IDEA
International Dialogue for Evaluation of Allergens

Development of the RCPL and its key elements

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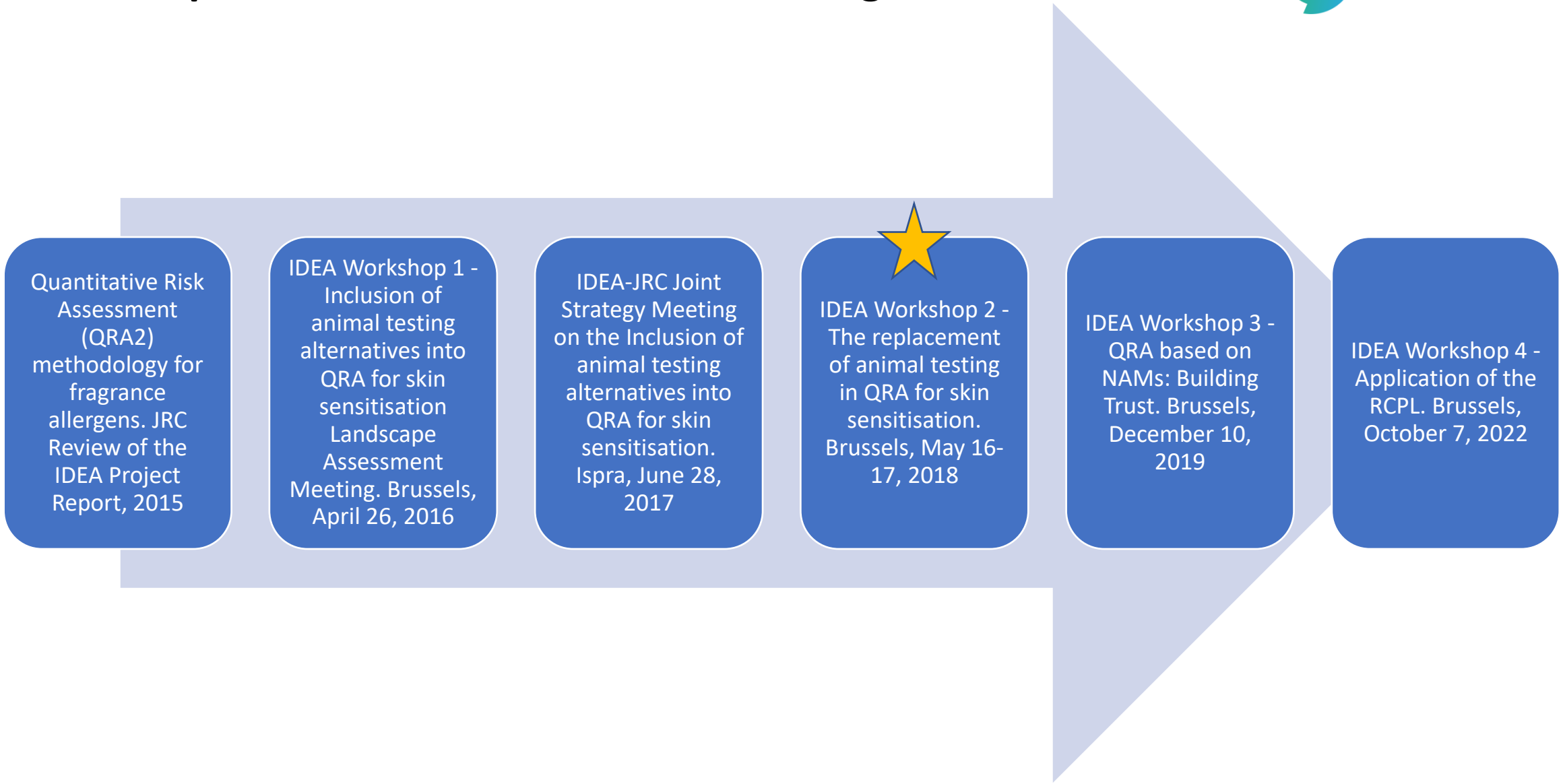
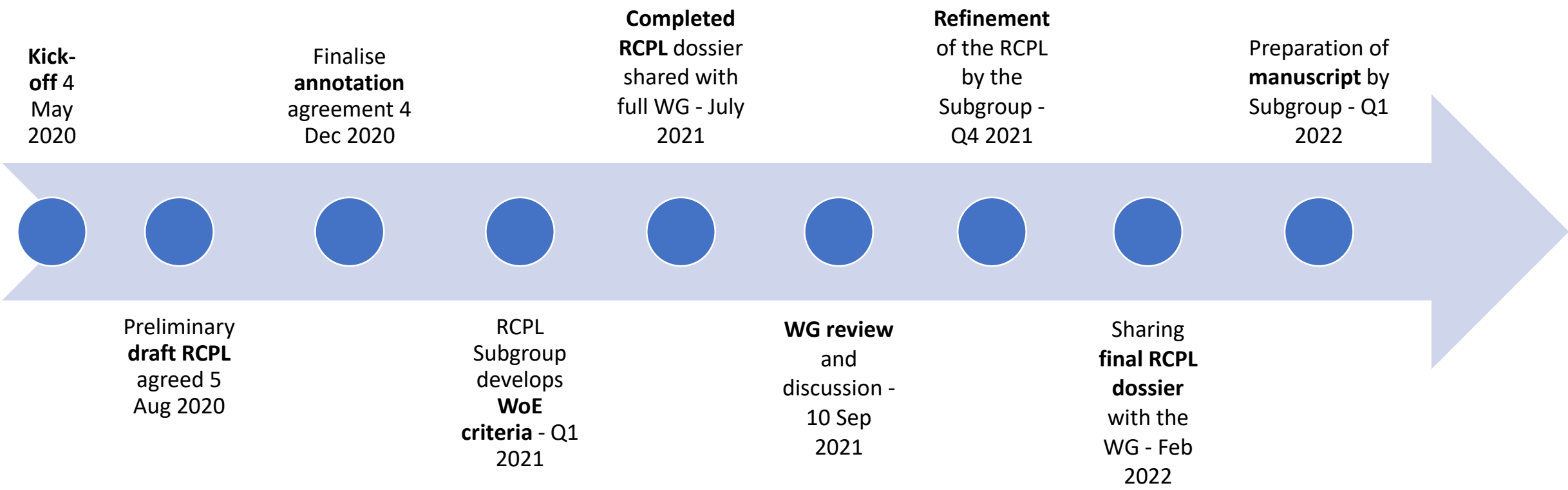
Development of IDEA RCPL – How did we get here?



