Male Sprague-Dawley rat hepatocytes were plated onto Matrigel-coated dishes and incubated for 48 hr with Williams' Medium E supplemented with 0.25 U/ml insulin, 10^{-7} M triamcinolone acetonide and antibiotics. Cultures were then incubated for 24 hr with medium alone, or with medium containing either a prototypical P450 inducer or SQ1 (10^{-7} M to $3x10^{-5}$ M). After treatment, hepatocytes were harvested for estimation of P450 and HMG-CoA reductase mRNA levels by slot blot hybridization. Doses of SQ1 of $3x10^{-6}$ and lower were nontoxic to the cultured hepatocytes, as judged by cell morphology, total RNA recovery and MTT activity. SQ1 treatment increased the amounts of CYP2B and HMG-CoA reductase mRNA that were detected in the cultured hepatocytes, but had no effect on CYP1A1, CYP3A or CYP4A mRNA levels. The maximal increase in CYP2B mRNA level that was produced by SQ1 treatment was approximately 50% of that produced by treatment with 10^{-4} M phenobarbital, and occurred at the lowest dose (10^{-7} M) of SQ1 that was tested. This concentration of SQ1 is the same as that previously reported to produce maximum inhibition of cholesterol biosynthesis in isolated hepatocytes, and is within one order of magnitude of the concentration reported to maximally inhibit squalene synthase activity *in vitro*. Supported by NIH grant HL50710 and by NIEHS Center grant ES06639.

682 XENOBIOTIC-ENHANCED EXPRESSION OF CYP2E1 mRNA AND PROTEIN IN FGC-4 RAT HEPATOMA CELLS.

K J Woodcroft, J- Y Lee, and R F Novak. *Institute of Chemical Toxicology, Wayne State University, Detroit, MI.*

The FGC-4 rat hepatoma cell line was examined as a potential adjunct to primary cultured rat hepatocytes to examine mechanisms of xenobiotic-mediated regulation of CYP2E1 expression. CYP2E1 mRNA and protein expression were examined by Northern and Western blot analyses, respectively, in FGC-4 cells in response to chemicals known to enhance hepatic CYP2E1 protein expression *in vivo*, and in primary cultured rat hepatocytes. Treatment of FGC-4 cells for 24 h with 100 mM ethanol (ETH), isopropanol (ISO), acetone (ACE), 25 mM pyridine (PYR), or 50 mM pyrazine (PYZ) increased CYP2E1 protein levels ~ 4-, 6-, 12-, 8-, or 27-fold, respectively, compared to untreated cells. Corresponding CYP2E1 mRNA levels were increased ~ 3-, 2-, 4-, 5-, or 3-fold with ETH, ISO, ACE, PYR, or PYZ treatment, respectively. The increase in CYP2E1 mRNA expression monitored in FGC-4 cells is in contrast to the effects of these xenobiotics on CYP2E1 expression in primary cultured rat hepatocytes, in which 2E1 protein levels are increased by 2- to 8-fold with no corresponding increase in 2E1 mRNA. The increase in CYP2E1 mRNA expression in FGC-4 cells following treatment with these chemicals was both concentration- and time-dependent. mRNA degradation studies demonstrated that PYR-or PYZ-mediated elevation of 2E1 mRNA levels in FGC-4 cells was not a result of increased stabilization of CYP2E1 mRNA. These data show that CYP2E1 expression in FGC-4 cells in response to xenobiotics differs mechanistically from that observed in primary cultured rat hepatocytes, the latter paralleling that reported *in vivo*. These data demonstrate that FGC-4 cells do not constitute an alternative to primary cultured rat hepatocytes for mechanistic studies on regulation of CYP2E1 expression. Supported by NIEHS grants ESO3656 and P30 ESO6639.

683 DIMETHYL SULFOXIDE (DMSO) DECREASES CYP3A TURNOVER IN PRIMARY CULTURED RAT HEPATOCYTES.

R C Zangar and R F Novak. *Institute of Chemical Toxicology, Wayne State University, Detroit, MI.*

DMSO treatment of rats or primary cultured rat hepatocytes increased immunodetectable CYP3A protein levels in the absence of elevated CYP3A mRNA levels, indicating post-transcriptional regulation of this P450. In cultured cells treated with phenobarbital, microsomal CYP3A levels increased ~2-fold in response to the addition of 0.01% DMSO. CYP3A protein levels were increased ~3.5- and 5-fold following 0.1% DMSO treatment for 6 and 12 hr, respectively. Polysomal distribution analyses failed to show a shift in CYP3A mRNA in response to DMSO treatment, suggesting that increased translation was unlikely to be a major mechanism by which CYP3A protein content was elevated. In the presence of the cycloheximide, which inhibits protein synthesis, loss of CYP3A protein was substantially decreased in the presence of DMSO. CYP3A-catalyzed testosterone 2β-hydroxylase activity was measured in intact cultured hepatocytes after removal of DMSO-containing medium. The results showed that DMSO treatment also increased CYP3A metabolic activity. DMSO did not increase CYP2B expression in any of the studies undertaken. These results suggest that DMSO specifically increased CYP3A protein levels as a result of decreased protein turnover without permanent loss of enzymatic activity. Supported by NIH grants ES03656 and ES05657 and EHS Center Grant P30 ES06639 from the National Institutes of Environmental Health Sciences.

