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Poster F326

Web Application to Classify and Subcategorize Skin Sensitizers Using Human Data

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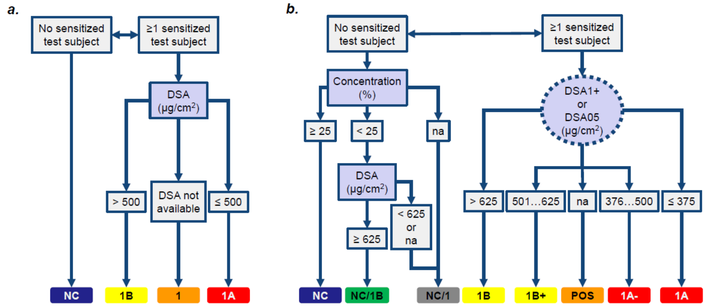
Introduction

- To support the evaluation of non-animal approaches for assessing skin sensitization hazard, the National Toxicology Program Interagency Center for the Evaluation of Alternative Toxicological Methods (NICEATM) and the German Federal Institute for Risk Assessment (BfR) curated data from two types of human predictive patch tests (HPPT), the human repeat insult patch test and human maximization test.
- These data were used to develop a modified approach for assigning skin sensitization potency subcategorizations based on the United Nations Globally Harmonized System of Classification and Labelling of Chemicals (GHS; Herzler et al. 2024).

Figure 1. (a) Standard GHS Classification Approach (b) Modified Classification Approach
Figure Adapted from Herzler et al. 2024



- The modified classification approach **(Figure 1b)** improves upon the standard GHS classification approach by incorporating potency using the DSA1+ (the dose that sensitizes one test subject) or DSA05 (the dose that sensitizes 5% of test subjects) dose metric and introducing ambiguous subcategorizations to address uncertainty. The approach also uses weight of evidence (WoE) to consolidate multiple HPPT results into a single GHS classification for each chemical.
- To allow users to apply this modified classification approach to their own HPPT data, we have developed an open-source web application, **HPPT App**. The App provides an interactive platform allowing users to input data, visualize results, and understand classification outputs.

HPPT App - User Interface (UI)

- The HPPT App is a web application that enables users to classify their own HPPT data for skin sensitization potency using a WoE approach developed by the Human Data Subgroup of the Expert Group on Defined Approaches for Skin Sensitization of the Organisation for Economic Co-operation and Development.
- The user interface was designed to provide an intuitive layout for data input and results display. There is also an About page that provides background information and a Help page with contact details for questions or feedback.

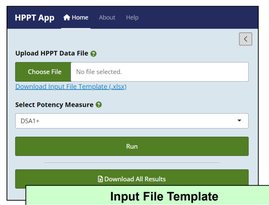
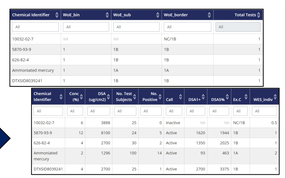


Figure 2. Application User Interface Workflow



Input File Template

chemical_identifier	concentration_percent	dsa_ug_cm2	n_testsubjects	n_positive
10032-02-7	6	3888	25	0
DTXSID8039241	4	2700	25	1
Ammoniated mercury	2	1296	100	14
5870-93-9	12	8100	24	5
626-82-4	4	2700	30	2

- The App home page allows the user to input HPPT data via file upload. It also provides a template for the data file.
- The user chooses if classification will be based on DSA1+ or DSA05.
- Required inputs include chemical identifier, test concentration, dose per skin area (DSA), number of sensitized test subjects and number of positive responses.

- Input data are processed using custom scripts that implement the modified GHS classification criteria, allowing for real-time classification and subcategorization of skin sensitizers.
- Data manipulation libraries were used to preprocess and analyze the data efficiently.
- To classify a substance for skin sensitization hazard, the HPPT App first derives DSA1+ and DSA05.
- Classifications are assigned for each record, and results are consolidated into one overall classification per substance.