Key steps in the process

1. RCPL development was **a key conclusion of 2018 IDEA WS on AAT** and its criteria was further explored in 2019 IDEA WS
2. **IDEA RCPL WG established**
   - Industry members and partners
   - Observers: SCCS and DG Grow
   - IDEA Management Team
   - IDEA Supervisory Group (Ian Kimber)
3. **Objective**: The RCPL was aimed to comprise chemicals [both fragrance and non-fragrance materials] that vary in potency ranging in a continuous scale from extreme sensitizers to sensitizers having only very weak or no sensitising potential (based on weight of evidence of human and animal data), and to be used to help evaluate NAMs for potency prediction

IDEA RCPL WG progress outline



Key steps in the process

4. **Selection of chemicals**
   - Fragrance chemicals selected from OECD LLNA Reference database (db) and IFRA Standards for skin sensitisation
   - Selection of the candidate RCPL (39 chemicals) covering the whole potency scale
   - Additional non-fragrance extreme sensitisers added
5. **Annotation and format of the LLNA and human data to consider as source data**
   - LLNA and human data in OECD db was compared to RIFM db data
   - OECD db was the most useful source of information for mouse data whereas RIFM db was particularly valuable for human data

Key steps in the process

6. **Consolidation of metrics for LLNA and human source data (EC3 and DSA04)**
7. **Definition of Potency Value (in µg/cm2)**
8. **Workflow developed for WoE integration of the LLNA and human data**
    - Building the criteria for the integration of the LLNA and human data became far more challenging and a key value of the final RCPL
    - Process was iterative, pragmatic and built from the quality of source data
    - For data integration ideas considered e.g., a) averaging data and working with averages and ranges, b) graphical analysis of all source data.
9. **Review by the WG and further refinement**
10. **Issue of final RCPL**

Final output: IDEA RCPL Potency Values

Name	Pre / Pro - Hapten	Potency Value [µg/cm²]
5-Chloro-2-methyl-4-isothiazolin-one (CMIT)		2.3
2,4-Dinitrochlorobenzene (DCNB)		3.4
1,4-Phenylenediamine (PPD)	Pre	3.9
Glutaraldehyde		20.0
trans-2-Hexenal		39.3
1,4-Dihydroquinone	Pre	47.5
Benzyl bromide		50.0
1,1,3-Trimethyl-2-formylcyclohexa-2,4-diene (Safranal)		106
Methyl 2-nonynoate (Methyl octine carbonate)		109
Methyl 2-octynoate (Methyl heptine carbonate)		125
Isoeugenol	Pre	325
Phenylacetaldehyde		750
Allyl phenoxyacetate		775
Cinnamic aldehyde		885
3-Propylidenephthalide	Pre	925
4-Hydroxy-2,5-dimethyl-3(2H)-furanone (Furaneol)		1181

Name	Pre / Pro - Hapten	Potency Value [µg/cm²]
Citral		1450
p-Mentha-1,8-dien-7-al (Perillaldehyde)		2175
Benzaldehyde		4094
Lyral (HICC)		4275
Hydroxycitronellal		5275
Cinnamic alcohol	Pre / Pro	5775
Eugenol	Pre / Pro	7357
Geraniol	Pre / Pro	9197
Coumarin		11792
Carvone		17573
Benzyl salicylate		17715
Hexyl cinnamic aldehyde		23620
Benzyl Alcohol	Pro	>25000
Benzyl benzoate		>25000
Isomethylionone (α-)		>25000
Methyl salicylate		No PV derived- very weak/non-sensitiser
Vanillin		No PV derived - very weak/non-sensitiser

Pre- and prohaptens based on Patlewitz et al., 2016 and Casati et al., 2016

Key elements of the RCPL

1. Definitions: Potency Value (PV)
2. Guidance for the derivation of LLNA EC3 values
3. Guidance for the derivation of human NOEL and LOEL (DSA04) values
4. Derivation of Potency Values WoE Workflow
5. Review of the PVs derived

Definitions: Potency Value (PV)

1. **PV** reflects an overall assessment of the relative skin sensitising potency of a chemical based upon consideration of relevant human data, LLNA EC3 values, and expert judgement. It is an inflection point where skin sensitisation starts and therefore described as the lowest concentration of a test chemical (in µg/cm^2) that will result in the initiation of a skin sensitisation response. The PV is designed to characterize a substance property.

2. On the other hand, a **NESIL** (No Expected Sensitisation Induction Level) identifies a concentration of a chemical that will not result in the induction of skin sensitisation. This has value in the risk assessment process but does not necessarily reflect accurately the skin sensitising potency of a chemical.

3. A **NOEL** (No Observable Effect Level) and **LOEL** (Lowest Observable Effect Level) also have value in risk assessments, but neither necessarily reflects accurately the sensitising potency of a chemical.

