SKIN SENSITIZATION TEST

GARD™skin—200 genomic biomarkers

[OECD TGP 4.106]

The GARD™ technology platform

GARD™ – Genomic Allergen Rapid Detection – is a unique and highly versatile genomic-based *in vitro* testing platform for assessment of various toxicological endpoints. GARD™ makes use of unique genomic biomarker signatures comprising genes involved in pathways known to be relevant to the toxicological outcome. Supported by modern technologies for pattern recognition, the large informational content provided by this approach enables mechanism-of-action based decisions, which becomes evident in a consistently high predictive accuracy.

GARD™ skin – Highest accuracy on the market

GARD™skin is based on a dendritic-like cell line called SenzaCells and classifies test compounds as either skin sensitizers or non-sensitizers based on the readout from a genomic biomarker signature of 200 genes. The assay exhibits the highest accuracy on the market, well-balanced between high sensitivity and high specificity.

	DPRA (TG 442C)	ARE-NRF2 (TG 442D)	h-CLAT (TG 442E)	2 out of 3 ITS	GARD (TGP 4.106)
Accuracy	80%	83%	77%	83%	88%
Sensitivity	78%	84%	80%	84%	90%
Specificity	83%	78%	67%	78%	83%

Figure 1. GARD™skin exhibits the highest accuracy for hazard identification of skin sensitizing chemicals on the market. Results are based on published data from a retrospective meta-analysis performed on a coherent dataset (n=69) of chemicals tested in all assays. *Roberts D.W, Regulatory Toxicology and Pharmacology, 2018.*

GARD™skin is in the OECD test guideline programme

In a formal validation study performed in compliance with OECD guidelines, GARD™skin demonstrated excellent reproducibility as well as outstanding predictive performance. A full validation report of the results has been submitted to ECVAM for scientific review.

Accuracy	82%
Sensitivity	93%
Specificty	96%
Within Laboratory Reproducibility	82-89%
Between Laboratory Reproducibility	92%

Figure 2. GARD™skin is progressing towards formal OECD approval. The table summarize results from a validation study involving the blinded classification of 28 test substances in three laboratories. So far, no other test, *in vitro* nor *in vivo* has shown such high accuracy.

Available GARD™ assays

GARD™skin: *In vitro* classification of skin sensitizers with outstanding accuracy.

GARD™potency: First-in-class assay for CLP/GHS classification of skin sensitizers.

GARD™air: First-in-class assay for specific identification of respiratory sensitizing chemicals.

GARD™skin performance

GARD™skin alone outperforms each of DPRA, ARE-Nrf2 luciferase or h-CLAT, alone or in any combination as a 2 out of 3 strategy, in terms of sensitivity, specificity and accuracy.
Prof. David Roberts

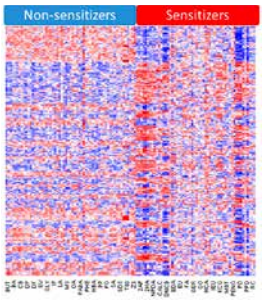
GARD™skin for REACH dossiers

The REACH Regulation allows the use of non-adopted *in vitro* methods in case they meet the EURL ECVAM criteria for entering pre-validation. For the GARD™ assay this criterium is met.
ECHA, official statement

GARD™skin enables a robust way to meet the demands for 3R with reliable outcome as the test is performed *in vitro* on a human cell line.

How is GARD™skin the most accurate assay on the market?

The 200 genes in GARD™skin are well-characterized and describe many diverse processes associated with skin sensitization. In contrast to the limited mechanistic information obtained from single biomarkers, the large informational content of this approach provides a more holistic view of the immunological response to skin sensitizing chemicals, which is reflected by its high predictive accuracy.



The genomic biomarker signature in GARD™skin covers several mechanistic events of sensitization.

How to GARD™ your products in six steps:

1. **GARD™ Input Finder:** Dose-response to find the GARD™ input concentration where 90% of SenzaCells survive.
2. **GARD™ Main Stimulation:** SenzaCells are exposed at the GARD input concentration.
3. **RNA extraction:** Following 24h of cellular stimulations, total RNA is isolated using an RNA extraction kit.
4. **Gene expression profiling:** Gene expression of the 200 genes in the biomarker signature is measured using NanoString.
5. **GARD™ data analysis application:** Gene expression pattern is compared to patterns of reference samples in an equation developed using a machine learning algorithm.
6. **Results:** Report provided on test substance classification as sensitizer or non-sensitizer (Y/N).

About SenzaGen

SenzaGen is dedicated to the development of innovative *in vitro* methods for safety testing of various toxicological endpoints across different industries. SenzaGen performs GARD™ in its own laboratory and through CRO partners around the world.

Contact us

EUROPE. Andy Forreryd, Scientific Liaison MGR
andy.forreryd@senzagen.com

Americas. Joshua Schmidt, Business development MGR
Joshua.schmidt@senzagen.com

APAC & ROW. Julia Huijie Li, Business Development Coordinator
julia.li@senzagen.com

GARD™skin, applicable for:

- Agrochemicals
- Cosmetics
- Industrial chemicals
- UVCB:s
- Natural extracts
- Screening of pharmaceutical candidates
- "Difficult-to-test" compounds and more...

GARD prediction signature

Recognition of foreign substances e.g. TLRs, RXR, AGR

Immunological self-defence mechanisms e. g. CD80, CD86

Cellular stress responses e.g. NRF2-pathway

Communication e.g. chemotaxis receptors

GARD™skin: At a glance

Measures: Genomic readout of 200 genes relevant to sensitization. Mechanistic coverage of several KEs in the AOP.

How: State-of-the-art machine learning tools used to develop the model. Transparent Y/N classification; no expert judgement required.

Performance: Accuracy > 90%, false positives and false negatives typically < 5%.

Reproducibility: Low variation within and between laboratories.

Compliant: OECD TGP 4.106 and accepted by ECHA for REACH dossiers.

Turnaround time: 4-8 weeks.

Sample requirements:0.5 g (solids) or 1 ml (liquids). Can be adapted to lower amounts.