of the test substance at 6 different concentrations. Wells containing anti-IgE and compound 40/80 were also tested for each animal as positive controls for IgE and non IgE mediated histamine release. Histamine release was reported as a percentage of the total cellular histamine content.
Although results did not show any clear indication of specific IgE mediated histamine release towards the test substance, the assay did show reproducible responses when anti-IgE and compound 40/80 were incubated with WBC's (histamine release up to 50% and 30% of total cellular histamine respectively). It is concluded that a valid method to detect IgE and non-IgE mediated histamine release from basophils has been developed for use in the cynomolgus monkey.

PS 303 EVALUATION OF PLASMACYTOID DENDRITIC CELL-BASED ASSAY TO DETERMINE CHEMICAL ALLERGENICITY.

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Human dendritic cells have been used to evaluate the allergenicity potential of chemicals and develop alternatives to existing animal models utilized throughout industry to monitor products for contact sensitization. Development of such non-animal alternative assay systems for hazard assessment directly addresses REACH (Registration, Evaluation, and Authorization of Chemicals) legislation. In this study, we investigated whether CD86 expression in plasmacytoid dendritic cells (pDC) can be used to identify contact allergens. Human DC were generated from CD34+ progenitor cells and the pDC fraction (CD123+/CD11c-) was harvested using FACS sorting. The pDC were exposed to an expanded list of chemical allergens (n=49) or irritants (n=42). Concentrations of each chemical that resulted in >50% viability as determined by FACS analysis of propidium iodide stained cells were used. Allergens were identified based on stimulation index (SI) calculated by the fold increase in CD86 expression. A preliminary prediction model was developed: materials with SI ≥1.5 in at least 50% of the pDC donors (n=2-5 donors) were labeled as allergens; materials with SI < 1.5 were labeled as non-allergens. Of the 91 materials tested, historical data for 71 materials were available from mouse local lymph node assay (LLNA) and human studies; these data were used to analyze the sensitivity, specificity, and accuracy of the pDC based assay system versus the LLNA method and human response. Evaluation of the pDC method resulted in sensitivity=95%, specificity=81%, accuracy=89%; for the same 71 materials, the LLNA gave sensitivity=85%, specificity=84%, and accuracy=85%. Thus, performance of the pDC was comparable to that of the LLNA. In conclusion, the pDC method appears to be a sensitive and specific predictor of allergenicity. The assay is advantageous because high throughput screening of chemicals is possible, donor-to-donor variation can be monitored, the cells are of human origin, and the assay is considerably more cost effective than the LLNA or other in vivo tests.

PS 304 THE SENSITIZATION POTENTIAL OF FURFURYL ALCOHOL.

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Furfuryl alcohol-based resins are commonly used as binding agents in foundry sand-moulds and as a corrosion inhibitor in mortar, grout, and cement. When mixed with foundry sand and exposed to heat or acid catalysts furfuryl alcohol initiates polymerization or "curing" of the sand. As the curing proceeds, it causes the sand to become dimensionally stable and furfuryl alcohol is vaporized. Furfuryl alcohol is considered by the EPA to be a high volume production chemical with over 1 million pounds produced annually in the United States. This coupled with numerous uses provide considerable potential for exposure of workers and the general public to furfuryl alcohol and furfuryl alcohol-based resins. The potential for exposure to furfuryl alcohol exists though pulmonary, oral, and dermal routes of exposure. It has been reported to be highly toxic in laboratory animals with exposure to higher concentrations producing signs of central nervous system depression, such as headache, drowsiness, nausea and vomiting. Although furfuryl alcohol was nominated and evaluated for carcinogencity potential by the NTP, studies evaluating immunotoxicity are lacking. Limited human exposure data reports a higher incidence of asthma in foundry mold workers exposed to furan resin, suggesting a potential immunological effect. These studies were executed to evaluate the immunotoxic potential of furfuryl alcohol following exposure including the dermal route.
Furfuryl alcohol was tested in a combined irritancy local lymph node assay (LLNA) that also examined irritancy. It was identified to be an irritant at high concentrations and a sensitizer with an EC3 value of 25.6% resulting in classification as a mild sensitizer. Significant increases were observed in the B220+ and IgE+B220+ cell populations in the draining lymph nodes after exposure to furfuryl concentrations of 25% and 75% respectively. No elevation in total serum IgE levels were observed after exposure to any concentration of furfuryl alcohol. These results suggest that furfuryl alcohol may function as a T-cell mediated sensitizer.

