Society of Toxicology

40th Annual Meeting

An Official Journal of the
Society of Toxicology
Supplement

TOXICOLOGICAL SCIENCES

Formerly Fundamental and Applied Toxicology

The Toxicologist

Oxford University Press

Volume 60, Number 1, March 2001

lung and olfactory tissues with minimal effect in the liver. Further, CYP2E1 protein expression increased in lung, olfactory, and liver tissues. To compare the effects of MAN and acetone, male F344 rats received a single acetone dose (5 ml/kg) by gavage. After 12 hr, acetone resulted in a significant increase in the levels of CYP2E1 mRNA and protein in nasal and lung tissues, with no obvious changes noted in the liver. These data suggest that treatment of rats with MAN causes increased expression of CYP2E1 in the lung and olfactory tissues, which may be caused by acetone and/or parent MAN. Previous whole body autoradiography studies also showed an early rapid uptake and persistence of MAN-derived radioactivity in the olfactory tissue for up to 24 hr after MAN administration. In conclusion, these results showed that MAN, similar to acetone, induces the expression of CYP2E1 at both the transcriptional and post-transcriptional levels in rat nasal and lung tissues. Further, increased expression of CYP2E1 in the liver of MAN-treated rats is apparently limited to post-transcriptional mechanisms. Induction of CYP2E1 in the olfactory tissue may increase the *in situ* metabolism of MAN causing the observed toxicity.

1928 CYTOKINE-MEDIATED SUPPRESSION OF P450 1A1 IN HEPA-1C1C7 CELLS BY POKEWEED MITOGEN.

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This study investigated the effects of pokeweed mitogen (PWM) on regulation of P450 1A1 expression in an *in vitro* model, using a murine hepatoma cell line Hepa-1c1c7 and a murine macrophage cell line RAW 264.7 cell cultures. The effect of PWM on P450 1A1 activity was measured by 2,3,7,8-tetrachlorodibenzo-p-dioxin (TCDD)-inducible P450 1A1-specific 7-ethoxyresorufin O-deethylase (EROD) in Hepa-1c1c7 cells. PWM added directly to Hepa-1c1c7 cells had no effect on the TCDD-inducible EROD activity; in contrast, TCDD-inducible EROD activity and P450 1A1 mRNA levels were markedly suppressed when Hepa-1c1c7 cells were incubated with PWM-treated RAW 264.7 conditioned media in a dose dependent manner. Concomitant treatment of PWM and pentoxifylline, a TNFa synthesis inhibitor, to RAW 264.7 cells decreased the suppressive effects of PWM on TCDD-inducible EROD activity. In PWM-exposed RAW 264.7 cell cultures, TNFa and IL-6 levels increased in a dose-dependent fashion. When treatment of antibodies to TNFa or/and IL-6 to PWM-treated RAW 264.7 conditioned media, the suppression of EROD activity was abolished. These results suggested the suppression of P450 1A1 by PWM was mediated exclusively by TNFa and IL-6, released from macrophages. [Supported by KOSEF Grant 1999-2-214-001-5 and RCPM]

1929 JP-8 JET FUEL INDUCES CYP2B1, CYP2E1, AND GSTPI BUT NOT CYP1A1 IN MURINE LIVER.

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JP-8 jet fuel is a complex mixture of aliphatic and aromatic hydrocarbons used predominantly in military and commercial airline environments. Recent reports have implicated jet fuel as an immune and respiratory toxicant. The purpose of this investigation was to determine if JP-8 modulates the expression of phase I and II hepatic enzymes to include CYP1A1, CYP2B1, CYP2E1 and GSTpi. B6C3F1 female mice were exposed to 1000 mg/kg/day of JP-8 or vehicle only (olive oil) by oral gavage for 7 days. After exposure to JP-8, protein expression of CYP1A1, CYP2B1, CYP2E1 and GSTpi metabolizing enzymes was determined by Western blot. Secondly, transient and persistent alterations in the expression of these enzymes were determined. From these studies, it was demonstrated that JP-8 increased the expression of CYP2B1 and CYP2E1 but not CYP1A1 in the liver microsomal fraction. It was also determined that protein levels of GSTpi (glutathione-s-transferase) were increased in the liver cytosolic fraction. The expression of these enzymes returned to control levels following a 7-day recovery. These results indicate that JP-8 modulates the expression of P450s and GSTpi but that recovery to control levels is complete and rapid. These findings may facilitate a greater understanding of the mechanisms of JP-8's pleiotropic toxicity and could be applicable toward the use of molecular biomarkers to determine exposure risk in military and commercial airline environments.

