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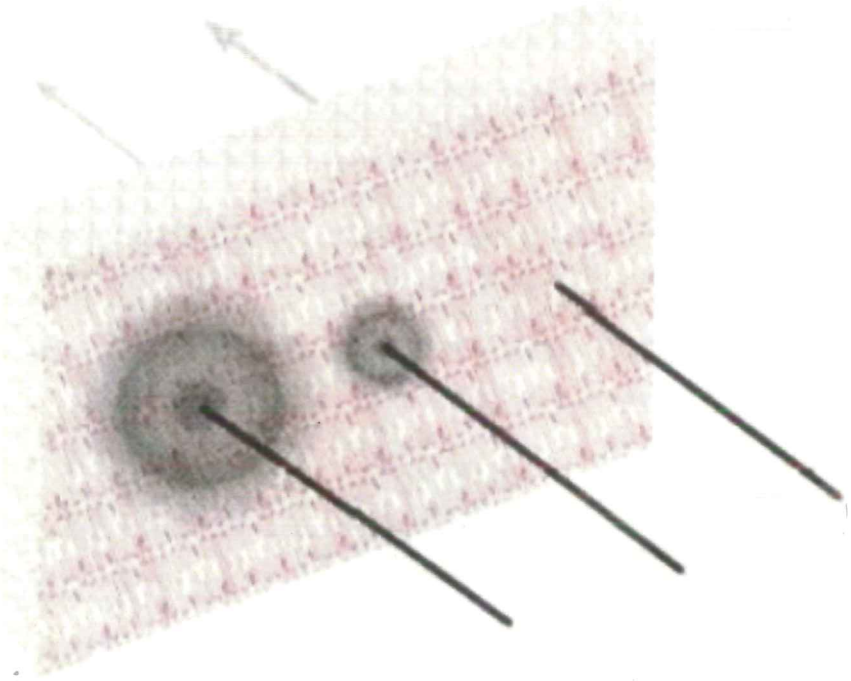

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# IRRITATION ASSESSMENT THROUGH HISTOPATOLOGY AND TEWL ON FRESH HUMAN SKIN OF THE PESTICIDES ISOPROTURON AND BENTAZON: AN ALTERNATIVE METHOD

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Traditionally, skin irritation assessments have been based on topical application of the test substances to rabbit skin (Draize test) but its reproducibility have been questioned. In addition, new European regulatory requests have pushed for advancements in alternative test methods to replace the current animal test for skin corrosion and irritation. It is in this respect that the emergence of new tools such as *in vitro* methods offers a promising future.

In the present work, an *in vitro* skin irritation test based on human skin from plastic surgery has been developed. The equipment used to measure the penetration through viable human skin was a flow through diffusion cell system (Franz cells). Two test-substances (Isoproturon and Bentazon), at different concentrations were applied topically to the epidermis surface at 32°C after the cell viability was controlled by measuring the trans epidermal water loss (TEWL). After 3h, we assessed the cell viability and examined the skin samples for histological features (normal appearing epithelium except slight thickening). The positive control was 10% SLS and the negative control was NaCl 0.9% during 3h.

The two herbicides chosen as test substances were: Isoproturon (3-(4-isopropylphenyl)-1,1-dimethylurea; CAS no. 34123-59-6), and Bentazon (3-isopropyl-2,1,3-benzothiadiazine-4-on-2,2-dioxide; CAS no. 25057-89-0). Both herbicides are frequently used in agriculture to kill weeds. These two substances are currently not classified as irritants but cases have been reported as such in the scientific literature.

Our results indicate no change in TEWL and histopathology after 3h of human skin exposures, even at high concentrations (double-blind controlled).

We conclude that Isoproturon and Bentazon do not cause irritation to human skin after 3h of exposure in this test system. Our next step is to assess a new apoptosis marker based on our histopathologic samples. These first results indicate that *in vitro* assays using fresh human skin is a promising alternative to *in vivo* rabbit skin irritation tests for the assessment of skin irritation.

**Acknowledgements**
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**References**

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