Regulatory approval of medical devices according to MDR using *in vitro* data from GARDskin Medical Device for skin sensitization assessment

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Summary

- The GARD®skin Medical Device assay is a novel method for assessment of skin sensitizing properties in leachables from solid materials, according to ISO 10993 standards.
- Here, we demonstrate how GARD®skin Medical Device data, combined with endpoint data from skin irritation and cytotoxicity testing, were successfully used in a data package for biocompatibility assessment of a novel medical device.
- Following review by a notified body, an obtained CE-mark for the medical device Tinearity® G1 highlights unique opportunities to comply with the European Medical Device Regulation 2017/745 (MDR) using only *in vitro* data sources.

Introduction

Historically, safety evaluation of potential skin sensitizers in medical device extracts relied on *in vivo* tests like the Guinea Pig Maximization Test and the Buehler Occluded Patch Test. However, a shift to *in vitro* methods for the biocompatibility evaluation of medical devices is occurring, facilitated by the inclusion of such methods in the ISO 10993 standard series including skin irritation (10993-23) and skin sensitization endpoints (10993-10).

The GARD®skin assay (OECD TG 442E[1]) is an *in vitro* method for the assessment of skin sensitization and is included in the updated version of the ISO 10993-10:2021. The assay involves a biomarker signature composed of 196 genes, combined with the application of a machine learning algorithm named Support Vector Machine (SVM) and categorizes substances into either sensitizers or non-sensitizers. Remarkably, GARD®skin is the first OECD TG 442 method demonstrated to function compatibly with oil, a non-polar extraction vehicle frequently employed in *in vivo* assessment of medical devices.

Here we present the utilization of *in vitro* testing results, including GARD®skin Medical Device data, for obtaining CE-marking under the European Medical Device Regulation 2017/745 (MDR) for an innovative tinnitus treatment medical device Tinearity® G1 **(Fig. 1)**.

The medical device was classified as a non-sensitizer in both polar and non-polar extracts in the GARD®skin Medical Device assay. These results, alongside cytotoxicity and *in vitro* skin irritation testing data, and chemical characterization were employed for the biological evaluation of Tinearity® G1 and submitted for regulatory filing in a weight of evidence approach.

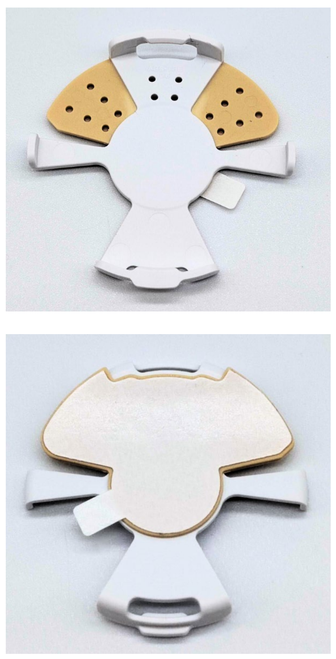


Figure 1: Tinearity® G1

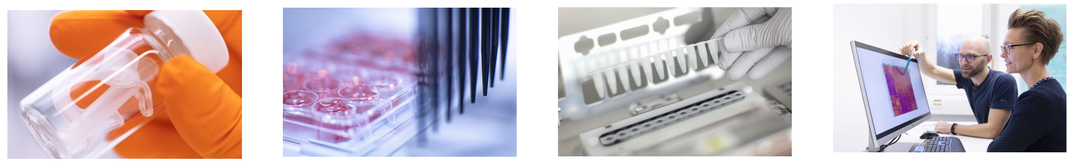
Methods

The GARD®skin Medical Device assay is based on the standard GARD®skin assay, extraction procedures are performed according to ISO 10993-12:2021.

The GARD®skin assay integrates genomics and machine learning for skin sensitization hazard assessment. Compounds are tested using a dendritic cell-like cell line, SenzaCell™, employing a single input concentration based on maximum solubility or cytotoxicity (<500µM). After RNA extraction and gene expression measurement, these gene levels serve as input values for a Support Vector Machine prediction algorithm. The algorithm assigns test samples with decision values (DVs), the sign of which is evaluated by a prediction model; a test item with a mean positive DV is classified as a sensitizer, while a test item with a mean negative DV is classified as a non-sensitizer. The assay process is outlined in **Figure 2**.

In addition, *in vitro* skin irritation (ISO 10993-23) and cytotoxicity (ISO 10993-5) tests were performed according to established standardized protocols.

In this study, extractions were conducted using the intact test item at a ratio of 3cm²/ml. For skin irritation and skin sensitization testing, extractions were carried out with saline and super refined olive oil under conditions of 72±2 hours at 37±1°C. For cytotoxicity testing, a medium with serum was utilized, with an extraction duration of 72±2 hours at 37±1°C.



Step 1. Prepare extracts from test items according to ISO 10993-12:2021.

Step 2. Expose cells (the SenzaCell™ cell line) to the extracts at determined concentration.

Step 3. Measure the gene expression levels of 196 biomarkers, the genomic biomarker signature.

