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P-307 - SUBSTRATE INHIBITION OF URACIL PHOSPHORIBOSYLTRANSFERASE (UPRT) IN DISTINCT MICROBIAL SPECIES AND ITS IMPLICATIONS IN THE PHENOTYPE OF URACIL AUXOTROPHS

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Background

Unlike the established practice, the supply of exogenous uracil cannot fully rescue uracil auxotrophy in Ura- mutants of some species of industrial and/or medical significance (e.g. *Ashbya gossypii*, *Candida guilliermondii*, *C. albicans*, *Leishmania donovani*), which require uridine for proper growth [1-4]. Here we reveal the molecular mechanism underlying this phenotype in *A. gossypii* and disclose common features with protozoan parasites and other flavinogenic/pathogenic fungi.

Method

Analysis of the *A. gossypii Agura3* (Ura- mutant) radial growth on minimal medium containing distinct pyrimidine supplements allowed the detection of a bottleneck in the *A. gossypii* pyrimidine salvage pathway at the Uracil Phosphoribosyltransferase (AgUPRT) level. This enzyme catalyzes the production of uridine monophosphate (UMP) from uracil and phosphoribosyl pyrophosphate (PRPP), being encoded by *AgFUR1*. Recombinant AgUPRT was produced and purified from *Escherichia coli*, and its activity characterized spectrophotometrically [4].

Results & Conclusions

Characterization of recombinant AgUPRT revealed that it is susceptible to substrate inhibition by uracil, thus explaining the hypersensitivity of *A. gossypii Agura3* to uracil and its requirement for uridine [1]. This UPRT substrate inhibition mechanism, together with similar Ura- phenotypes, is also present in medically relevant protozoan parasites [4] and phenotypic evidences indicate that it likely exists in other flavinogenic fungi [2]. Substrate inhibition of AgUPRT favours the preservation of PRPP for use in purine synthesis/salvage. While in *A. gossypii* and other flavinogenic fungi (e.g., *C. guilliermondii*) purine synthesis is crucial to support the highly active biosynthesis of riboflavin [1-2], in human pathogens such as *C. albicans* and *L. donovani* the intracellular availability of purines is essential during infection [4-5].

References & Acknowledgments

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