Transferability of the GARDskin assay to two independent laboratories

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Introduction

The Genomic Allergen Rapid Detection (GARD) skin assay is an *in vitro* assay for the assessment of skin sensitizers. The assay is based on gene expression analysis of SenzaCells, a human myeloid cell line, test substance exposure.

GARDskin is currently in a ring trial (OECD TGP 4.106). Here, we present the data from the initial transfer study that involved two laboratories independent from the development laboratory.

Conclusions

Two test laboratories were able to classify the eleven tested substances correctly in three independent transfer experiment studies. This paves the way for a successful final validation of GARDskin where 28 blinded chemicals currently are tested by three independent laboratories.

The GARDskin assay

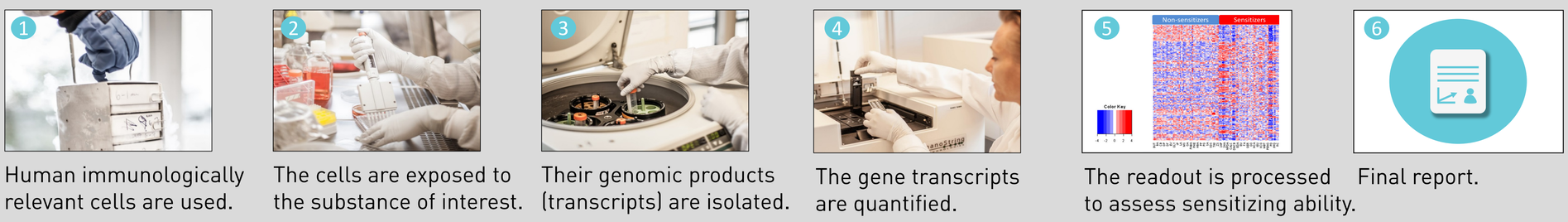

Figure 1. The dendritic cells (DCs) are sentinels of the immune system during sensitization. The GARDskin assay is based on SenzaCells, a human myeloid cell line similar to DCs. The assay protocol includes stimulation of the cells by the test substance and after incubation the transcripts are harvested. The NanoString technology is used for quantification of 200 genes. A bioinformatics multivariate technology is used for gene expression analysis.

Study design and result

Table 1. List of selected test substances and controls used in the ring trial.

Test substances	Sensitizer (Y/N)
DNCB	Y
p-Phenylenediamine (PPD)	Y (pos ctrl)
2-Aminophenol	Y
2-Nitro-1,4-PPD	Y
2-Hydroxyethylacrylate	Y
Resorcinol	Y
Geraniol	Y
Hexyl cinnamaldehyde	Y
Chlorobenzene	N
1-Butanol	N
DMSO	N (neg ctrl)

Eleven chemicals (Table 1) known to be sensitizers or non-sensitizers were analysed according to the GARDskin SOP. The assay was repeated three times at two contract research laboratories independent from the developing laboratory.

P-Phenylenediamine and DMSO was used as positive and negative controls respectively.

All chemicals, including control (11/11) were predicted to their correct class (sensitizer/non-sensitizer) generating an accuracy of 100% (Figure 2) in both laboratories in all three experiments.

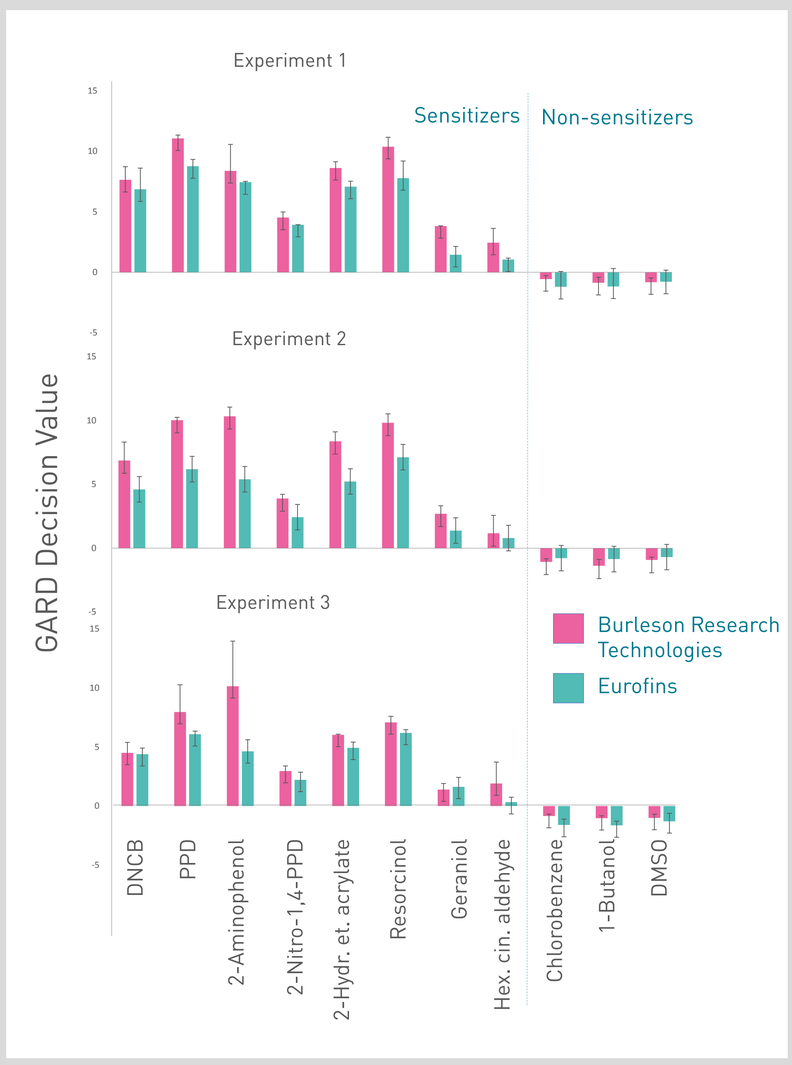

Figure 2. Decision values (DVs) of the 11 chemicals. DV>0 = sensitizer, DV<0=non-sensitizer. The error bars represent one standard deviation of three replicates.

References: Johansson *et al.* ALTEX, 2017, Johansson *et al.* BMC Genomics, 2011, Forreryd *et al.* BMC Genomics, 2014, Johansson *et al.* Toxicol Sci, 2014.

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