Step 4. GARD® Data Analysis Application makes a binary prediction based on gene expression analysis.

Figure 2. GARD®skin Medical Device in four steps
Testing of the extracts was performed according to the GARD®skin Medical Device protocol which combines the extraction protocol as described in ISO 10993-12:2021 and the GARD®skin protocol described in OECD TG 422E. In short: (Step 1) Extracts from the medical device were prepared according to ISO 10993-12:2021. (Step 2) Cells were exposed to the extract under 24h. (Step 3) Total RNA were isolated from the cells, and the gene expression of the 196 genes in the GARD® Prediction Signature was measured. (Step 4) Gene expression data were uploaded into the cloud-based GARD® Data Analysis Application (GDAA) harbouring the data analysis pipeline.

Results

Figure 3 displays outcomes from *in vitro* safety testing focusing on the skin sensitization endpoint. The positive and negative controls were accurate, and Tinearity® G1 was predicted to be non-sensitizing across both extraction vehicles. Moreover, *in vitro* evaluations for skin irritation and cytotoxicity endpoints revealed no hazards.

Subsequently, these testing results were integrated into a weight of evidence approach for risk assessment. This approach was combined with toxicological evaluation of the material, forming a comprehensive biological evaluation process.

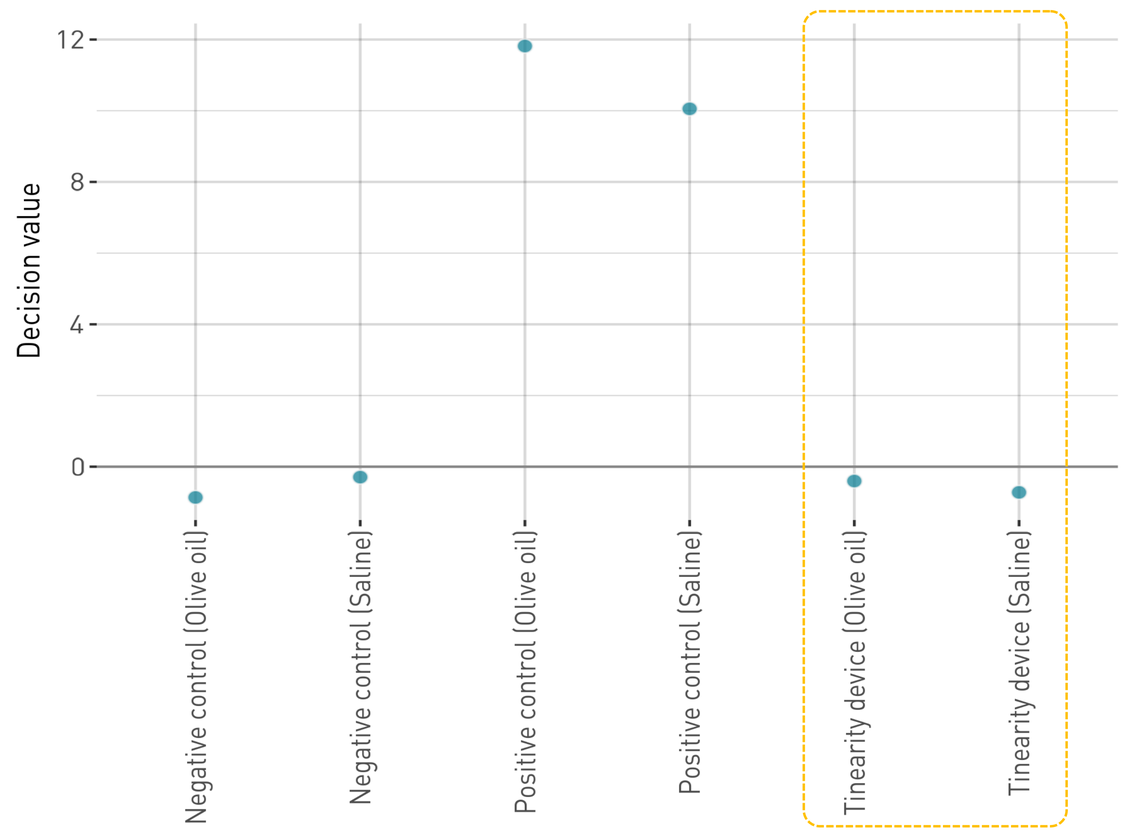


Figure 3: Mean Decision Values obtained from GARD®skin Medical Device testing for Controls and Tinearity® G1 device in the two extraction vehicles olive oil and saline.

Conclusion

It is demonstrated that the GARD®skin assay (OECD TG 442E[1]), an *in vitro* method for the assessment of skin sensitization together with cytotoxicity, *in vitro* irritation and chemical characterization support the biological safety of the device Tinearity® G1. The tinnitus treatment medical device Tinearity® G1 obtained CE-marking under European Medical Device Regulation 2017/745 (MDR) based on negative results for each endpoint and chemical characterization.

References

[1] OECD 2022, Test No. 442E: In Vitro Skin Sensitization, OECD Guidelines for the Testing of Chemicals, Section 4, OECD Publishing

Poster presentation at
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