Guidance for the derivation of LLNA EC3 values

1. The LLNA studies included in the OECD database had a predefined criteria applied to them which have been adopted for the RCPL
2. Where the EC3 had been extrapolated, further evaluation criteria were applied (partially based on Ryan et al. 2007).
    - EC3s not fulfilling it were excluded (however, the chemical remained in the list if appropriate human data was available e.g., Methyl 2-nonynoate)
3. Medium-like-location parameter (MLLP) as the averaging metric for EC3s (Hoffman et al 2018; OECD TG 497) was adopted
4. Where the difference between the OECD MLLP EC3 value (in $\mu g/cm^2$) and the RIFM weighted mean EC3 value was greater than 2-fold, the RIFM LLNA data were reviewed. If the difference was less than 2-fold, the OECD MLLP EC3 value was selected for the RCPL

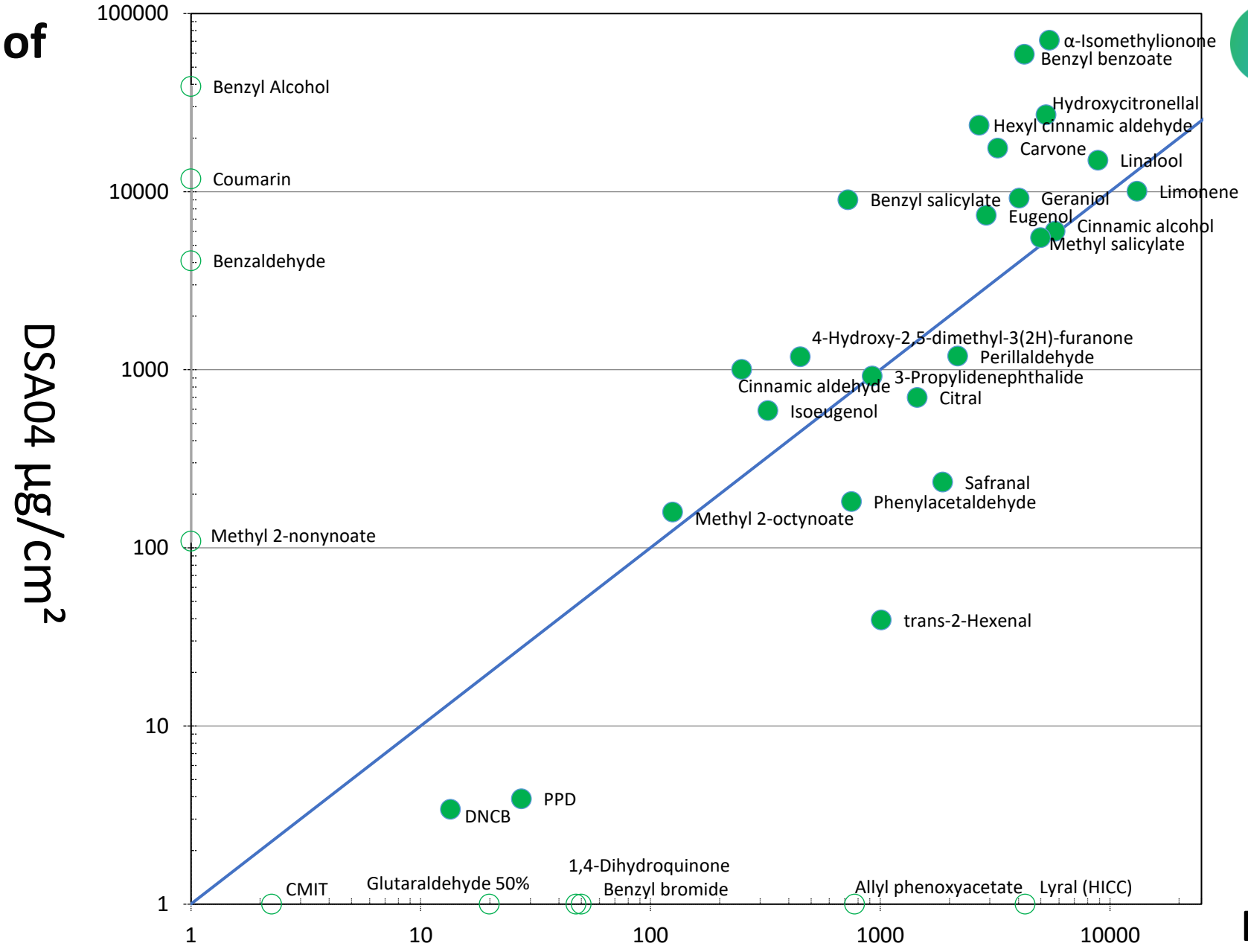
Guidance for the derivation of human NOEL and LOEL (DSA04) values

1. Results of human studies with well-defined, experimental exposure conditions during the induction phase, followed by a well-defined and well-documented elicitation challenge under medical surveillance were considered
2. Exposure level had to be expressed as the concentration of chemical per unit area of skin (measured as µg of chemical per cm^2 of skin) or, at least, to have reported details required for derivation of this value
3. HRIPTs with 100 subjects or more had the highest value as regards to fulfilling the criteria, HMTs to a lower extent
4. LOELs have been favoured as an expression of human data because, unlike NOELs, they reflect a signal of sensitisation, rather than the absence of sensitisation.

Guidance for the derivation of human NOEL and LOEL (DSA04) values (cont.)

5. When none of the available HRIPTs and HMTs reported cases of sensitisation, the NOEL was established at the highest exposure level reported in a fully valid HRIPT or, if no fully valid study was available, at the highest exposure level that could be supported from an evaluation of the body of available data.
6. A DSA (Dose per Surface Area) value was deemed to provide a sound basis for harmonising LOEL values for chemicals causing different incidences of sensitization in the panel of subjects
7. The DSA04 is the dose per unit area of skin estimated to result in an incidence of sensitisation of 4% of the exposed study population
8. A DSA04 value was selected in preference to any other DSA value (such as DSA02 or DSA05) because it was found to correlate well with LLNA EC3 values.

