

DPRA-MS for raw materials and complex mixtures

All chemicals used in consumer products can cause skin sensitization leading to Allergic Contact Dermatitis (ACD).

The molecular basis of chemical allergy resides in the ability of an allergen (hapten) to modify endogenous proteins, by chemically bonding with their reactive functional groups.

The OECD Test Guideline N°442C, describes the Direct Peptide Reactivity Assay (DPRA) which addresses the human health hazard endpoint skin sensitisation, following exposure to a test chemical. Skin sensitisation refers to an allergic response following skin contact with the tested chemical, as defined by the United Nations Globally Harmonized System of Classification and Labelling of Chemicals (UN GHS).

Principle: Raw materials are incubated (24h) with synthetic peptides containing either cysteine (1:10) or Lysine (1:50)

Method : LC-MSMS that will specially assay the reactive peptides (P-CYS and P-LYS)

Results: Percentage depletion of cysteine and lysine relative to controls and classification of the substance (Sensitizers or non-sensitizers)

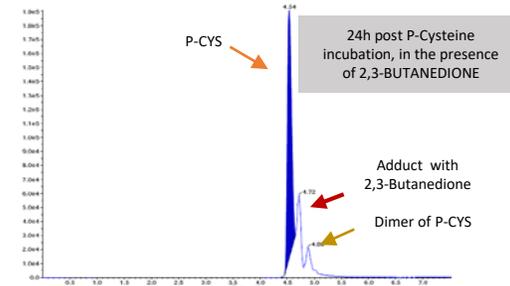
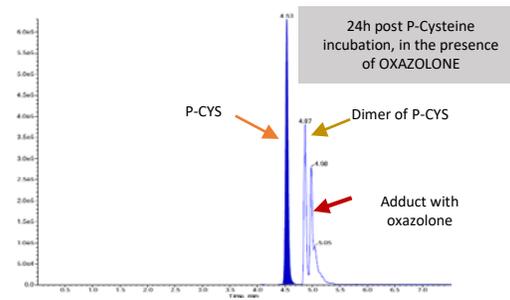
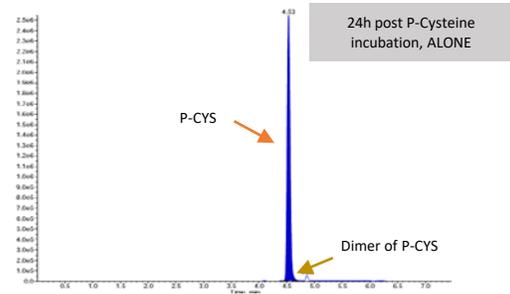
In Vitro Toxicity

DPRA : LC-MSMS Advantages over LC-UV

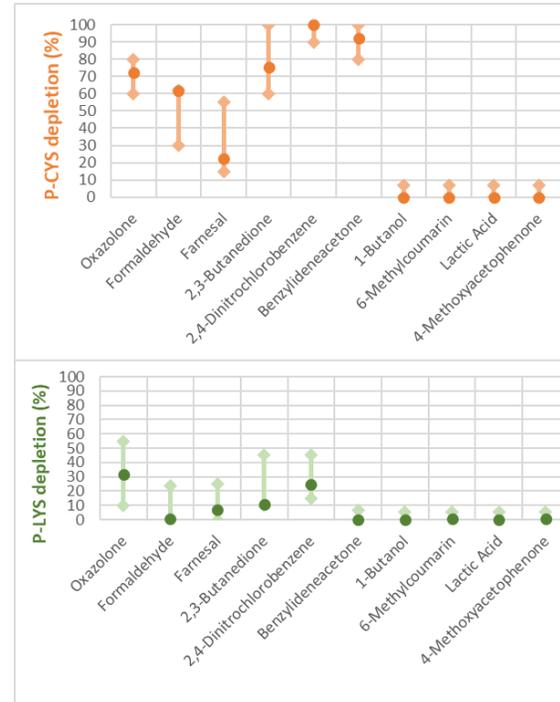
Even though initiated with LC-UV (220 nm), the employment of LC-MS by its specific measurement of the test peptides avoids misleading conclusions:

- No Co-elution issues to distinguish the native peptide from potential
- Distinguish CYS-peptide reactivity vs. oxidation leading to CYS-Peptide dimerization
- Decrease the risk of misleading conclusions
- Get information concerning the sensitization potential of solvent soluble complex mixtures

Accuracy of detection, using Mass Spectrometry, independently to sample complexity



The 10 reference chemicals for passing the DPRA proficiency (OECD 442C TG) P-CYS and P-LYS



Acceptance criteria using LC-UV
Proficiency results using LC-MSMS

The 10 reference chemicals were accurately classified according to the OECD Test Guideline N°442C

Applications

A- PURE RAW MATERIAL TESTING WITH A DEFINED M.W.

Application of the current OECD Test Guideline N°442C, with one accepted deviation : The use of MSMS detection instead of LC-UV detection, justified by:

- Scientific relevance of LC-MSMS specificity to at least supersede the LC-UV
- Methods validated for both reactive peptides for recommended solvents.

B- APPLICATION TO COMPLEX MIXTURES OR BOTANICAL EXTRACTS

In the particular case of complex mixture, which is mentioned in the TG has not led to a formal way of evaluating these substances due diversity of these mixtures. However, some recommendations are provided, serving as roots for a safety evaluation. Hence, the study plan has to be adapted to the test item. The assay performed in agreement with the study plan to provide a percentage of depletion for both peptides.

Offers And Services

- DPRA-MS tests for raw materials, including solvent soluble mixtures
- GLP compliance or screening format