2 to 144 hours after oral administration of 100 umoles/kg of sodium arsenite to adult female Sprague-Dawley rats. In rat liver the mean inorganic arsenic concentrations were 0.11, 2.15, 1.85, 1.86, 1.82, 0.68, 0.51, 0.20 and 0.10 ug/g at 0, 2, 4, 8, 16, 24, 48, 72 and 144 hours after arsenite administration, respectively. At these same time points, rat hepatic HO activities were 1.0, 10.8, 6.8, 16.3, 21.2, 5.2, 3.8, 0.8 and 0.5 times control HO values. At these time points 86, 46, 61, 66, 80, 89, 95, 97 and 98% of the total rat liver arsenic was present as DMA, an arsenite metabolite that does not induce HO. A hepatic concentration of 0.49 ug/g of additional inorganic arsenic was present before or at the time of HO enzyme induction. In rat kidney lower inorganic arsenic concentrations and less HO enzyme induction occurred after oral arsenite administration. This study links tissue dosimetry for arsenic with a biological effect (HO enzyme induction) in a pharmacokinetic and pharmacodynamic view of its action in two tissues that are targets for arsenic-induced carcinogenesis. (Disclaimer: This is an abstract of a proposed presentation and does not necessarily reflect EPA policy.)

1664 DIMETHYLARSINIC ACID EFFECTS ON SIX BIOCHEMICAL PARAMETERS IN B6C3F1 MICE.

S Ahmad, W A Anderson and K T Kitchin. *Environmental Carcinogenesis Division, NHEERL, US Environmental Protection Agency, Research Triangle Park, NC.*

Historically, methylation of the inorganic forms of arsenic (arsenate or arsenite) was interpreted as a detoxification pathway; however, recent data from free radical, biochemical and carcinogenic studies of dimethylarsinic acid (DMA) suggest that methylation of arsenic may be a toxification pathway instead. DMA has tested positive as both a complete carcinogen (urinary bladder and fibrosarcomas) and as a promoter of carcinogenesis (lung, liver, kidney, urinary bladder and thyroid gland). Adult female B6C3F1 mice were given 720 mg/kg of DMA by gavage at one of three points - 2 hours before, 15 hours before, and both 21 and 4 hours before sacrifice. We determined the DNA damage, the reduced and oxidized glutathione (GSH, GSSG) content, cytochrome P-450 content, ornithine decarboxylase (ODC) activity and plasma alanine aminotransferase activity. Significant ($P < 0.05$) decreases (15 to 37%) in liver GSH and GSSG contents were observed in all exposures to DMA. This data suggests that DMA can lower cellular defenses against electrophiles and free radicals. DNA damaging and eventually carcinogenic effects may be mediated by GSH depletion and/or the enhancement of oxidative stress. A general trend toward a DMA-induced DNA damage was observed, particularly in the liver ($P < 0.1$, one tail), but no comparisons reached the $P < 0.05$ level of statistical significance. Pulmonary and hepatic ODC activities were reduced (19 to 59%) by prior DMA treatment. ODC is involved in cell proliferation. Overall, our DMA mouse results are quite different from our prior rat results. Rats and mice differ greatly in DMA pharmacokinetics, largely because of large binding of DMA to cysteine 125 on the β chain of rat hemoglobin. These biochemical studies show that mice are much less responsive to DMA than rats. (Disclaimer: This is an abstract of a proposed presentation and does not necessarily reflect EPA policy.)

1665 TOUGHNESS OF SKIN IN ARSENIC EXPOSED MICE: ROLE OF GM-CSF AND TGFa.

T Yoshida[1], H Aikawa[1], H Yamauchi[2], K Sakabe[1], F Kayama[3], W Fujimoto[4], D R Germolec[5] and M I Luster[6]. [1]*Tokai Univ. School of Medicine,* [2]*Saint Marianna Medical College,* [3]*Jichii Medical College,* [4]*Okayama Univ. School of Medicine, Kanagawa, Japan and* [5]*NIEHS, NIH, RTP, NC. USA; and* [6]*NIOSH, Morgantown, WV, USA.*

Hyperkeratosis on the palms of the hand and soles of the feet are characteristics of arsenic (As) poisoning. However, toughness of skin has not been described as a manifestation of As poisoning. We previously reported that As induces GM-CSF and TGFa mRNA and protein production in human keratinocyte culture. In the present studies sodium arsenite was provided at 1 and 10mg/L to C57B/6n male and female mice via drinking water for 50 days. As concentrations were determined in hair and urine. Pieces of skin were excised from shaved ventral skin by scissors for histpathological and molecular analysis. Increase skin toughness dominantly in male was apparent in exposed groups when skin sections were prepared. Skin sections were prepared and stained for Haematoxilin and Eosin. GM-CSF in serum were measured by ELISA. RNA was extracted from skin and the expression of GM-CSF and TGFa mRNA were examined by RT-PCR. Although the epidermis appeared normal, the dermis in exposed animals showed moderate thickening of the collagen bundles. Seven of 12 exposed male mice and 4 of 12 female mice showed significantly thickened collagen bundles. GM-CSF in serum were elevated in 10mg/L, but not significantly. RNA expressions for GM-CSF and TGFa were enhanced moderately in exposed groups compared to controls. These results indicate that a number of changes occur in the skin of As-exposed mice, including thickening of collagen bundles and increased growth factor expression.