Selection of DSA04

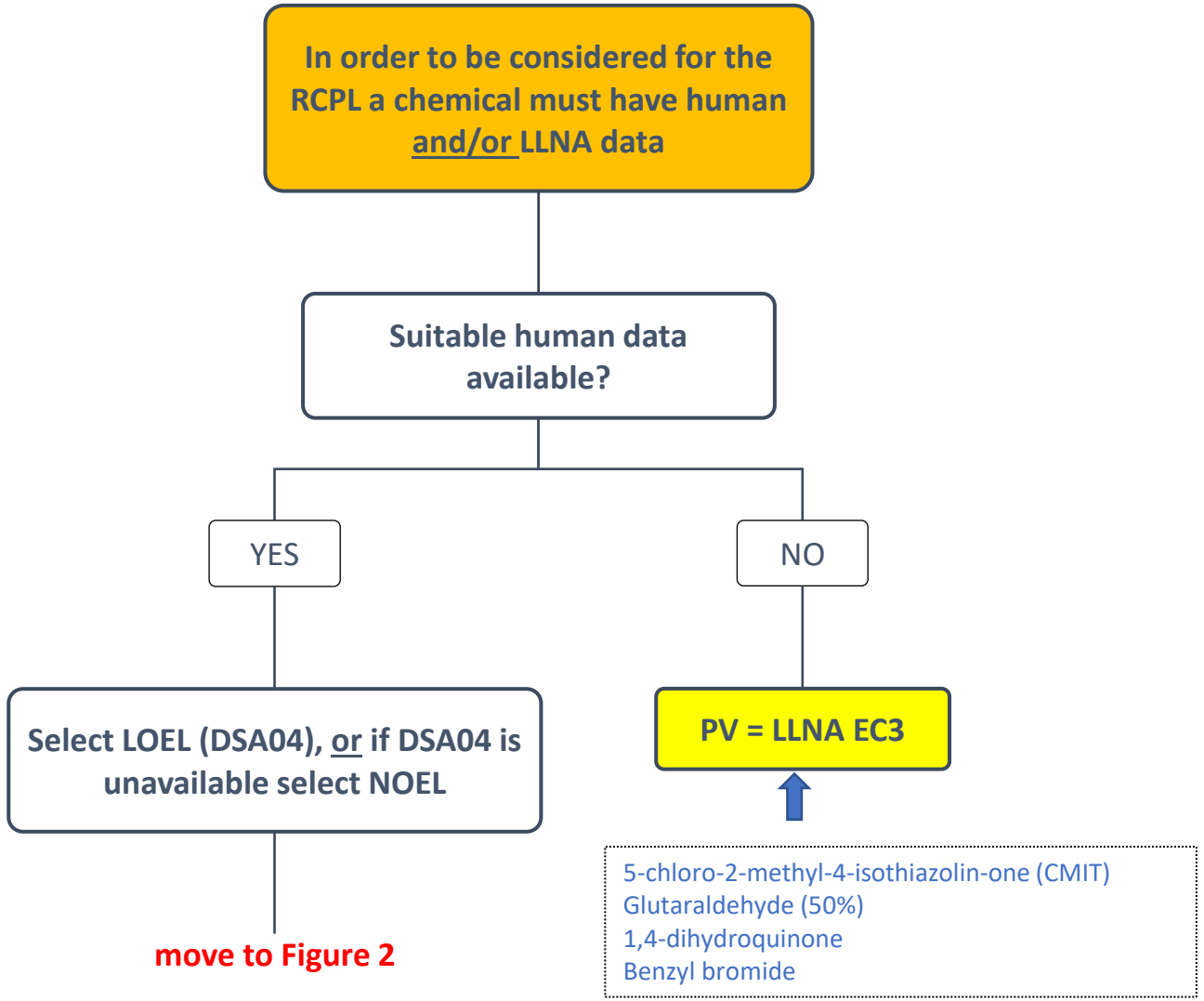


Applying the selected LLNA and human data metrics into the workflow

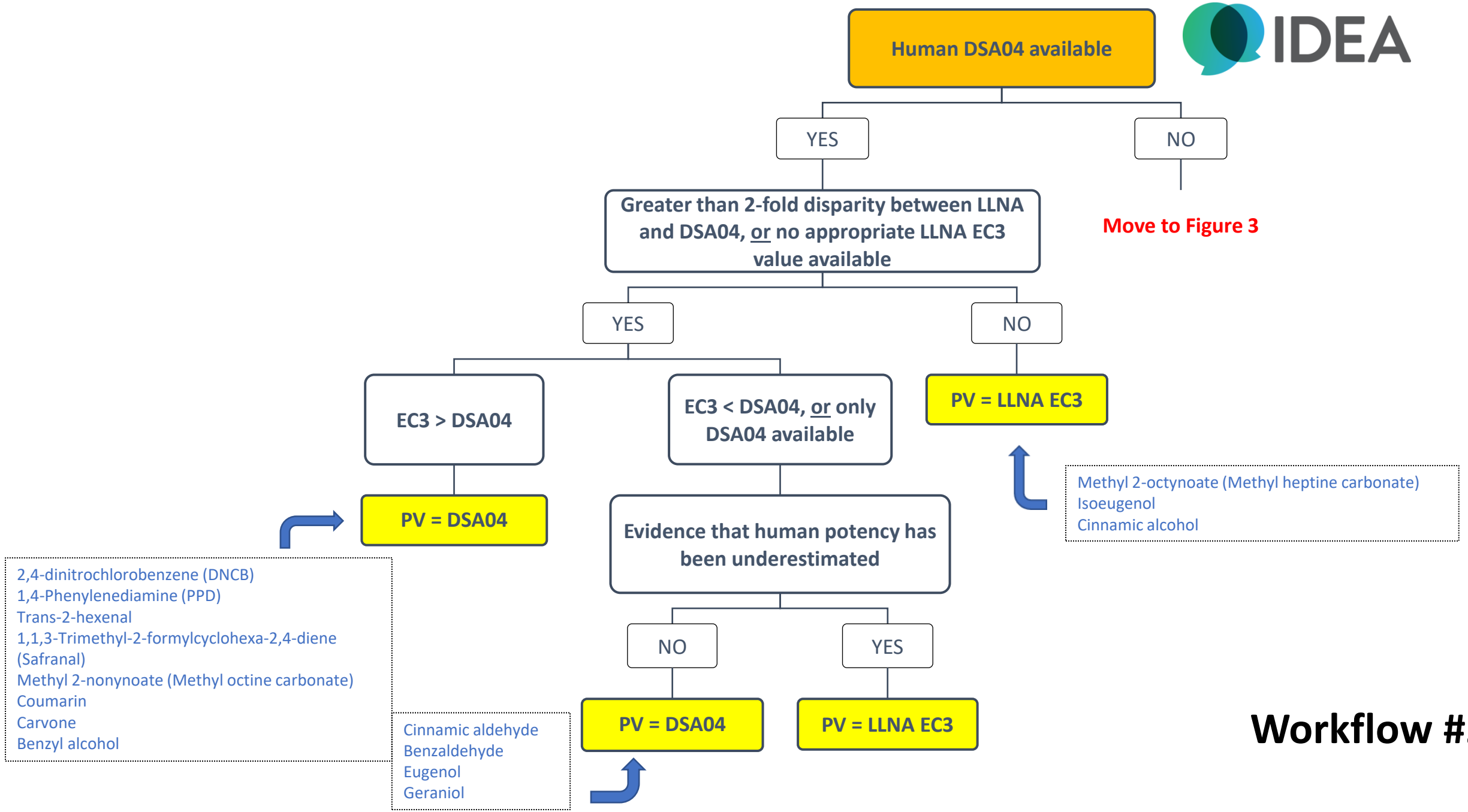
Weight of evidence applied for derivation of Potency Values.

Chemical	CAS No	Human LOEL/DSA04 [μg/cm^2]	Human NOEL [μg/cm^2]	LLNA EC3 [μg/cm^2]	Workflow number and Rationale	Potency Value [μg/cm^2]
5-Chloro-2-methyl-4-isothiazolin-one (CMIT)	26172-55-4	None	None	2.3	1	2.3
2,4-Dinitrochlorobenzene (DNCB)	97-00-7	LOEL (HRIPT) of 7 giving DSA04 of 3.4	NA	13.5	2 >2-fold difference between LLNA and DSA04 EC3 > DSA04	3.4
1,4-Phenylenediamine (PPD)	106-50-3	LOEL (HRIPT) of 7 giving DSA04 of 3.9	NA	27.5	2 >2-fold difference between LLNA and DSA04 EC3 > DSA04	3.9
Glutaraldehyde	111-30-8	LOEL inadequate[a]	None	20.0	1	20.0
trans-2-Hexenal	6728-26-3	LOEL (HRIPT) of 236 with higher uncertainty in extrapolation to DSA04 since HRIPT incidence was > 3 × 4% incidence. Extrapolated DSA04 of 39.3 was used in absence of conflicting human data	NA	1013	2 >2-fold disparity between LLNA EC3 and DSA04 EC3 > DSA04	39.9
1,4-Dihydroquinone	123-31-9	None	None	47.5	1	47.5
Benzyl bromide	100-39-0	None	None	50.0	1	50.0
1,1,3-Trimethyl-2-formylcyclohexa-2,4-diene (Safranal)	116-26-7	LOEL (HRIPT) of 250 giving DSA04 of 106 was used although the subject number was just 53 because another HRIPT using 99 subjects reported an incidence <1/3x of 4% at 59.1 resulting in a higher uncertainty of the extrapolated DSA04 (234)	NA	1875	2 >2-fold disparity between LLNA and DSA04 EC3 > DSA04	106

