PROGRESS

evic international group

EVIC international Neuronal net's - No. 16

Assessment of the allergic potential without animal testing

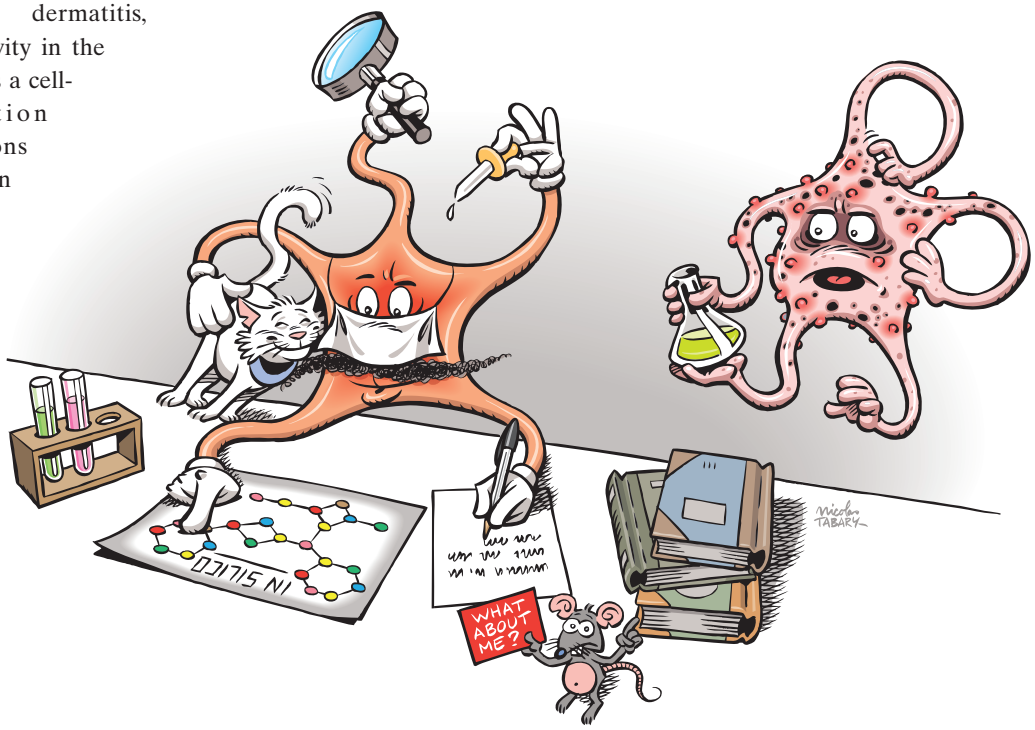
In the cosmetic field, the end of the transitional period for the implementation of the new European regulation (11 July 2013) leads to the final ban of animal testing, link hitherto considered as essential for the safety assessment of cosmetics as constituent ingredients. One consequence is the inability to assess by this way the adverse effects that may result from bioaccumulation after repeated contacts. This affects directly the investigation of allergenicity in cosmetic products, a main potential risk which involves a long process whose impacts are just perceived at the end of a long induction phase.

In addition, this sector decision is in contradiction with other regulatory requirements which concern number of raw materials used by the cosmetic industry. Indeed REACH program and CLP Regulation impose ranks of risk based on experimental data that can just currently be established using laboratory animals. This is particularly the case when differenciations have to be made between non-sensitizing and sensitizing substances or strong and slight sensitizers.

The resort to new methods of investigation of the allergic potential is essential and remains a high priority in the cosmetic field where this risk is considered to be in constant increase, the evolution being faster in the more developed populations.

What do we mean by delayed contact allergy?

The allergic contact dermatitis, classified as type IV hypersensitivity in the classification of Gell & Coombs, is a cell-mediated delayed reaction characterized by eczema lesions appearing in contact with certain substances. This reaction takes place in two stages including an efferent phase of sensitization and an efferent phase of revelation. The prerequisite for triggering an allergic contact reaction is the penetration of a substance through the skin, implying most commonly small size substances and sufficient liposolubility to cross the epidermis. To ensure the substance becomes allergenic it is important that it can bind to proteins (including membrane proteins).