684 TIME-DEPENDENT DEPRESSION OF TOTAL CYTOCHROME P450 AND FORM-SPECIFIC ACTIVITIES IN MICE FOLLOWING INFECTION WITH THE MURINE RETROVIRUS, LP-BM5 (MAIDS).

S Shedlofsky[1], J E Snawder[2], R Tosheva[1]R Avidiushko[3], and D Cohen[3]. [1]*V.A. Medical Center, Dept. Of Medicine, University of Kentucky, Lexington, KY,* [2]*National Institute for Occupational Safety and Health/ Centers for Disease Control and Prevention, Cincinnati, OH and* [3]*Dept. of Microbiology and Immunology, College of Medicine, University of Kentucky, Lexington KY.*

The cytochrome P450 enzyme system is important in the metabolism of many endo- and xenobiotic agents, including many therapeutic drugs. We and others have demonstrated that the acute, LPS-induced inflammatory response and its associated cytokines play a role in the modulation of cytochrome P450. This report describes the effects of murine retrovirus LP-BM5 infection on total P450 and P450 form-dependent metabolic activities in mouse hepatic microsomes. Mice were inoculated with a mixture of LP-BM5 and sacrificed at 2, 4, 6, 8, 14 and 20 weeks post-infection to evaluate the effect of infection and progression to MAIDS on total P450, p-nitrophenol hydroxylase (PNP) and the dealkyation of benzyloxy-, ethoxy- and pentoxy-resorufins (BROD, EROD and PROD) in hepatic microsomes. The expression of mRNA coding for the cytokines, interferon (IFN), interleukins 1 and 10 (IL 1 and IL 10) and TGF was also examined in lung. Infection of mice with LP-BM5 produced a slight decreases in total P450 at 2 weeks post-infection, these minor losses were seen in all the activities examined. Total P450 and associated activities were not further altered until week 20; while EROD remained at 70% of controls, BROD, PROD and PNP in infected animals were only 40% of the control values. Lung-mRNAs for IFN, IL-1, IL-10 and TGF all increased after infection and peaked by week 8. TGF and IL-10 remained elevated out to week 20; IL1 and IFN slowly declined after week 8. This report describes the effect of MAIDS on hepatic P450 and its associated activities. If similar responses occur in humans with AIDS, careful monitoring of the efficacy and safety of administered drugs is warranted.

685 EFFECT OF PHENOBARBITAL, PREGNENOLONE-16α-CARBONITRILE AND ISONIAZID ON CYP450 ISOZYMES IN THE SPECIES *AMEIVA EXSUL*.

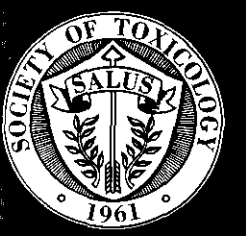
L Santos, A Hupka & G Winston[1]. *PSM, Puerto Rico & LSU, Louisiana*[1].

Ameiva exsul, a ground lizard common to Puerto Rico, has been tentatively established as a sentinel model of terrestrial areas of contamination. In our previous studies, β-naphthoflavone (βNF) was used as a potential inducer of CYP4501A1 and induction of this isozyme in the lizard was demonstrated. Further studies using different inducers have established the presence of other CYP450 isozymes in *Ameiva exsul*. The inducers include: phenobarbital, pregnenolone-16α-carbonitrile and isoniazid. Isozyme specific enzyme activities measured include: ethoxyresorufin-O-deethylase (EROD), pentoxyresorufin-O-deethylase (PROD), nitrophenol hydroxylase (PNPH), and erythromycin-N-demethylase (EMDM). Selective inhibition studies and Western blot analysis support results of P450 isozyme associated enzyme studies. Results of our studies indicate that CYP4501A1 is present in this species as CYP4501A2. CYP4502B does not seem to be present and some evidence suggests that a third isozyme from the subfamily CYP4501A might be present. Preliminary results also indicate that CYP4502E and CYP4503A might be present but not the forms usually associated with rats or humans.

686 EFFECTS OF 5-BROMO-2 DEOXYURIDINE (BrdU) ON HEPATIC CYTOCHROME P450 CONTENT AND β-OXIDATION ACTIVITY IN RATS AND MICE.

M Applegate, L Sulecki, and L B Biegel. *DuPont-Haskell Laboratory, Newark, DE.*

Standard FIFRA toxicity tests are frequently supplemented with analysis of various biochemical markers of toxicity. Haskell Laboratory routinely performs analysis of hepatic cytochrome P450 content, hepatic β-oxidation activity, (biochemical analysis) and cell proliferation rates. These analyses are typically conducted when past experience with a test compound or structural similarity to other compounds, which effect these parameters, suggest that



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Preface

This issue of *The Toxicologist* is devoted to the abstracts of the presentations for the symposium, platform, poster / discussion, workshops, roundtables, and poster sessions of the 36th Annual Meeting of the Society of Toxicology, held at the Cincinnati Convention Center, Cincinnati, Ohio, March 9-13, 1997.

An alphabetical Author Index, cross referencing the corresponding abstract number(s), begins on page 371.

The issue also contains a Keyword Index (by subject or chemical) of all the presentations, beginning on page 395.

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