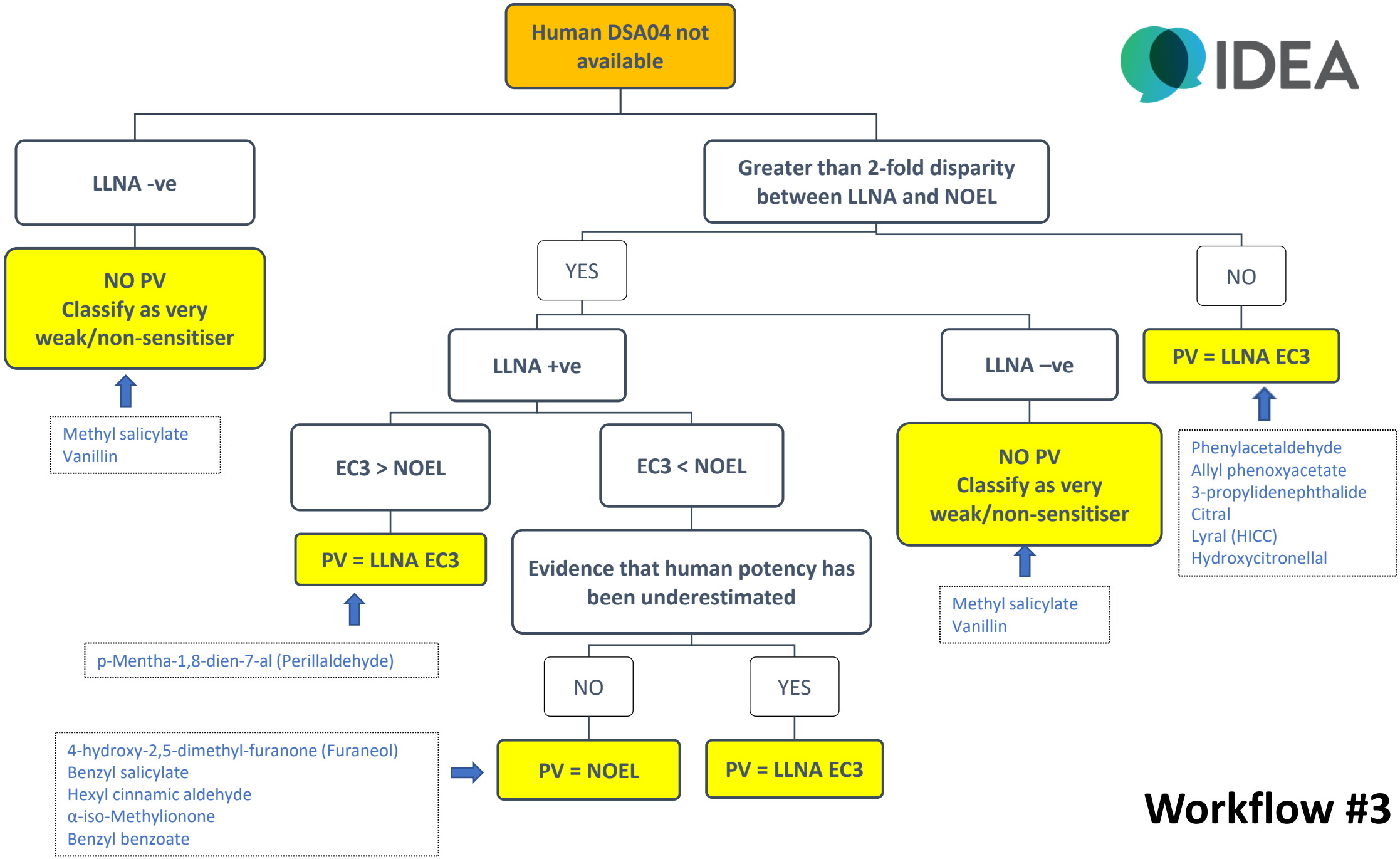
WoE for Derivation of PVs



Workflow #1

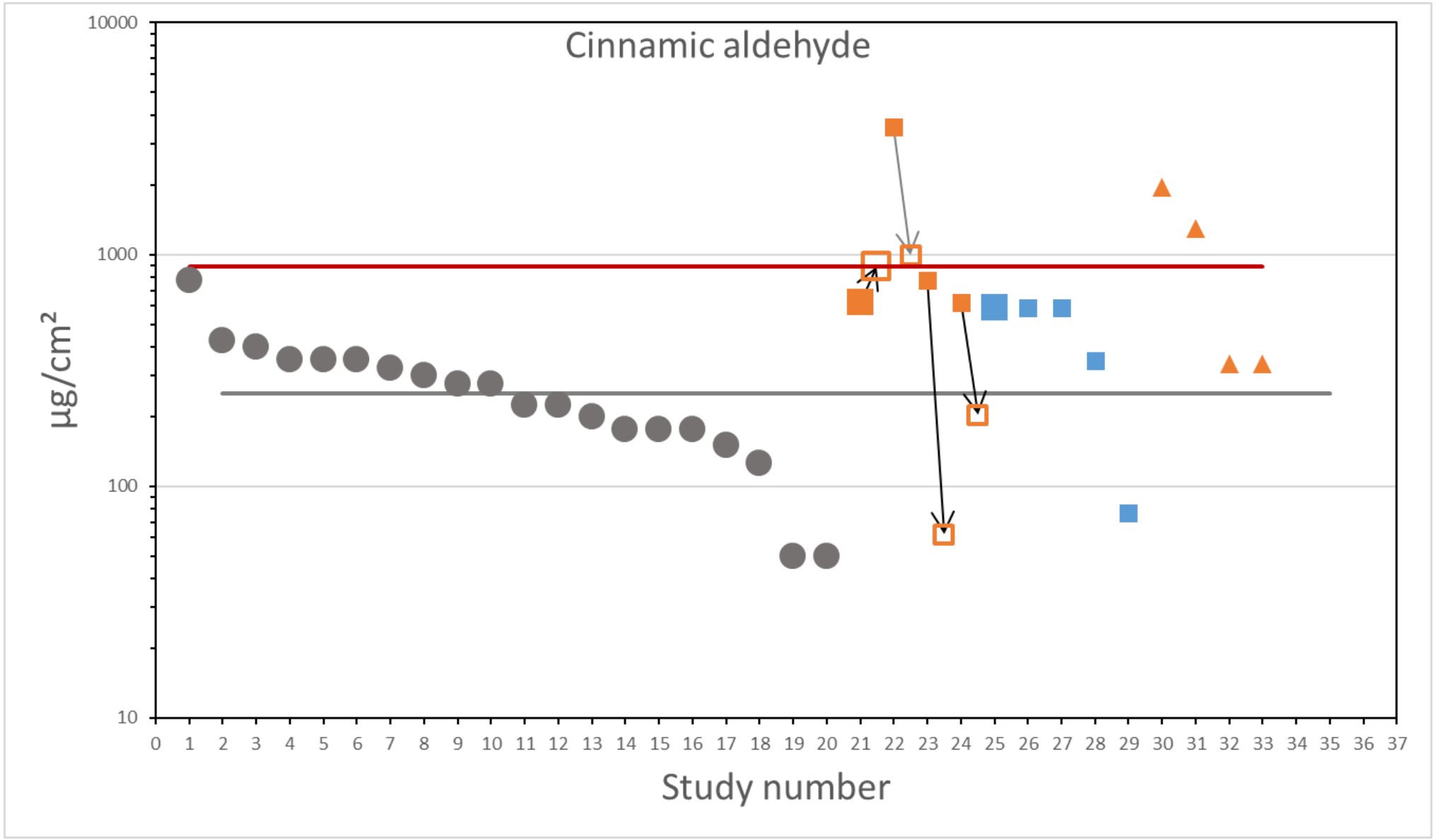

Human DSA04 available
YES
NO
Move to Figure 3
Greater than 2-fold disparity between LLNA and DSA04, or no appropriate LLNA EC3 value available
YES
NO
EC3 > DSA04
EC3 < DSA04, or only DSA04 available
PV = LLNA EC3
Methyl 2-octynoate (Methyl heptine carbonate)
Isoeugenol
Cinnamic alcohol
PV = DSA04
2,4-dinitrochlorobenzene (DNCB)
1,4-Phenylenediamine (PPD)
Trans-2-hexenal
1,1,3-Trimethyl-2-formylcyclohexa-2,4-diene (Safranal)
Methyl 2-nonynoate (Methyl octine carbonate)
Coumarin
Carvone
Benzyl alcohol
Evidence that human potency has been underestimated
NO
YES
PV = DSA04
PV = LLNA EC3
Cinnamic aldehyde
