kappaBalpha, showed no change in expression levels. Taken together, these results suggest that exposure to low doses of propanil affects a multitude of important signal transduction proteins involved in T cell activation and IL-2 production. (Supported by NIH grant ES07512.)

1429 MICROARRAY ANALYSIS OF GENE EXPRESSION PATTERNS INDUCED BY IRRITANT AND SENSITIZING CHEMICALS.

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Chemical-induced dermatitis continues to be an important occupational health problem. Despite decades of investigation, the molecular mechanisms underlying chemical induced hypersensitivity and irritation remain unclear due to the complicated interplay between properties of different chemicals and the immune system. In this study gene expression patterns induced by Toluene Diisocyanate (TDI, IgE-inducing sensitizer), Oxazolone (OXA, cell-mediated hypersensitivity inducing sensitizer) or Nonanoic acid (non-sensitizing irritant) were investigated using gene microarrays. Female BALB/c mice were dermally exposed on the ears once daily for 4 consecutive days. On day 5 the lymph nodes draining the exposure sites were collected and used for mRNA extraction. For TDI and OXA exposures, the concentrations used induced similar quantities of mRNA in the draining lymph nodes. The extracted mRNAs were reverse transcribed into cDNAs and then *in vitro* transcribed into biotin-labeled cRNAs. The hybridization of labeled cRNAs to GeneChip Mu6500 oligonucleotide arrays (Affymetrix,CA) and scanning were conducted by Research Genetics, Inc. AL. Of the 6,500 genes on the arrays, there were 19 genes whose expression levels were significantly different between TDI- and nonanoic acid-treated samples, 18 genes between OXA and nonanoic acid samples and 33 genes between TDI and OXA samples. These include immune response-related genes, transcriptional factors, signal transducing molecules and Expressed Sequence Tags. Microarray analysis identified differentially expressed genes which can be further investigated by conventional methods. Candidate genes will be chosen for further evaluation following exposure to a panel of chemical sensitizers and irritants. Further studies will be conducted to define the functional roles of these genes.

1430 A STUDY ON AN EXPOSURE MARKER FOR SAFROLE.

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Safrole is a known animal carcinogen. It exists in sassafras oil, black pepper, ginger, and *Piper* betle inflorescence, etc. The latter is an unique ingredient of the Taiwanese betel quid. To assess the contribution of safrole in the induction of oral and liver cancer among the Taiwanese betel quid chewers, it is desirable to have an assessable biomarker for exposure. It has been shown that the main urinary metabolite of safrole is the glucuronide conjugate of 1,2-dihydroxy-4-allylbenzene (DHAB). DHAB standard was synthesized via a demethylation of eugenol. Its structure was confirmed by NMR and a ≥95% purity by NMR, TLC, and HPLC analyses. Urine sample preparation involved a 16-hour anaerobic hydrolysis with β-glucuronidase and extraction with ethylacetate. Quantitative analysis was accomplished by an isocratic HPLC on an ODC column with UV detection. The linearity of the standard curve (3.5 to 28 μg/mL) was good (r≥0.997), all relative errors were <±12%, and recovery was 70.2±6.7% (n=4). When male Wistar rats were given safrole in corn oil, at 0, 75, 150, and 300 mg/kg, both the first and second day urines showed a linear dose-response relationship of r=0.90 and 0.99, respectively. The method was then applied to 11 human subjects, 7 betel quid non-chewers and 4 chewers. The results showed that DHAB of the non-chewers ranged from 0.75 to 17.75 μg/mg creatinine (crn) with a median of 1.64. For the chewers, it was 1.47~12.16 with a median of 4.17 μg/mg crn, which was ~2.54 times of the non-chewer's. Thus, it is concluded that DHAB can be the biomarker of exposure to safrole. (Supported by grant DOH88-HR-802, ROC.)

1431 EFFECT OF GENETIC POLYMORPHISMS OF CYP2A6 AND 2E1 ON URINARY COTININE LEVELS IN JAPANESE.

T. Kawamoto. *University of Occupational and Environmental Health, Kitakyushu, Japan.* Sponsor: K. Toshihiro.

Among nicotine metabolites, urinary cotinine has been popularly used as an exposure biomarker for cigarette smoking. However, it is well known that there are individual differences in nicotine kinetics and metabolism. In order to clarify the effects of the genetic polymorphims of drug metabolizing enzymes on nicotine metabolism, urinary cotinine levels in Japanese students and office workers were studied in the relation with cytochrome P450IA1 (CYP1A1), CYP2A6, CYP2E1, aldehyde dehydrogenase 2 (ALDH2), N-acetyltransferase 2 (NAT2) and glutathione S-transferase M1 (GSTM1). As a result, cigarette consumption and homozygous CYP2A6 deletion significantly affect urinary cotinine levels. The genetic polymorpshim in 5'-flanking region of CYP2E1 can be also considered as a clue of individual differences in nicotine metabolism. In a smoking challenge test, cumulated urinary cotinine excretion in the homozygously CYP2A6 deleted individuals was about one seventh as compared to the controls that have CYP2A6*1 allele.