- Result tables can be viewed interactively and downloaded as a CSV or an Excel worksheet.
- List of result tables:
  - Overall Weight of Evidence GHS Classification
  - Classification Based on Individual WoE Methods
  - Individual Classifications by Record
  - Reproducibility of HPPT-based WoE Classifications

Classifications Based on Individual Methods

- Substances are first assigned extrapolated classifications (Ex.C) for each individual test outcome (**Figure 3**). The Ex.Cs are then consolidated using three WoE approaches (Herzler et al. 2024; **Figure 4**) :
  - **WES** (WoE score) provides a score for each test substance based on individual test outcomes.
  - **MLLP** (median-like location parameter) provides the value at the median position of individual test outcomes.
  - **MSPE** (median sensitization potency estimate) is a modified version of the MLLP.
- The HPPT App returns three different GHS category modes:
  - **GHS$_{BIN}$**: substance classified in a binary manner as Category 1 (sensitizer) or NC (not classified).
  - **GHS$_{SUB}$**: substance assigned to one of three classes: 1A strong sensitizer, 1B weak sensitizer, or NC.
  - **GHS$_{BORDER}$**: substance assigned to one of five classes: 1A strong sensitizer, 1* (sensitizer, but subclassification not possible), 1B weak sensitizer, NC/1B ambiguous (substance may or may not be a sensitizer, but 1A can be ruled out), or NC.

Figure 3. Interactive Output Table Showing Ex.C and WES for Each Record

Chemical Identifier	Conc (%)	DSA (ug/cm2)	No. Test Subjects	No. Positive	Call	DSA1+	DSA05%	Ex.C	WES_indiv
5870-93-9	12	8100	24	5	Active	1620	1944	1B	1
626-82-4	4	2700	30	2	Active	1350	2025	1B	1
Ammoniated mercury	2	1296	100	14	Active	93	463	1A	2
DTXSID8039241	4	2700	25	1	Active	2700	3375	1B	1
10032-02-7	6	3888	25	0	Inactive	*NA*	*NA*	NC/1B	0.5

Each field can be sorted. Here the Call field is sorted to show the Active values.

Figure 4. Sections of Interactive Output Table with GHS Classifications based on WES, MLLP, and MSPE and Summary of Ex.C's for Each Chemical

Chemical Identifier	WES	WES_GHS_bin	WES_GHS_sub	WES_GHS_border	MLLP_bin	MLLP_sub	MLLP_GHS_bin	MLLP_GHS_sub	MLLP_GHS_border	MSPE	MSPE_GHS_bin	MSPE_GHS_sub	MSPE_GHS_border
10032-02-7	0.5	*NA*	*NA*	NC/1B	*NA*	*NA*	*NA*	*NA*	*NA*	*NA*	*NA*	*NA*	*NA*
5870-93-9	1	1	1B	1B	1620	1620	1	1B	1B	1620	1	1B	1B
626-82-4	1	1	1B	1B	1350	1350	1	1B	1B	1350	1	1B	1B
Ammoniated mercury	1	1	1A	1A	93	93	1	1A	1A	93	1	1A	1A
DTXSID8039241	1	1	1B	1B	2700	2700	1	1B	1B	2700	1	1B	1B

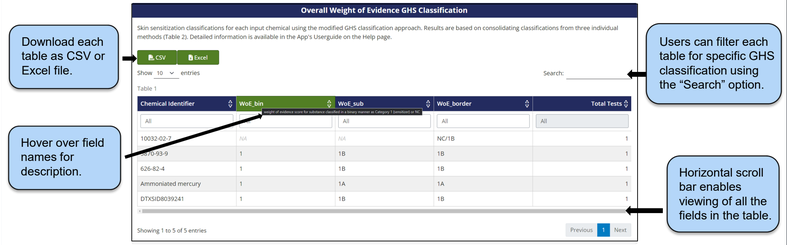
User-provided chemical identifier can be chemical name, CASRN, or DTXSID.

NC	NC/1B	NC/1	1B	1B+	POS	1A-	1A
0	1	0	0	0	0	0	0
0	0	0	1	0	0	0	0
0	0	0	1	0	0	0	0
0	0	0	0	0	0	0	1
0	0	0	1	0	0	0	0

Each field can be filtered.

Overall WoE GHS Classifications

Figure 5. Interactive Output Table with Overall WoE GHS Classifications



- Each chemical is assigned overall WoE GHS classifications based on the concordant outcome across the MLLP, MSPE, and WES (**Figure 5**).

