Poster Presentation

MS009.P25

Understanding IMPDH and ADSS in Cryptococcus neoformans andtheir inhibitors design

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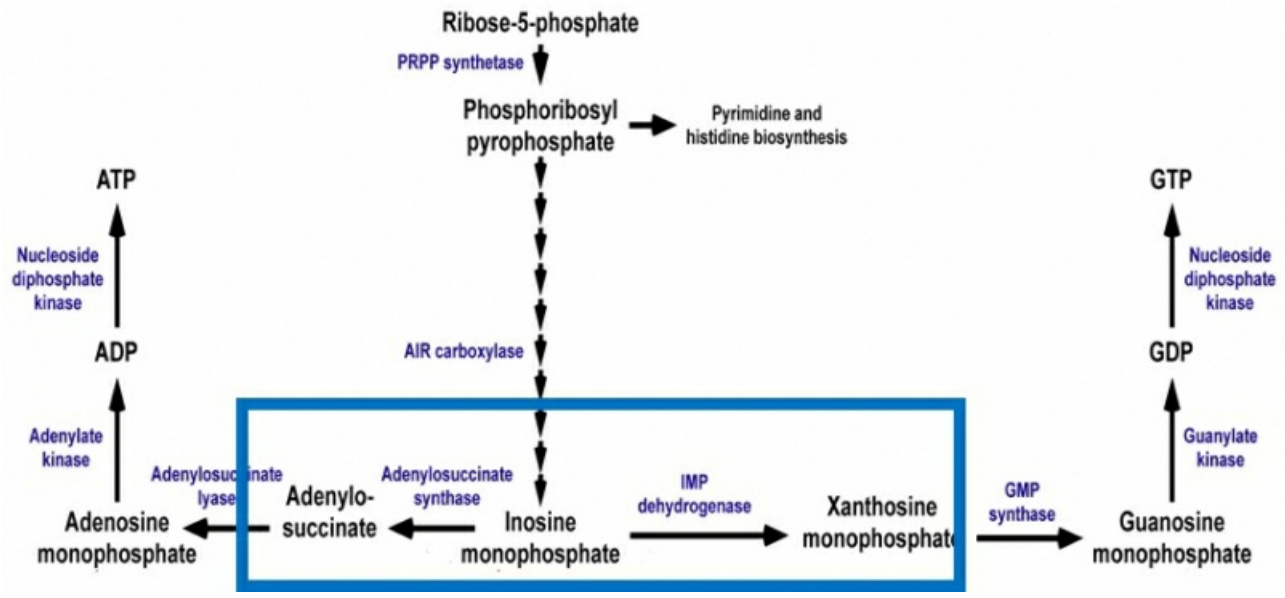
Infections caused by fungi and bacteria are a major global problem. In humans, cryptococcosis is one of the life-threatening infection caused by fungi Cryptococcus neoformans. The treatment is based on three antifungal agents: Amphotericin B, Fluconazole and Flucytosine. However, there are significant side effects associated with these drugs and increased drug resistance in some cases. The discovery of new antifungals is essential to fight against the rise of invasive infections caused by fungal pathogens.

In all organisms, purine metabolic pathway is essential for the biosynthesis of adenosine triphosphate (ATP) and guanosine triphosphate (GTP) nucleotides. In de novo purine biosynthetic pathway, Inosine monophosphate (IMP) acts as a branch point substrate for the formation of ATP and GTP through two different directional pathways catalyzed by Adenylosuccinate synthetase (ADSS) and Inosine monophosphate dehydrogense (IMPDH). To design inhibitors of C. neoformans IMPDH (CnIMPDH) the high-throughput screening was conducted with 114,000 drug-like compounds from Walter and Eliza Hall Compound Collection before to onset of my thesis work. One of the interesting hit set that I have been studying is based on benzothiophene-1,1-dioxide moiety and by modification of aromatic substituents on this hit molecule we found potent CnIMPDH inhibitors with antifungal activity. The studies for identifying the binding site of these inhibitors through kinetics and crystallography are under progress and will be published soon.

Regarding C. neoformans ADSS (CnADSS), the structural elements essential for the substrate (IMP) and cofactor (GTP) binding are studied using protein crystallography technique. This information could be useful for the design of inhibitors through computational studies. The plan of utilizing computational studies is to substitute the traditional high throughput screening (HTS) that requires lot of time and cost for identification of hits and their development to lead molecules. The in vitro assays on identified hits for inhibition of CnADSS (IC50) and finding the site of binding through crystallography are in progress.

[1] Morrow CA, Valkov E, Stamp A, Chow EWL, Lee IR, Wronski A, Williams SJ, Hill JM, Djordjevic JT, Kappler U, Kobe B, Fraser JA. (2012). De novo GTP Biosynthesis Is Critical for Virulence of the Fungal Pathogen Cryptococcus neoformans. PLoS Pathog 8(10): e1002957.

[2] Blundell RD, Williams SJ, Arras SDM, Chitty JL, Blake KL, Ericsson DJ, Tibrewal N, Rohr J, Koh YQAE, Kappler U, Robertson AAB, Butler MS, Cooper MA, Kobe B, Fraser JA. (2016). Disruption of de Novo Adenosine Triphosphate (ATP) Biosynthesis Abolishes Virulence in Cryptococcus neoformans. ACS Infectious Diseases. 2(9):651-63.



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