Nenemycin is a newly discovered 31-membered cyclodepsipeptide antibiotic isolated from an aquatic Streptomyces. This cyclic molecule contains ester and peptide bonds and is biosynthesized by the combined action of multiple enzymes. Among those is NenD1, an argininosuccinate lyase involved in the synthesis of 2S,3R-2,3-diaminobutanoic acid (DABA), a modified amino acid later added to the cycle of the nenemycin biosynthesis. The structure of NenD1 has been determined at 2.7 Å, showing the homotetrameric arrangement of the enzyme. A new crystal form was obtained after reductive methylation of lysine residues and the resulted 2.9 Å structure reveals features not present in the original. The active site is located at the interface of three subunits and its architecture was compared with other similar enzymes. Our study provides further insights into the synthesis mechanism of the unusual amino acid.

**Keywords:** Argininosuccinate lyase, Antibiotics