ELUCIDATING THE FUNCTION OF "ORPHAN" HUMAN CYTOCHROME P450S
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Roughly one-fourth of the 57 human P450 genes do not have well-established functions in the oxidation of endogenous or xenobiotic chemicals and are still considered orphans. One major approach to elucidate their functions involves heterologous expression and in vitro definition of substrates1. We used an Escherichia coli system to achieve active expression of several orphan P450s. In some cases, the expression levels were markedly increased by the introduction of synonymous codons preferred by E. coli and coexpression of molecular chaperones2. In a medium-throughput bacterial genotoxicity assay, the orphan P450 2W1 exhibited broad substrate specificity in the activation of a spectrum of chemical carcinogens3, while P450s 2S1 and 4F11 showed negative results4. The orphan P450s 20A1 and 27C1 were assayed using several potential (targeted) substrates but no reactions were observed5,6. Similar trials with P450 4X1 showed oxidation of anandamide to 14,15-EET ethanolamide7.

Another strategy is untargeted searches with extracts of human tissues as sources of substrates. Incubation of recombinant P450s with liver extracts and 16O2/18O2 mixtures and comparisons of multiple LC-MS profiles with the program DoGEX identified a series of fatty acids as substrates for P450 4F11. This approach also identified a number of fatty acids for the known "xenobiotic" P450s 1A2, 2C8, and 2C98. These findings may reflect the sensitivity of fatty acids in the current LC-MS procedures. We have recently modified the method to increase the sensitivity in untargeted searches.

References:

Biography
Zhong-Liu Wu was born in 1975. After receiving her Ph.D. degree from Shanghai Institute of Organic Chemistry, Chinese Academy of Sciences in 2003, she joined Prof. F. Peter Guengerich’s research group at Vanderbilt University as a postdoctoral researcher. In 2007, she moved to Chengdu Institute of Biology, Chinese Academy of Sciences as a full professor. Her research is focused on the diverse chemical function and application of oxidoreductases, such as cytochrome P450s, epoxygenases, and related reductases.