Reproducibility of HPPT-based WoE Classifications

- The HPPT App also estimates reproducibility for chemicals with at least two unambiguous test results by comparing Ex.C to WoE$_{BIN}$ and WoE$_{SUB}$ classifications, indicating agreement or disagreement with overall classification outcome.

Figure 6. Interactive Output Tables Showing Mean Reproducibility of the WoE$_{BIN}$ (a) and WoE$_{SUB}$ (b) Classifications

a.

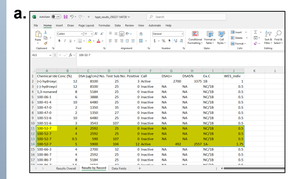
Chemical Identifier	WoE_bin	WoE_bin Reproducibility	Total Tests (Reproducibility)
Mercuric chloride	1	100	6
Malathion	1	100	4
Mafenide	1	100	3
Linalyl acetate	1	100	2
Hydroxycitronellal	1	100	12
Hydralazine	1	100	3
Hexahydrocoumarin	1	100	2
Hexachlorophene	1	80	5
Geraniol	1	100	3
Formaldehyde	1	100	5

b.

Chemical Identifier	WoE_sub	WoE_sub Reproducibility	Total Tests (Reproducibility)
Diethyl maleate	1A	75	4
Ethyl acrylate	1A	50	4
Farnesol	1B	100	5
Formaldehyde	1A	100	5
Geraniol	1B	66.67	3
Hexachlorophene	1B	60	5
Hexahydrocoumarin	1B	50	2
Hydralazine	1A	100	3
Hydroxycitronellal	1B	100	12
Linalyl acetate	1B	100	2

Download Output Tables

Figure 7. (a) Download File with All Query Results (b) Individual Table Output with Custom Filtering (Red) and Sorting (Blue) (c) Download File with Refined Individual Table Output



b.

Chemical Identifier	Conc (%)	DSA (ug/cm2)	No. Test Subjects	No. Positive	Call	DSA1+	DSA05%	Ex.C	WES_indiv
100-52-7	All	All	All	All	All	All	All	All	All
100-52-7	5	5900	104	12	Active	492	2557	1A	1.75
100-52-7	4	2592	25	1	Inactive	*NA*	*NA*	NC/1B	0.5
100-52-7	4	2592	25	0	Inactive	*NA*	*NA*	NC/1B	0.5
100-52-7	0.5	590	107	0	Inactive	*NA*	*NA*	NC/1B	0.5

c.

	A	B	C	D	E	F	G	H	I
1	HPPT App								
2	Table 3								
3	Chemical Identifier	Conc (%)	DSA (ug/cm2)	No. Test Subjects	No. Positive	Call	DSA1+	DSA05%	Ex.C	WES_indiv
4	100-52-7	5	5900	104	12	Active	492	2557	1A	1.75
5	100-52-7	4	2592	25	1	Inactive	*NA*	*NA*	NC/1B	0.5
6	100-52-7	4	2592	25	0	Inactive	*NA*	*NA*	NC/1B	0.5
7	100-52-7	0.5	590	107	0	Inactive	*NA*	*NA*	NC/1B	0.5

Discussion and Conclusion

- Approaches currently used to assign skin sensitizers to GHS potency subcategories do not consider the number of sensitized test subjects, potentially leading to an underestimation of potency.
- The modified approach based on HPPT data enhances the evaluation of non-animal skin sensitization tests by incorporating potency into the GHS classification decision tree and applying WoE methods. However, the approach involves complex decision tree logic that may be challenging for non-computational users.
- This user-friendly, open-source R Shiny-based HPPT App was developed to facilitate public use of the modified classification approach and allow users to easily classify their HPPT data in a way that reflects human potency.

References

- Herzler M et al. Use of human predictive patch test (HPPT) data for the classification of skin sensitization hazard and potency. Archives of Toxicology (2024) 98:1253–1269. https://doi.org/10.1007/s00204-023-03656-4
- Strickland J et al. A database of human predictive patch test data for skin sensitization. Archives of Toxicology (2023) 97:2825–2837. https://doi.org/10.1007/s00204-023-03530-3

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