Benzaldehyde
Eugenol
Geraniol

Workflow #2

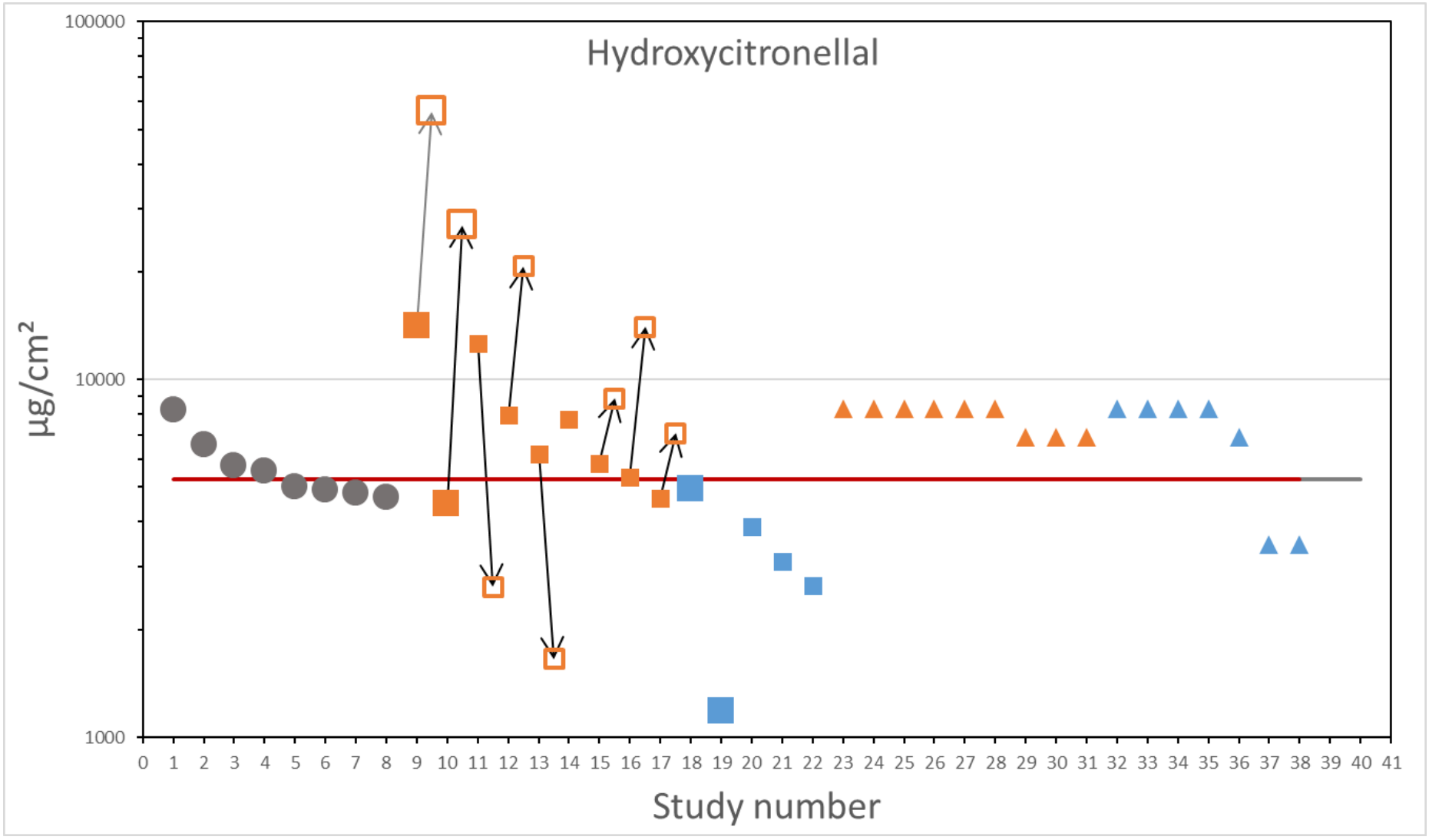

Human DSA04 not available
LLNA -ve
Greater than 2-fold disparity between LLNA and NOEL
NO PV
Classify as very weak/non-sensitiser
YES
NO
LLNA +ve
LLNA –ve
PV = LLNA EC3
Methyl salicylate
Vanillin
EC3 > NOEL
EC3 < NOEL
NO PV
Classify as very weak/non-sensitiser
Phenylacetaldehyde
Allyl phenoxyacetate
3-propylidenephthalide
Citral
Lyral (HICC)
Hydroxycitronellal
PV = LLNA EC3
Evidence that human potency has been underestimated
Methyl salicylate
Vanillin
p-Mentha-1,8-dien-7-al (Perillaldehyde)
NO
YES
4-hydroxy-2,5-dimethyl-furanone (Furaneol)
Benzyl salicylate
Hexyl cinnamic aldehyde
α-iso-Methylionone
Benzyl benzoate
PV = NOEL
PV = LLNA EC3

