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Cunninghamellla elegans: A Fungal Model for Mammalian Xenobiotic Metabolism

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The filamentous fungus *Cunninghamella elegans* has been widely used as a microbial model for mammalian xenobiotic metabolism (Clark and Hufford, 1991) as well as for the biodegradation of environmentally relevant chemicals (Cerniglia, 1997). The fungus has shown the ability to metabolize a wide variety of xenobiotics, including polycyclic aromatic hydrocarbons and pharmaceutical drugs, especially by hydroxylation, N-demethylation and N-oxidation. Numerous studies have been focused on the metabolism of tricyclic antihistamine and antidepressant drugs by this fungus. This study also deals with the fungal metabolism of malachite green, an N-methylated diaminotriphenylmethane dye, which is widely used as an antifungal agent in the aquaculture industry and a dye in the textile industry (Cha et al., 2001). Metabolites were elucidated by high performance liquid chromatography/mass spectrometry, indicating that this fungal system produced similar metabolite profiles to those observed in rat liver. Malachite green was enzymatically reduced to leucomalachite green, and also converted to N-demethylated and N-oxidized metabolites, including primary and secondary arylamines. The results indicated that reduction and N-demethylation occur simultaneously by the cytochrome P450 system. A cytochrome P450 gene from *C. elegans* was cloned and sequenced, suggesting that the *C. elegans* P450 clustered close to the cyp51 family rather than fungal cytochrome P450s (Wang et al., 2000). Thus, *C. elegans* is a suitable microbial model for triphenylmethane dye metabolism.

Although the fungus produces phase I and phase II enzymes, little is known about the enzymes involved in the biotransformation of xenobiotics. Glutathione *S*-transferases (GSTs; EC 2.5.1.18) are a family of multifunctional enzymes that catalyze the conjugation of a wide variety of electrophilic compounds with glutathione and are phase II enzymes of xenobiotic detoxification. GSTs are found in most aerobic eucaryotes and procaryotes, and occur as multiple enzyme forms. Mammalian cytosolic GSTs have been divided into alpha, pi, mu and theta classes on the basis of primary structure, substrate specificity and immunological properties. More recently different classes of GSTs, such as sigma, kappa, zeta and omega have been identified. A GST has been purified and characterized from *C. elegans* (Cha et al., 2001). Using a degenerate oligonucleotide probe based on the N-terminal sequence of the purified protein, the structural gene for GST in *C. elegans* was cloned and sequenced by screening a cDNA library. The deduced amino acid sequence showed 14-25% sequence identity to the sequences of other known classes of GSTs. Further screening of a cDNA library with the gene probe revealed the presence of another GST isozyme in this fungus with 85% sequence identity. Mutagenesis analysis indicated that Tyr-9 in the N-terminal region is essential for catalysis of the enzyme. Overall results suggest that the GSTs from *C. elegans* belong to a novel class of GST proteins. Therefore, the GSTs from *C. elegans* were proposed to be GST class gamma and designated as GSTG1-1 and GSTG2-2, respectively (Cha et al., 2002). Further investigation on the structure and function of new microbial GSTs may provide new insights into GST evolution, possible detoxification pathways and ecological fates of toxic chemicals in the environment.

References

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