1930 INDUCTION OF MOUSE HEPATIC CYP2A5 IS ASSOCIATED WITH OXIDANT INJURY AND ER STRESS.

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Hepatic cytochrome P450 2a5 (Cyp2a5) is induced in mice during hepatitis and liver injury. While the regulatory mechanisms are unclear, recent studies suggest oxidative stress may play a role. We tested the hypothesis that induction of murine hepatic Cyp2a5 occurs as a consequence of oxidant injury targeted primarily to the endoplasmic reticulum (ER). Increases in Cyp2a5 mRNA levels by the hepatotoxin pyrazole were abrogated in mice pretreated with vitamin E. In contrast, glutathione depletion by buthionine sulfoximine markedly up-regulated Cyp2a5 in vivo. N-acetylcytsteine also prevented pyrazole-mediated Cyp2a5 induction in mouse hepatocytes in primary culture. To determine whether cellular stress was localized to the ER we examined the expression of the ER stress gene, glucose regulated protein 78 (grp78), following pyrazole treatment. Grp78 mRNA levels were maximally elevated 12 h following pyrazole treatment whereas Cyp2a5 mRNA levels peaked at 72 h. Immunohistochemistry revealed that pyrazole-mediated induction of Cyp2a5 and grp78 occurs concurrently in zone 3 hepatocytes where glycogen stores are deficient. In primary mouse hepatocytes, pyrazole increased grp78 and Cyp2a5 mRNA levels in a dose-related fashion. To determine whether Cyp2a5 induction was a direct consequence of ER stress, mouse hepatocytes were treated with the ER stress inducers thapsigargin, tunicamycin or the oxidized form of dithiothreitol (DTTox). Only DTTox resulted in Cyp2a5 induction suggesting that redox cycling is critical in Cyp2a5 regulation. These novel findings show that induction of a CYP is a direct consequence of oxidant stress targeted to the ER underscoring the unique regulatory control of Cyp2a5 expression during liver injury.

1931 CYP1A2 PROTECTS AGAINST METHEMOGLOBINEMIA PRODUCED BY 4-AMINOBIPHENYL.

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4-Aminobiphenyl (ABP) is a potent human urinary bladder carcinogen that is present in amounts greater than 100 ng per cigarette in side stream cigarette smoke. As with many hazardous chemicals, a metabolite of ABP is thought to be the ultimate carcinogen, and *N*-hydroxylation by CYP1A2 is believed to be responsible for the initial step in metabolic activation. After activation, metabolites of ABP have been shown to oxidize hemoglobin to methemoglobin (metHb) and produce adducts with hemoglobin and DNA. To evaluate the role of CYP1A2 in metHb formation, we dosed male and female *Cyp1a2(-/-)* and *Cyp1a2(+/+)* mice with topical ABP (10 mg/kg) and spectrally monitored metHb for 24 h. There were only modest sex-related differences in metHb formation. Although *Cyp1a2(-/-)* and *Cyp1a2(+/+)* animals accumulated similar levels of metHb measured 2 h post-exposure (39.5 ± 3.1% vs 34.4 ± 3.3% MetHb, respectively), differences at 24 h following treatment were observed in *Cyp1a2(-/-)* compared to *Cyp1a2(+/+)* mice (13.6 ± 1.6% and 6.7 ± 0.4% MetHb, respectively). Pretreatment of animals with dioxin, (15 µg/kg, 48 h pretreatment) to induce members of the aromatic hydrocarbon receptor gene battery, decreased metHb by approximately 50% in both *Cyp1a2(-/-)* and *Cyp1a2(+/+)* animals at both treatment times. We conclude that mouse CYP1A2 does not participate in ABP metabolism leading to metHb but rather protects against such formation. Participation of CYP1A2 in the formation of DNA adducts and carcinogenesis by ABP is currently being evaluated. As these data are analyzed, we will be able to estimate the relationship between metHb formation and ABP-induced DNA adduct formation. ***—Supported in part by NIH grants P30 ES06096 and RO1 ES06321.***

1932 THE REDUCTION OF CYP450 IN THE SWISS WEBSTER MOUSE BY METHYLPHENIDATE (RITALIN ™).

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There have been many case reports of drug interactions with methylphenidate (MPH) that indicate MPH is inhibiting one or more of the CYP450 hepatic enzymes. Therefore, the effect of MPH on the hepatic CYP450 content and catalytic activity of CYP1A2, CYP2E1 and CYP3A was studied. Male Swiss Webster mice were treated with a single i.p. dose of MPH and total hepatic CYP450 was determined. MPH decreased CYP450 in a dose-dependant manner. 50 mg/kg and 100 mg/kg MPH decreased CYP450 to 54.2 ± 0.2 % ($p<0.05$) and 32.4 ± 11.8 % ($p<0.01$) of control values respectively. The effect of MPH on various isoforms of CYP450 was then determined. CYP1A2, which is involved in the metabolism of caffeine, imipramine, tricyclic antidepressants (TCA) and R-warfarin was also decreased in a dose-dependant manner by MPH with 50 mg/kg and 100 mg/kg reducing catalytic activity to 40.5 ± 2.4 % ($p<0.05$) and 23.8 ± 0.1 % ($p<0.01$) respectively. CYP2E1, which is involved in the metabolism of paracetamol, halothane and isoflurane, was inhibited. 50 mg/kg and 100 mg/kg of MPH reduced catalytic activity to 53.1 ± 13.2 % ($p<0.05$) and 38.2 ± 9.3 % ($p<0.01$) respectively. CYP3A, which is involved in the metabolism of many of the benzodiazepines as well as some