PS 305 IS ORTHO-PHTHALALDEHYDE A SAFE ALTERNATIVE TO GLUTARALDEHYDE?

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Although ortho-phthalaldehyde (OPA) has been recommended as an alternative to glutaraldehyde for the sterilization and disinfection of heat-sensitive medical equipment, its toxicity has not been thoroughly investigated. The present study was designed to evaluate the dermal irritation and sensitization potential of OPA. Results of the Epiderm Skin Irritation Test identified OPA as a dermal irritant and furthermore, demonstrated that OPA is a more potent skin irritant than glutaraldehyde. Consistent with the in vitro results, exposure to 0.75% OPA induced irritancy when evaluated in a combined irritancy local lymph node assay (LLNA) exposed to 0.75% OPA. A concentration-dependant increase in lymphocyte proliferation was observed after OPA exposure with a calculated EC3 value of 0.051%, classifying this chemical as an extreme sensitizer. IgE-inducing potential was evaluated by phenotypic analysis of draining lymph node cells and measurement of total and OPA-specific serum IgE levels in the mice. The 0.1% and 0.75% exposed groups yielded significant increases in the IgE+B220+ cell population in the lymph nodes while only the 0.75% exposed group demonstrated significant increases in IL-4 mRNA in the draining lymph nodes and total and OPA specific serum IgE levels. A significant elevation in OPA-specific IgG1 was also observed after exposure to 0.75% OPA. These results demonstrated the dermal irritation and sensitization potential of OPA in an animal model raising concern about the skin irritation and sensitization potential of OPA among healthcare workers who are potentially exposed to the chemical.

PS 306 COMPARISON OF CONTACT ALLERGEN-INDUCED GENE EXPRESSION CHANGES IN HUMAN PERIPHERAL BLOOD MONONUCLEAR CELL-DERIVED DENDRITIC CELLS AND THE DENDRITIC CELL SURROGATE CELL LINE MUTZ-3.

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Since the use of dendritic cell surrogate cell lines in the development of in vitro skin sensitization test methods holds several advantages over the use of peripheral blood mononuclear cell-derived dendritic cells (PBMC-DC), expression analysis of 29 potentially predictive genes derived using PBMC-DC were evaluated in the DC surrogate cell line MUTZ-3. A pilot set of 5 allergens (DNBS, 2-MBT, CAL, EUG, ISOEUG) and 3 non-allergens (SA, LA, SLS) were tested in two independent experiments in the MUTZ-3 cells. If a gene response was positive to allergen treatment or negative to a non-allergen treatment in at least one of the two experiments it was considered as positive or negative, respectively. The response of the cell line to treatment with the non-allergens was predominantly negative. In the MUTZ-3 cells the only 'false' positive gene expression changes (CCL23, IL3RA, and MRC1) occurred following treatment with salicylic acid. Overall the response of the cell lines to non-allergen treatment was more predictive than the response of PBMC-DC which had 12 genes with false positive responses to salicylic acid treatment and two false positive genes each with lactic acid and sodium lauryl sulfate. For the MUTZ-3 cells 6 genes failed to respond to any of the 5 test allergens. In comparison, 17 of the 29 genes were positive with all 5 allergens with PBMC-DC. This is not surprising since the gene list was developed based on the response of PBMC-DC. The best performing gene with the MUTZ-3 cells was CCL4 which was positive for all five allergens and negative for the three non-allergens. A number of other genes were positive in the MUTZ-3 cells with four of the five allergens and negative with all non-allergens: ARGHDIB, CD1E, CTSH, EPB41L2, S100A4 and SLAM. The MUTZ-3 cell line shows some promise as a DC-surrogate for use in a gene expression-based method. However additional testing with an expanded test set of chemicals will be needed.

PS 307 RECONSTRUCTED HUMAN EPIDERMIS INTEGRATING LANGERHANS CELLS (RHE-LC) RESPONSE TO CONTACT SENSITIZERS.

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The most common manifestation of immunotoxicity in humans is allergic disease resulting from industrial or environmental exposure to sensitizers. A range of different in vitro models have been used, in order to understand the mechanisms through which chemical allergens induce allergic contact dermatitis in humans, and to develop in vitro assays to assess the potential of a chemical to induce skin sensitization