1666 PARTIAL PURIFICATION OF AN ARSENATE REDUCTASE FROM HUMAN LIVER AND CHARACTERIZATION OF ITS COFACTOR REQUIREMENTS.

T R Radabaugh[1] and H V Aposhian[2,3]. [1]*Committee on Genetics,* [2]*Dept. of Molecular and Cellular Biology,* [3]*Center for Toxicology, Univ. of Arizona, Tucson, AZ, USA.*

An arsenate reductase from human liver has been purified 200 fold using DEAE-Sephacel, Sephacryl S-200 HR, Activated Thiol Sepharose 4B and Reactive Brown 10. The starting liver cytosol reduced 860 pmoles arsenate/30 min/mg protein. The activity is associated with a heat labile protein which is greater than 30 kDa which required both a thiol and a heat stable cofactor less than 3 kDa in size. To determine the enzyme's thiol requirement, the ability of glutathione, thioredoxin and lipoic acid were tested to see if they would stimulate reduction of arsenate in the absence of other thiols. In the presence of enzyme, either glutathione or lipoic acid stimulated reduction but were less than 20 times as active as dithiothreitol in this respect. Thioredoxin did not stimulate activity. Mg^{+2}, Ca^{+2}, Cu^{+2}, Mn^{+2}, and Zn^{+2} did not stimulate enzyme activity. In addition, enzyme activity was not inhibited by the presence of 5 mM ethylenediamine tetraacetic acid. (Supported by Superfund Program NIEHS Grant #ES-04940 and NIEHS Center Grant #P30-ES-06694.)

1667 CHRONIC ARSENITE EXPOSURE INDUCES RESISTANCE TO ACUTE TOXICITY OF ARSENIC AND ITS METABOLITES AND CROSS TOLERANCE TO CADMIUM.

E H Romach[1] and M P Waalkes[2]. [1]*PAI, Durham, NC;* [2]*NCI at NIEHS, Research Triangle Park, NC, USA.*

Arsenic is a known human carcinogen. A previous study in our laboratory showed that chronic (>18 weeks), low dose (500 nM) exposure to arsenite induces malignant transformation in a rat liver epithelial cell line (TRL1215). This study sought to determine if the adverse effects of metals were altered in these transformed cells. Arsenite transformed cells (ATC) and untransformed cells (UTC) were exposed to arsenite, arsenate, monomethyl arsenate (MMA), dimethylarsenic acid (DMA) and cadmium for 24 hours and cell viability was determined. The LC_{50} for arsenite was 67 μM in ATC as compared to 26 μM in UTC. ATC were very tolerant to the acute toxic effects of arsenate (LC_{50} >500 μM) compared to UTC (LC_{50} 54 μM). After metallothionein (MT) induction with zinc (80 μM) pretreatment for 24 hours, UTC showed minimal changes in sensitivity to arsenite and arsenate (15% and 20% increase in LC_{50}, respectively) compared to ATC (155% and 826% increase in LC_{50}, respectively). Prior studies have shown that MT is hyperexpressible in ATC. DMA had a LC_{50} of 32 mM in ATC as compared to a LC_{50} of 7 mM in UTC. Differences in sensitivity to MMA were modest but significantly different between ATC (LC_{50} 68 mM) and UTC (LC_{50} 50 mM). Cellular accumulation of arsenic was 2.7-fold higher in UTC (9.5 ng/10^6cells) than in ATC (3.5 ng/10^6cells). Thus, UTC accumulate more arsenic than ATC and this may, in part, account for their increased sensitivity to acute arsenic toxicity. ATC also showed a cross tolerance to the toxic effects of cadmium. The LC_{50} of cadmium in ATC was 51% greater than that seen in UTC. The results of this study show that ATC are tolerant to the acute toxic effects of arsenite, arsenate, MMA, and DMA, and cross tolerant to cadmium. This tolerance may be based, at least in part, on reduced uptake of arsenicals.

1668 METALLOTHIONEIN-I/II NULL MICE ARE MORE SENSITIVE THAN CONTROLS TO CHRONIC ARSENIC TOXICITY.

J Liu[1], Y P Liu[2], and M P Waalkes[3]. [1]*PAI, Durham, NC;* [2]*Univ. Kansas Medical Center, Kansas City, KS;* [3]*NCI at NIEHS, Research Triangle Park, NC, USA.*

Metallothionein (MT) is low-molecular weight, sulfhydryl-rich, metal-binding protein. MT can protect against the toxicity of cadmium, mercury, and copper, but the role of MT in arsenic (As)-induced toxicity is less certain. To better define the ability of MT to modify As toxicity, metallothionein-I/II null (MT-null) and control mice were given repeated sc injections of As(III)

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