyaspartate <1> (<1> the enzyme is strictly D-specific as to the α -position, whereas it does not distinguish between threo and erythro forms at the β -position [1]) (Reversibility: ?) [1]

P glycine + glyoxylate

S D-erythro-3-phenylserine <1> (Reversibility: ?) [1]

P ?

S D-threo-3-3,4-methylenedioxyphenylserine <1> (Reversibility: ?) [1]

P ?

S D-threo-3-phenylserine <1> (Reversibility: ?) [1]

P ?

S allo-threonine <1> (Reversibility: ?) [1]

P ?

S Additional information <1> (<1> the enzyme shows no activity towards L-erythro-3-hydroxyaspartate, L-threo-3-hydroxyaspartate, L-threonine, L-allo-threonine, L-erythro-3-phenylserine, L-threo-3-phenylserine, L-erythro-3-3,4-methylenedioxyphenylserine, and L-threo-3-3,4-methylenedioxyphenylserine [1]) (Reversibility: ?) [1]

P ?

Inhibitors

EDTA <1> (<1> 82% inhibition at 1 mM [1]) [1]

Cofactors/prosthetic groups

pyridoxal 5'-phosphate <1> [1]

Metals, ions

Co²⁺ <1> (<1> 2.0fold increase of specific activity in the presence of 1 mM Co²⁺ [1]) [1]

Mg²⁺ <1> (<1> 5.4fold increase of specific activity in the presence of 1 mM Mg²⁺ [1]) [1]

Mn²⁺ <1> (<1> 8.1fold increase of specific activity in the presence of 1 mM Mn²⁺ [1]) [1]

Additional information <1> (<1> K⁺ and Na⁺ do not have an effect on the enzymatic activity [1]) [1]

Specific activity (U/mg)

0.36 <1> (<1> after 50fold purification, using DL-threo-3-hydroxyaspartate as substrate, in 0.02 mM HEPES buffer, pH 8.0, at 30°C [1]) [1]

0.6 <1> (<1> crude extract, in 0.02 mM HEPES buffer, pH 8.0, at 30°C [1]) [1]

14.6 <1> (<1> after 50fold purification, using D-threo-3-3,4-methylenedioxyphenylserine as substrate, in 0.02 mM HEPES buffer, pH 8.0, at 30°C [1]) [1]

17.5 <1> (<1> after 50fold purification, using D-erythro-3-3,4-methylenedioxyphenylserine as substrate, in 0.02 mM HEPES buffer, pH 8.0, at 30°C [1]) [1]

20 <1> (<1> after 50fold purification, using D-threonine as substrate, in 0.02 mM HEPES buffer, pH 8.0, at 30°C [1]) [1]

25 <1> (<1> after 50fold purification, using D-allo-threonine as substrate, in 0.02 mM HEPES buffer, pH 8.0, at 30°C [1]) [1]

30 <1> (<1> after 50fold purification, using D-erythro-3-hydroxyaspartate as substrate, in 0.02 mM HEPES buffer, pH 8.0, at 30°C [1]) [1]

95 <1> (<1> after 50fold purification, using D-threo-3-phenylserine as substrate, in 0.02 mM HEPES buffer, pH 8.0, at 30°C [1]) [1]

99 <1> (<1> after 50fold purification, using D-erythro-3-phenylserine as substrate, in 0.02 mM HEPES buffer, pH 8.0, at 30°C [1]) [1]

K_m-Value (mM)

0.4 <1> (D-erythro-3-hydroxyaspartate, <1> in 0.02 mM HEPES buffer, pH 8.0, at 30°C [1]) [1]

pH-Optimum

9 <1> [1]

pH-Range

7.5-10 <1> [1]

Temperature optimum (°C)

35 <1> [1]

4 Enzyme Structure

Molecular weight

80000 <1> (<1> gel filtration [1]) [1]

Subunits

homodimer <1> (<1> 2 * 43000, SDS-PAGE [1]; <1> 2 * 41633, calculated from amino acid sequence [1]) [1]

5 Isolation/Preparation/Mutation/Application

Purification

<1> (ammonium sulfate fractionation, hydroxyapatite column chromatography, DEAE-Toyopearl column chromatography, phenyl-Toyopearl column chromatography, Superdex 200 gel filtration, and Mono Q column chromatography) [1]

Cloning

<1> (expressed in Escherichia coli XL1-Blue MRF^c cells) [1]

6 Stability

pH-Stability

6.5-8.5 <1> (<1> the enzyme is stable between pH 6.5 and 8.5 for 30 min at 30°C [1]) [1]

Temperature stability

45 <1> (<1> the enzyme retains 50% activity upon heating at 45°C for 30 min [1]) [1]

References

- [1] Liu, J.Q.; Dairi, T.; Itoh, N.; Kataoka, M.; Shimizu, S.: A novel enzyme, D-3-hydroxyaspartate aldolase from *Paracoccus denitrificans* IFO 13301: purification, characterization, and gene cloning. *Appl. Microbiol. Biotechnol.*, **62**, 53-60 (2003)