DISSERTATION ABSTRACT

Hegelund Myrbäck, Tove. **Characterization of CYP3A in fish - expression, inhibition, induction and analyses of gene evolution.** Department of Zoophysilogy, Göteborg University, Box 463, SE-405 30 Göteborg, Sweden

The organism is constantly exposed to foreign substances, *e.g.* food and pollutants. Cytochrome P450 (CYP) enzymes, including the CYP3A isoforms, facilitate the excretion of lipophilic compounds, *e.g.* steroids, procarcinogens and drugs, to prevent toxic accumulation. Although CYP3A genes are evolutionarily well conserved, the response to certain inducers differ between species. In addition, several substances present in the environment are CYP3A inhibitors. Altered CYP3A metabolism could affect xenobiotic clearance and steroid hormone homeostasis.

Aims of the study

(I) To determine tissue- and cell-specific expression of CYP3A in adult killifish. (II) To study the impact of *N*-substituted azole fungicides (*i.e.* ketoconazole, propiconazole, miconazole, clotrimazole) on CYP3A and CYP1A expression and metabolism. (III) To study the induction of CYP3A using prototypical mammalian inducers, *i.e.* pregnenolone 16α-carbonitrile and azole fungicides, in rainbow trout and killifish. (IV) To isolate fish and reptile CYP3A sequences for evolutionary analyses of the subfamily.

Results and discussion

CYP3A expression was prominent in the gastrointestinal and respiratory tracts in killifish. Low levels were seen in kidney, spleen, brain and ovaries. Males generally had higher CYP3A levels than females. Inter-individual variations in the expression were seen. The results imply sex-dependent CYP3A regulatory mechanisms and/or metabolic functions. The azole fungicides primarily inhibited CYP3A and CYP1A activities, although the CYP1A protein levels were induced. Azole fungicides and PCN resulted in weak or no induction of CYP3A, indicating different regulatory mechanisms, compared to mammals. CYP3A56 was isolated from killifish and CYP3A42 from ball python. The genes were included in evolutionary studies of the gene family. The CYP3A gene family probably evolved through separate gene duplication events in most species. In rodents and primates, however, gene duplications probably appeared before speciation.

Keywords; cytochrome P450, CYP3A, tissue specific expression, inhibition, induction, azole fungicides, gene evolution, fish

ISBN 91-628-5695-2