Abstract

Lincomycin is an antibiotic used in clinical praxis. It is produced by *Streptomyces lincolnensis*. Lincomycin is composed of an amino-sugar and an amino-acid moiety linked by an amide bond. The amino-acid precursor is propylproline (PPL), whose biosynthesis undergoes the pathway derived from tyrosine. The modified PPL biosynthesis pathway was also discovered in pyrrolobenzodiazepines (PBD) and hormaomycin. In the biosynthesis of PBD the PPL precursor is further modified by reactions catalysed by specific enzymes missing in the biosynthesis of lincomycin. The genes encoding these enzymes could be transferred to the lincomycin biosynthetic gene cluster. In this way we could get producers of hybrid antibiotics with better properties and even antimalaric effects.

Six enzymes participate in PPL biosynthesis, which are encoded in the lincomycin biosynthetic gene cluster. The first two reactions of PPL biosynthesis pathway are proven, therefore, this work focuses on the third reaction that is supposed to be catalysed by protein LmbX according to literature. The proposed function of LmbX is a hydrolysis of C-C bond. However, LmbX belongs to the protein family of isomerases by sequence homology.

The protein LmbX was overproduced in this work and its activity was tested in the presence of the expected substrate. The products were measured on the liquid chromatography (UHPLC). The hydrolytic function failed to prove. Furthermore, the products of the prepared deletion mutants of genes of PPL pathway were analysed using UHPLC. The results indicate that LmbX is probably an isomerase that catalyzes the transfer of the double bond of the intermediate of PPL biosynthesis pathway and that the previously supposed scheme should be revid. In this work the new scheme corresponding with obtained results is proposed.