GARD® – the future of sensitization testing and safety assessment of chemicals, using a genomics-based platform

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BACKGROUND

Genomic Allergen Rapid Detection, GARD, is an *in vitro* test developed for the prediction of sensitizing chemicals. It is based on differential expression of disease-associated genomic biomarkers in a human myeloid dendritic cell line.

Here, we describe the development, scientific validation, applications and the current state of the GARD platform. The scientific rationale behind the use of genomic biomarker signatures are detailed, linked to the AOP in a biological context, and to advantages realized through multivariate computational prediction models in a technological context.

RESUME

The GARD assay is elastic, it can be used for several applications. Today, two are developed:

✓ GARDskin®
- Hazard identification of skin sensitizers
- Accuracy: ~90%
- Initiated ECVAM validation (OECD TGP no. 4.106)

✓ GARDair®
- Hazard identification of respiratory sensitizers
- Accuracy: ~85%

THE GARD PLATFORM

In vivo, dendritic cells are involved in initiating the immune response that occurs during sensitization. Therefore, a human myeloid cell line, similar to dendritic cells, is used for the GARD assay. The cells are stimulated with the compound of interest and after incubation the transcripts are harvested. The gene expression is analysed through a bioinformatics multivariate technology (1).

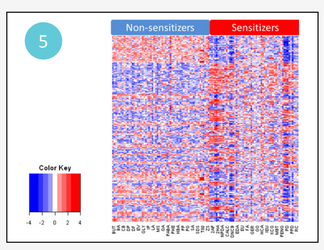
A brief overview of the GARD process

Human immunologically relevant cells are used.

The cells are exposed to the substance of interest.

Their genomic products (transcripts) are isolated.

The gene transcripts are quantified.



The readout is processed to assess sensitizing ability.

Final report.

During development of the GARD applications, total gene expression is analysed by microarrays, but after gene signature establishment, assays are transferred to the Nanostring platform (2).

Depending on requested end-point data the GARD platform can be transformed to different applications .

GARDskin®

The original GARD application assesses skin sensitization of chemicals and is denoted GARDskin. Regarding The platform cell line is stimulated with a chemical of interest and the gene expression of ~200 genes are analysed by a multivariate bioinformatics test method (1). The accuracy of GARDskin is assessed to ~90% (3), which can be compared to the traditional Local Lymph Node Assay (LLNA) with an accuracy of 72%. Based on additional genomic biomarkers relevant for skin sensitization the possibility to identify potency categories are suggested (4, manuscript in preparation).

GARDair®

An expansion of the GARD platform is the GARDair application, which assesses respiratory sensitizers. The method is unique and highly required, as predictive assays for assessment of respiratory sensitizers are greatly underdeveloped, with no validated, or even widely accepted, *in vivo* or *in vitro* method currently in use. The application is similar to the GARDskin application but another genetic profile consisting of ~400 genes is analysed. The assay was established with 10 chemicals of interest and in-house validated with another 25 chemicals. The accuracy for GARDair is assessed to ~85% (6).

VALIDATION

The importance of mature and reliable alternative test methods have to be emphasised. GARDskin is included in the OECD test guide line programme (TGP no. 4.106) and an ECVAM ring trial is set up at two external test laboratories to confirm the reproducibility. A method evaluation on chemicals provided by Cosmetics Europe Skin Task Force has been executed and the results are confirming that accuracies are constantly high (manuscript in preparation).

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