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Unraveling the catalytic mechanism of Tryptophan synthase, a drug target against *Mycobacterium tuberculosis*

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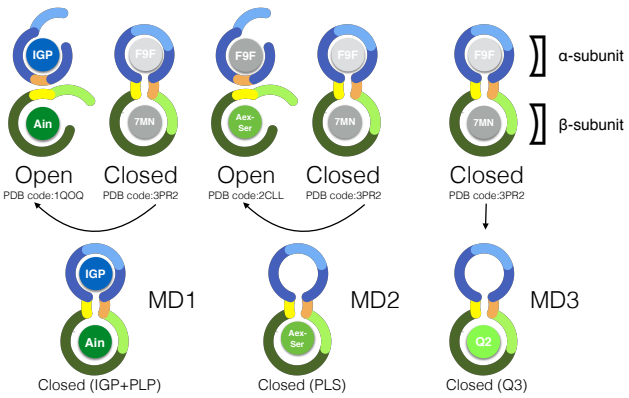
Graphical Abstract



Abstract.

Tryptophan Synthase (TSase) is a bifunctional enzyme that catalyzes the last two steps in the synthesis of tryptophan (TRP). Each reaction is catalyzed in different active sites that are located in separate α and β subunits. The active site of the α-subunit catalyzes the formation of indole and gliceraldeyde-3-phosphate (G3P) from indole 3-glycerolphosphate (IGP). Indole is then transported through a 25 ≈ physical tunnel to the active site of the β-subunit where it is added to a molecule of acrylate, derived from serine, to produce TRP, in a PLP dependent reaction[1].

Since TSase is absent in mammals, it is a promising target for the development of new antibiotics and vaccines against infectious bacteria, such as *Mycobacterium tuberculosis.*

The complex allosteric regulation of the enzyme has turn it difficult to co-crystalize the enzyme in its closed conformation with both substrates correctly placed in the α and β-active sites. In this work, we modulated three enzyme models for the posterior construction of QM/MM models: Model 1 (α-IGP and β-Ain);

	Model 2 (β-Aex-Ser); Model 3 (β-Q2). All the models were based on the crystallographic structure with PDB ID: 3PR2 and the ligands were either obtained from other crystallographic structures (PDB ID:1QOQ) or modulated from the analogs. Each of the three models were emerged in a box of waters and subjected to a MD simulation of 30 ns for detailed analysis and sampling of the interactions formed. The RMSd analysis of the last 20 ns of the three MD simulations did not evidence any abnormal fluctuation, and the equilibrated region presents a low RMSd average value of respectively 2.90 ± 0.17 Å for MD1; 2.64 ± 0.12 Å for MD2 and 2.37± 0.10 Å for MD3. We concluded that all the models are stable and can be the basis for further studies. Afterwards four ONIOM QM/MM hybrid model were built for geometry optimization, validation of the initial enzyme-ligand interaction, and posterior study of the catalytic mechanism of both α and β subunits of the enzyme.

Acknowledgments

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References

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