## Assessment of the Sensitising Potential of Textile Disperse Dyes and some of their Metabolites by Loose-fit Coculture-based Sensitisation Assay (LCSA)

Sonnenburg, A., Ahuja, V., Stahlmann, R., Wanner, R. Institute of Clinical Pharmacology and Toxicology, Charité, Berlin, Germany

Introduction: Certain textile disperse dyes are known to cause allergic reactions of the human skin, such as allergic contact dermatitis and contact urticaria. However, by now only few quantitative data on the sensitising potential of these dyes exist. We have tested 3 disperse azo dyes (Disperse blue 124 - DB124, disperse red 1 – DR1, Disperse yellow 3 – DY3), 3 products of azo-cleavage of these dyes (ANT - 2-Amino-5-nitrothiazole (DB124), AAA - p-Amino-acetanilide and ApC - 2-Amino-p-Cresol (both DY3)) and one disperse antraquinone dye (Disperse blue 1 – DB1) to achieve data on their sensitising and irritative potential.

Therefore we used a loose-fit coculture-based sensitisation assay (LCSA) of primary human keratinocytes and of allogenic DC related cells to emulate the *in vivo* situation of the human skin. Sensitisation was determined by analysing the expression of the DC maturation marker CD86 by flow cytometry. Estimation of the concentration required to cause a half-maximal increase in CD86-expression allowed quantitative risk assessment. Furthermore we used 7-AAD (7-amino-actinomycin D)-staining to achieve data on cell viability and thus the irritative potential of the tested substances. The dyes were categorised as weak or strong irritating substances by estimation of the concentration required to devitalize 50 % of the examined cells compared to a zero control.

Results: DB1, ANT and AAA were tested up to concentrations of 100, 200 and 300 ÿmol/l, respectively, and showed no sensitising potential. All other substances were categorised as extreme sensitisers. DB124 showed the strongest sensitising potential, followed by DY3, DR1 and ApC. The irritative potential correlated with the sensitising potential. We observed most pronounced cytotoxic effects for DB124. DY3, DR1 and ApC also turned out to be highly cytotoxic substances, whereas ANT and DB1 showed only weak irritative potential. AAA did not show any cytotoxic effect at the concentration range tested.

Conclusion: The LSCA proved to give adequate results for the sensitising potential assessment of coloured substances. In addition we were also able to achieve data on the irritative potential in the same series of tests. Hence the LCSA provides a stable test system to simultaneously analyse two crucial properties of substances relevant for allergy induction.