Applying a Next Generation Risk Assessment (NGRA) framework for skin sensitization to inconsistent New Approach Methodology (NAM) information

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ABIHPEC webinar

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on behalf of the ICCS Skin Sensitisation Working group

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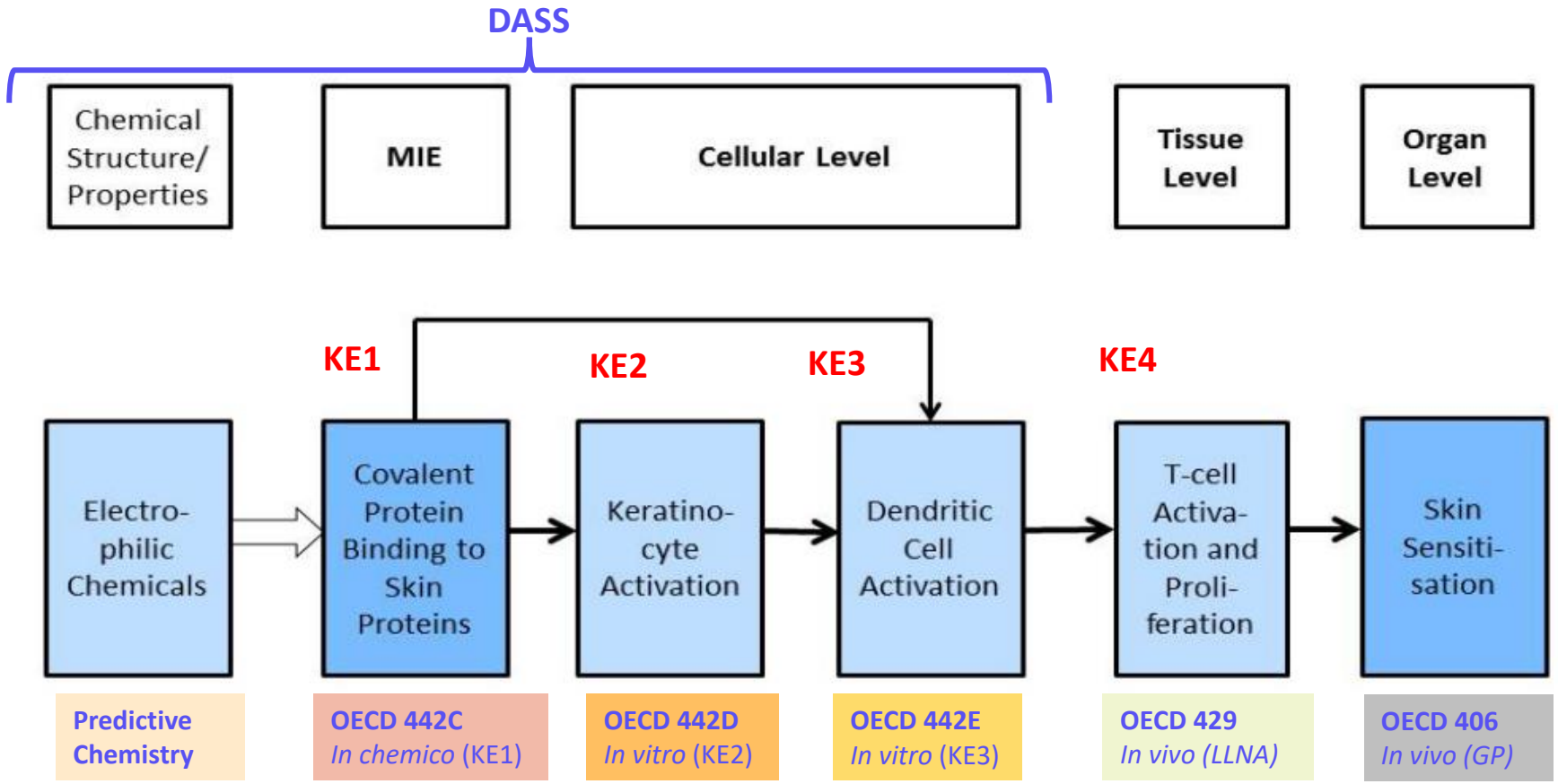
INTERNATIONAL COLLABORATION ON COSMETICS SAFETY

