

1 Nomenclature

EC number

5.5.1.15

Systematic name

terpentedienyl-diphosphate lyase (decyclizing)

Recommended name

terpentedienyl-diphosphate synthase

Synonyms

Cyc1 <2> [2]

CAS registry number

429681-55-0

2 Source Organism

- <1> *Streptomyces lividans* [3]
<2> *Kitasatospora griseola* [1,2]

3 Reaction and Specificity

Catalyzed reaction

geranylgeranyl diphosphate = terpentedienyl diphosphate

Substrates and products

S geranylgeranyl diphosphate <1,2> (<1> the triggering proton is lost at the end of the cyclization reaction [3]) (Reversibility: ?) [1,2,3]

P terpentedienyl diphosphate

Inhibitors

EDTA <2> [2]

geranylgeranyl diphosphate <2> (<2> above 0.05 mM [2]) [2]

Activating compounds

Additional information <2> (<2> 20% glycerol, 5 mM 2-mercaptoethanol, and 0.1% Tween 80 are required for the full activity of the Cyc1 [2]) [2]

Metals, ions

Mg²⁺ <2> (<2> the enzyme activity of the Cyc1 is highest at a concentration of 1 mM but slightly inhibited at a concentration of 10 mM (decreased 40%) [2]) [2]

Additional information <2> (<2> no activity is detected with other divalent metal ions such as Ca²⁺, Co²⁺, Cu²⁺, Fe²⁺, Mn²⁺, and Zn²⁺ at both 1 and 10 mM [2]) [2]

K_m-Value (mM)

0.0642 <2> (geranylgeranyl diphosphate) [2]

pH-Optimum

6.8 <2> (<2> Tris-HCl buffer [2]) [2]

Temperature optimum (°C)

25-30 <2> [2]

Temperature range (°C)

25-50 <2> (<2> 25-30: optimum, 50°C, no activity detected above [2]) [2]

4 Enzyme Structure

Molecular weight

50000 <2> (<2> gel filtration [2]) [2]

5 Isolation/Preparation/Mutation/Application

Purification

<2> [1]

Cloning

<2> [2]

<2> (heterologous expression of the cyclase genes in *Streptomyces lividans* and *Escherichia coli*) [1]

6 Stability

Temperature stability

30 <2> (<2> full activity after incubation at 30°C in 0.05 M Tris-HCl buffer for 1 h [2]) [2]

References

- [1] Dairi, T.; Hamano, Y.; Kuzuyama, T.; Itoh, N.; Furihata, K.; Seto, H.: Eubacterial diterpene cyclase genes essential for production of the isoprenoid antibiotic terpenecin. *J. Bacteriol.*, **183**, 6085-6094 (2001)
- [2] Hamano, Y.; Kuzuyama, T.; Itoh, N.; Furihata, K.; Seto, H.; Dairi, T.: Functional analysis of eubacterial diterpene cyclases responsible for biosynthesis of a diterpene antibiotic, terpenecin. *J. Biol. Chem.*, **277**, 37098-37104 (2002)
- [3] Eguchi, T.; Dekishima, Y.; Hamano, Y.; Dairi, T.; Seto, H.; Kakinuma, K.: A new approach for the investigation of isoprenoid biosynthesis featuring pathway switching, deuterium hyperlabeling, and ^1H NMR spectroscopy. The reaction mechanism of a novel streptomyces diterpene cyclase. *J. Org. Chem.*, **68**, 5433-5438 (2003)