1432 HUMAN SKIN SENSITIZATION POTENTIAL, *IN VIVO*: GLUCOWATCH® BIOGRAPHER.

D. R. Wilson and S. J. Fermi. *Cygnus, Inc., Redwood City, CA.* Sponsor: W. G. Reifenrath.

The GlucoWatch® biographer, developed by Cygnus, Inc., is a frequent, automatic, and non-invasive glucose monitor intended to provide diabetics with the information needed to control their glucose levels. Sponsored by Cygnus, Hill Top Research, Inc. (Miamiville, OH) performed a clinical trial evaluating the potential of the active GlucoWatch® biographer to induce contact sensitization (allergic skin response) in 31 diabetic and 56 non-diabetic subjects via Repeat Insult Patch Test. The active test system delivered iontophoretic current (up to 0.32 mA/cm² for 6 minutes during each 20 minute glucose measurement cycle). When worn, the following components of the biographer contacted the skin surface at distinctly different regions; i.e., device edge, active glucose collection gel pad (with glucose oxidase), blank gel pad (without glucose oxidase), skin contact adhesive area, and sweat sensors. These skin regions were evaluated separately. There were nine 14-hour biographer applications in the induction phase, and (after a 13 to 16-day break) a single 8-hour application in the challenge. 99 subjects initially enrolled in the study; 12 subjects dropped out, (9 diabetics and 3 non-diabetics). None of the diabetics dropped out for reasons related to diabetes. There were no serious or unexpected adverse events during the study. No subject scored greater than a 2 (moderate) for erythema or a 2 (light) for edema during the Induction or Challenge Period. Conclusion: There was no indication of contact sensitization in either diabetic or non-diabetic subjects.

1433 BIOMARKERS OF INTERNAL DOSE IN WORKERS EXPOSED TO CHLORONITROBENZENE.

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Para-chloronitrobenzene (p-CNB) has multiple pharmaceutical and agrochemical applications. P-CNB acts as a haematotoxic agent following metabolic activation to the phenylhydroxylamine. The aim of this study was to determine the occupational exposure level, and the internal dose, of p-CNB in exposed factory workers, from the Peoples Republic of China. Eight hour time weighted average (TWA) workplace exposure levels (n=21) ranged from 1.604 - 78.26mg/m³ (mean: 7.97mg/m³, UK 8hr TWA limit=1mg/m³). Urine samples were collected from workers for the determination of recent exposure. HPLC-UV analysis of spot urines from exposed workers (n=43) revealed three metabolites; p-chloroaniline (p-CA), 2-chloro-5-nitrophenol (CNP) and N-acetyl-S-(4-nitrophenyl)-L-cysteine (NANPC). P-CA was detected in 30 (mean: 0.470μ mol/mmol creatinine, range: 0.140-2.47μ mol/mmol creatinine); CNP was detected in 41 (0.620μ mol/mmol, 0.006-4.779μ mol/mmol) and NANPC was detected in 42 (0.819μ mol/mmol, 0.158-2.574μ mol/mmol) of the exposed samples. Metabolites were not detected in non-exposed controls (n=10), within the detection limits of the assay (0.167μ mol on column). Blood samples were collected for the determination of cumulative exposure dose. Haemoglobin (Hb) was hydrolysed in base. The released p-CA was derivatised with pentafluoropropionic anhydride and quantified by GC-MS using single ion monitoring and negative chemical ionisation. The mean adduct levels from exposed (n=43) and factory controls (n=17) were 8.09pmol/mg Hb (range: 0.15-17.06pmol/mg Hb) and 1.70pmol/mg Hb (0.14-8.22pmol/mg Hb) respectively. The presence of p-CA was confirmed by GC-MS electron impact ionisation. No significant correlation was found between external exposure and markers of internal dose, or between urinary metabolite and Hb adduct levels. The urinary metabolites provided markers of short-term exposure, reflecting the inter-day variability in external exposure dose. The presence of Hb adducts and absence of urinary

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