Poster G-3

Molecular mechanics studies of pyrophosphorylase, template enzyme involved in chitin metabolic pathway

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Short Abstract: Pyrophosphorylase of Crinipellis perniciosa (agent of witches' broom disease) was sequenced, and a homologous protein (1JV1) was obtained from PDB with 50.1% sequence similarity. 1JV1 structure was refined using molecular mechanics methods and this will be used as a template to obtain the 3D model of pyrophosphorylase structure of C. perniciosa by comparative modeling approach.

Long Abstract:

Crinipellis perniciosa (Stahel) Singer is the causal agent of witches' broom disease, which is endemic in Brazilian Amazon, and was, first reported to infect Theobroma cacao (cocoa) in the state of Bahia, Brazil, in 1989 [1]. The first consequence is a decreased production of cocoa about 60%, which changed the Brazilian position in international market from exporter to importer of cocoa. In other words, in the last 11 years Brazilian market of cocoa range from US$ 700 million to US$ 260 million [2]. C. perniciosa has a set of potential enzymes which can serve as target for the design of new inhibitor compounds. One of these enzymes belongs to the class of pyrophosphorylase, and is present in the metabolic pathway of synthesis of chitin [3]. The release of the amino acid sequence for this enzyme initiated studies to obtain its 3D structure by means comparative modeling [4]. The primary sequence of the pyrophosphorylase of C. perniciosa obtained from the previous studies was queried in blastp using Protein Data Bank (PDB). A human pyrophosphorylase (1JV1) was obtained from blastp which has 50.1% sequence similarity with C. perniciosa pyrophosphorylase. This homologous sequence was aligned with C. perniciosa pyrophosphorylase using CLUSTAL-W. The sequence from CLUSTAL-W revealed 9 gaps and 3 disulfide bridges. A set of molecular mechanics calculations using different non-bond cut-off values ranging from 3Ǻ to 20Ǻ was performed using MM3 methodology present in BioMedCache. Subsequently, the best non-bond cut-off value was determined to be 14 Ǻ and used to refine the crystal structure using Steepest-Descent (SD)-Conjugate Gradient (GC)-Steepest-Descent (SD) protocol. The energy for final optimization of the system in presence of explicit solvent (water) was found to be -2244 kcal/mol. This refined structure of (1JV1) to be used as a template to obtain the 3D model of pyrophosphorylase structure of C. perniciosa by comparative modeling approach.

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