

## Abstract

Insects account for more than one million of described species with an ecological and economic impact disproportional to their minute body size. Among the factors which have contributed to their evolutionary success, insect secondary metabolites such as defensive compounds and chemical signals are regarded to play a major role. This thesis aims at uncovering the molecular mechanisms underlying evolution of ubiquitous insect secondary metabolites – sex pheromones (SPs), i.e. chemical signals mediating mate finding and mating between individuals of the same species. The thesis focuses on a class of oxidoreductase enzymes, membrane fatty acid desaturases (mFADs), which introduce double bonds into hydrocarbon chains of fatty acyls and thus produce precursors of unsaturated fatty acid-derived SPs. mFADs are involved in SP biosynthesis in e.g. moths (Lepidoptera), flies (Diptera), cockroaches and termites (Blattodea), wasps and bees (Hymenoptera) - some of the most species-rich insect orders. Since SPs are principal to species reproductive isolation, uncovering the molecular basis of insect SP biosynthesis holds promises to contribute to answering fundamental questions concerning the insect ecology and evolution. The insect mFADs with diverse enzymatic specificities also represent a naturally available resource for study of enzyme function evolution.

This thesis explores mFADs in Hymenoptera (bumblebees - *Bombus*) and Lepidoptera (tobacco hornworm moth - *Manduca sexta*) as well as in non-insect organism (yeast – *Candida parapsilosis*). We demonstrate that the ability to produce a wide range of unsaturated fatty acids is inherent to mFADs across kingdoms (Publications I and III). We show that pheromone-biosynthetic mFADs can synthesize novel unsaturated SP precursors as a result of a single amino acid substitution, a mechanism which might have a high potential in generating novel SP components in moths and represents thus a possible molecular mechanism of SP evolution (Publication I). Our finding that the amino acid residue which controls *M. sexta* mFAD specificities resides in the kink of the mFAD substrate binding channel provides novel insights into mechanism of mFAD substrate specificity determination (Publication I). By study of mFADs from three bumblebee species we show that post-transcriptional regulation of mFAD activity represents an alternative possible regulatory mechanism of pheromone composition in hymenopterans (Publication II). Together, these findings expand our knowledge on determinants of mFADs enzymatic specificities and contribute to our understanding of the role which mFADs play in SP biosynthesis and evolution of SP communication in moths and bees.