Workflow #3

Data variability



Data variability



PVs vs NESIL

NESIL

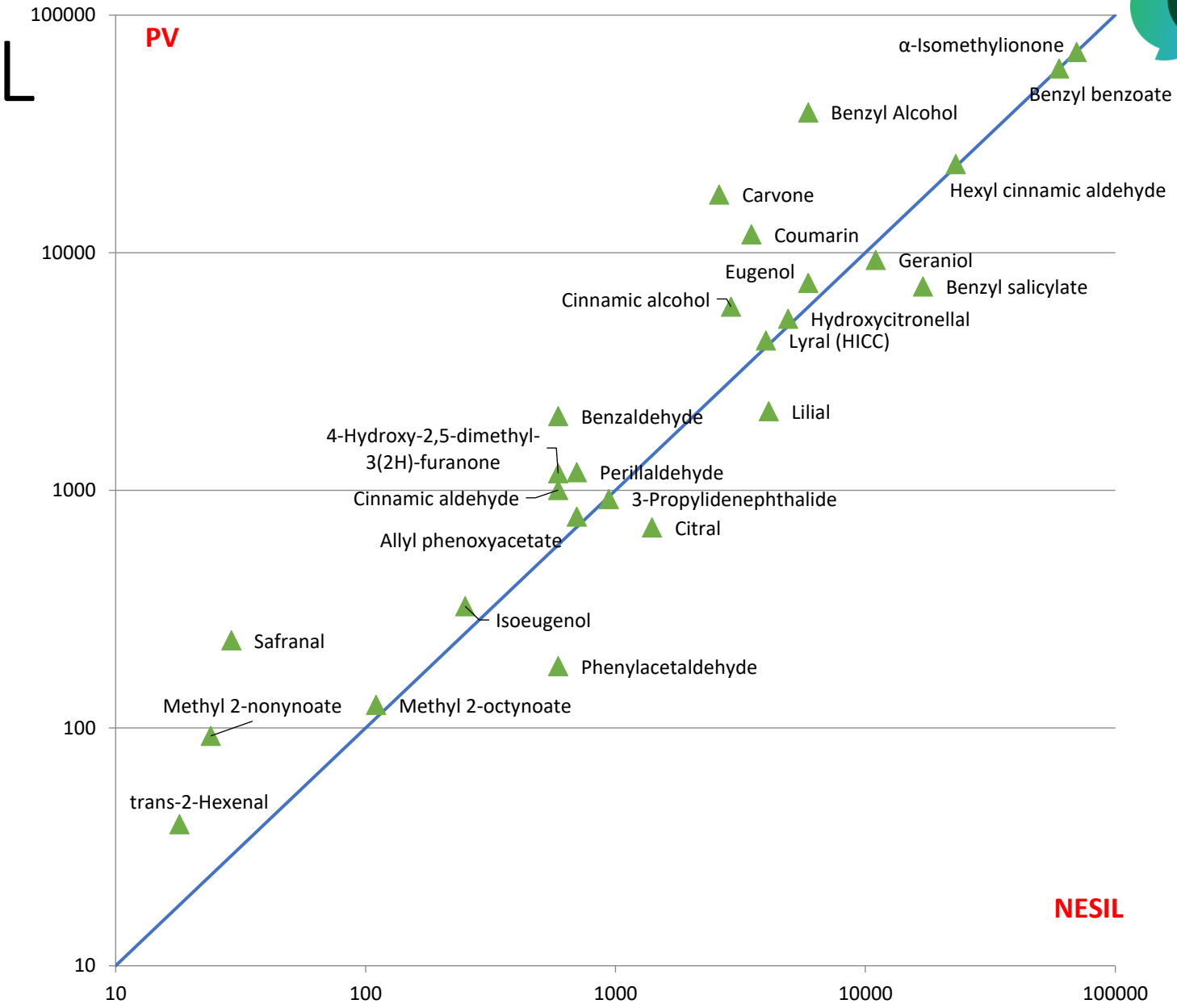
- estimated level of exposure (in dose per unit area of skin) at which sensitisation is expected not to develop under the conditions of an HRIPT.
- Based upon a NOEL value

PV

- the PV is a concentration (in dose per unit area of skin) derived from interrogation of available experimental human and animal data, at which it is estimated that skin sensitisation will first be induced

There is no set relationship between these metrics, although as expected in most cases the previously reported NESIL was lower than the PV

PV vs NESIL


PV
NESIL
100000
10000
1000
100
10
10
100
1000
10000
100000
α-Isomethylionone
Benzyl benzoate
Benzyl Alcohol
Hexyl cinnamic aldehyde
Carvone
Coumarin
Geraniol
Eugenol
Benzyl salicylate
Cinnamic alcohol
Hydroxycitronellal
Lyral (HICC)
Lilial
Benzaldehyde
4-Hydroxy-2,5-dimethyl-3(2H)-furanone
Perillaldehyde
Cinnamic aldehyde
3-Propylidenephthalide
Citral
Allyl phenoxyacetate
Isoeugenol
Safranal
Phenylacetaldehyde
Methyl 2-nonynoate
Methyl 2-octynoate
trans-2-Hexenal

What's next?

1. RCPL dissemination:
    a) Peer review publication
    b) Presentation in colloquia and conferences and sharing with other working groups
2. Explore and communicate its practical application, as it:
    a) Is the best-founded list developed with an objective and integrated approach from best available human and animal potency data
    b) Provides a flexible template for evaluating the accuracy of NAMs for measuring sensitising potency
3. The aim of this workshop is to kick start this.