THE INHIBITORY EFFECT OF POLYUNSATURATED FATTY ACIDS ON HUMAN CYP ENZYMES
Hsien-Tsung Yao, Yi-Wei Chang, Shih-Jung Lan, Chiu-Tong Chen, John T.A. Hsu, Teng-Kuang Yeh
Division of Biotechnology and Pharmaceutical Research, National Health Research Institutes, 35, Keyan Road, Zhunan Town, Miaoli County, Taiwan, R.O.C.

The inhibitory effect of saturated fatty acids (SFAs): palmitic acid (PA), stearic acid (SA) and polyunsaturated fatty acids (PUFAs): linoleic acid (LA), linolenic acid (LN), arachidonic acid (AA), eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA) on six human drug-metabolizing enzymes (CYP1A2, 2C9, 2C19, 2D6, 2E1 and 3A4) was studied. Supersomes from baculovirus-expressing single isoforms were used as the enzyme source. Phenacetin O-deethylation (CYP1A2), diclofenac 4-hydroxylation (CYP2C9), mephenytoin 4-hydroxylation (CYP2C19), dextromethorphan O-demethylation (CYP2D6), chlorzoxazone 6-hydroxylation (CYP2E1) and midazolam 1-hydroxylation (CYP3A4) were used as the probes. Results show that all the five examined PUFAs competitively inhibited CYP 2C9- and 2C19-catalyzed metabolic reactions, with IC50 (Ki) values ranging from 2.9 ~ 8.1 (1.7 ~ 4.7) µM and 4.4 ~ 15 (2.3 ~ 7.4) µM, respectively. Among these, AA, EPA and DHA tended to have greater inhibitory potencies (lower IC50 and Ki values) than that of LA and LN. In addition, these five PUFAs also competitively inhibited the metabolic reactions catalyzed by CYP1A2, 2E1 and 3A4 to a lesser extent (with IC50 values >30 µM and Ki values >10 µM). On the other hand, PA and SA, the saturated fatty acids, had no inhibitory effect on the activities of six human CYP isozymes at concentrations up to 200 µM. Incubation of PUFAs with CYP2C9 or CYP2C19 in the presence of NADPH resulted in the rapid metabolism of the fatty acids. These results indicate that the PUFAs are the potent inhibitors as well as the substrates of CYP2C9 and CYP2C19.

ROLE OF INFLAMMATORY MEDIATORS IN DOWN-REGULATION OF HEPATIC CYTOCHROME P450 IN THE RAT WITH TNBS-COLITIS
Yasuhiro Masabuchi1, Kanako Enoki2 and Toshiharu Horie2
1Laboratory of Clinical Pharmacy, Faculty of Pharmaceutical Sciences, Chiba Institute of Science, 15-8 Shioiri-cho, Choshi, Chiba 288-0025, Japan; 2Laboratory of Biopharmaceutics, Graduate School of Pharmaceutical Sciences, Chiba University 1-8-1, Inohana, Chuo-ku, Chiba 260-8675, Japan

Rats treated intracolonically with 100 mg/kg trinitrobenzene sulfonic acid (TNBS) dissolved in 30 % ethanol develop colitis. The colitis accompanies down-regulation of hepatic CYP3A2 and CYP2C11, and to a lesser extent, CYP2E1. In the present study, attempts have been made to protect against the down-regulation of hepatic P450s as well as against the development of colitis by using chemical agents to assess the role of inflammatory mediators in the liver effects. In the colitis model, higher levels of endotoxin, nitric oxide (NO) metabolites and interleukin-6, but not TNF-α appeared in the portal vein. Polymyxin B, which neutralizes endotoxin, partially prevented the decrease in CYP3A2 as assessed by testosterone 6β-hydroxylation activity in hepatic microsomes, without ameliorating colitis as assessed by myeloperoxidase (MPO) activity in colonic mucosa. Curcumin, which has anti-inflammatory properties, alleviated both the increase in MPO of the colon and the down-regulation of CYP3A2. Nimesulide, a preferential COX-2 inhibitor, which did not suppress the increase in MPO but the appearance of portal NO metabolites, protected rats against the down-regulation of hepatic CYP3A2. Similar results were obtained by gadolinium chloride, which inactivates macrophages. On the other hand, these treatments were less effective on the down-regulation of hepatic CYP2C11. It is suggested that endotoxin, cytokines and prostaglandins, which are possibly leaked from damaged colon and further activate Kupffer cells, are involved in down-regulation of hepatic CYP3A2 in the rats with TNBS-colitis.