Any substance in contact with the skin (hapten) does not therefore have the physicochemical structure causing it to become an allergen. So to simplify we note that the induction of allergy is caused by penetration of the hapten in the skin which became allergen by its uptake by Langerhans cells, its presentation to T lymphocytes cells, their proliferation and their spreading after implementation of a memory mechanism (induction phase).

When occurs a new contact with the allergen, it will be resubmitted to the Langerhans cell and T lymphocytes cells but the encounter will be directly on site in the dermis causing immediate expression of an lymphocyte mediated inflammatory reaction, resulting in a visible manner by the appearance of eczema (erythema, edema, vesicles), sometimes relocated from the point of contact (revelation phase). The induction phase usually lasts 5 to 10 days while triggering phase appears within 24 to 48 hours.

Historically how the sensitization potential was investigated?

The expression being dependent on the reacting ability of the immune system which is individual dependent, this type of investigation is necessarily limited to the study of the probability of occurrence of eczematous reaction resulting from prolonged contact of a given population with the possible allergen. This means that while triggering an allergic reaction indicates the presence of an allergen, the lack of response to the test does not allow a definitive conclusion. Experimental methods successively implemented attempted to reduce this interindividual variability, that has been possible through the use of sensitive and genetically close models. The first traditional methods using Guinea pigs (Buehler, Magnusson and Kligman tests) have been refined by methods attempting to quantify this effect (rLLNA and LLNA tests).

These methods have been successfully applied to raw materials and finished products. Most often, the absence of any sensitization induced in animals allowed subsequently its confirmation in humans ethically by means of challenge test today universally accepted (HRIPT): this is a final test of confirmation of the absence of allergic potential which remains valid.

Do we have models able to replace the re-orienteering tests performed up to now in animals?

The disappearance of tests using laboratory animals supposes to argue differently and gather a variety of elements providing the same level of information to the safety assessor. Two *in silico* approaches which are complementary are now used:

- The first approach is to study the physico-chemical profile of the substances likely to be in contact with the skin. It is particularly interested in the structure-activity relationship of molecules (QSAR).
- The second one aims to establish from the acquired data, the thresholds at which the allergenic potential of a substance can be expressed (TTC).

Meanwhile, three *in vitro* methods are in the final stages of validation now promising since they directly integrate the process of induction of allergic reaction.

- The DPRA test is based on a model purely chemical exploiting the fact that chemical allergens have electrophilic properties and are able to react with amino acids thus enabling to quantify the reactivity into four levels of risk.
- The MUSST test uses myeloid U937 human cells enabling to highlight the sensitizing potential by increasing the dose / effect of the expression of the CD86 marker in non-cytotoxic doses.
- The H-CLAT test is based on the use of THP1 cells relied on the change of the expression of the cell membrane markers. Sensitization potential is identified when the expression of receptors exceeds a certain threshold relative to the control.

All these means should quickly be able to replace traditional models.

However, in spite of these temporary difficulties there is no real problem; safety assessors in place should be able a priori to support their opinion based not only on pre-acquired data but also on all the information available (not just restricting to the toxicological one) to measure at the same time the intrinsic hazard of raw materials as well as the potential risk resulting from their incorporation into cosmetic formulations.

For lack of other robust historical data, results accumulated with traditional models will continue to be the reference for a long time especially for the evaluation of allergic reactions.

However the question arises since now for raw materials from new technologies or those of natural origin, more and more numerous whose compositions are still largely unknown. But then again in anticipation of the implementation of the promising alternative methods, the practical experience of the safety assessor should allow the needed overlaps to assure the consumers safety.

Next topic:

PROGRESS

EVIC international

122 rue Croix de Seguey
33000 Bordeaux - France
Tel : +33 5 56 95 59 95 - Fax : +33 5 56 95 05 22
E-mail : evicinternational@evic.fr
www.evicinternational.com

Editor: Philippe Masson - Concept: Tapez Dièse, Bordeaux
Printed on April 2013