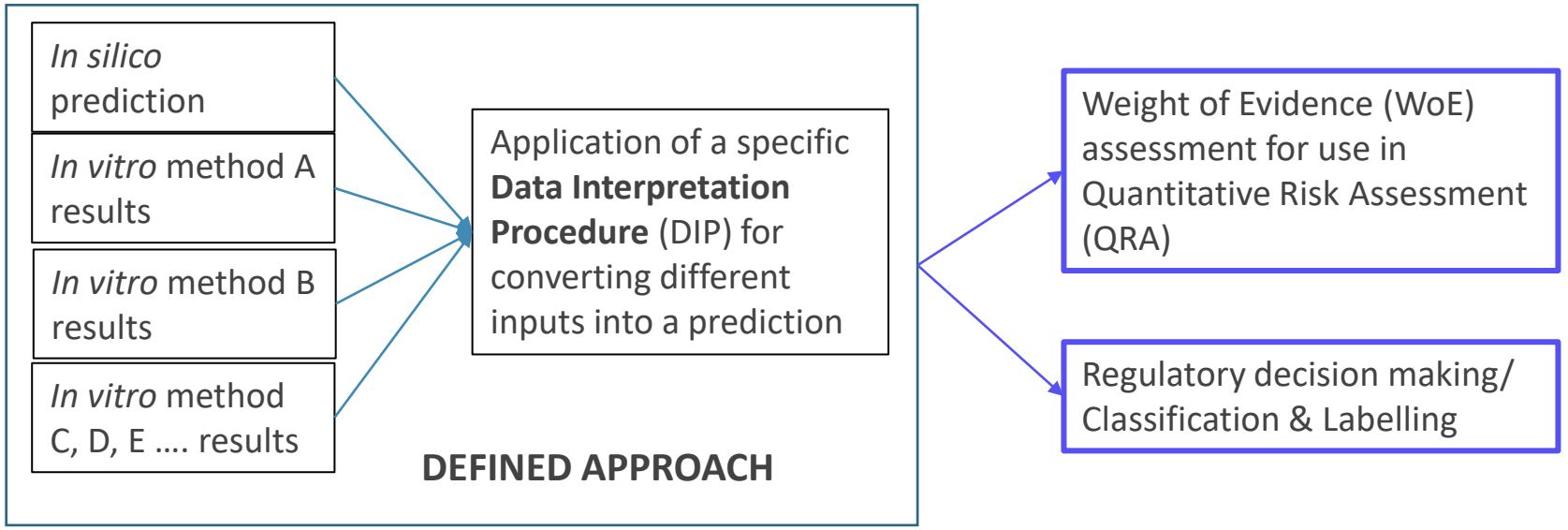
NAM developments along Skin Sensitisation AOP

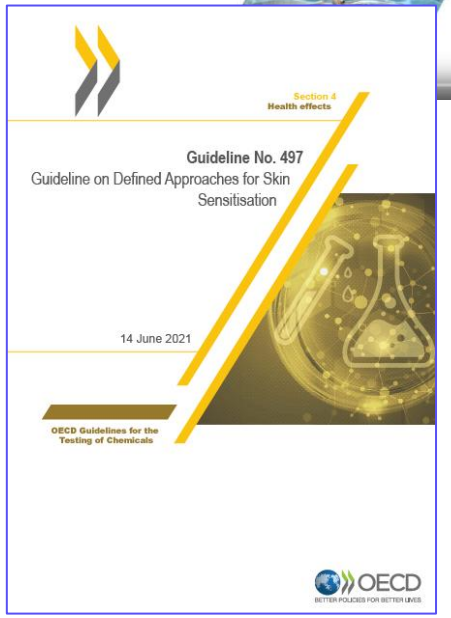


OECD, 2012. The Adverse Outcome Pathway for Skin Sensitisation Initiated by Covalent Binding to Proteins. Series on Testing and Assessment No. 168.

From IATA to Defined Approaches (DA)

- Remove expert judgement
- Are **not flexible** and **are suitable** for harmonisation





OECD 497
Defined Approach (DASS)

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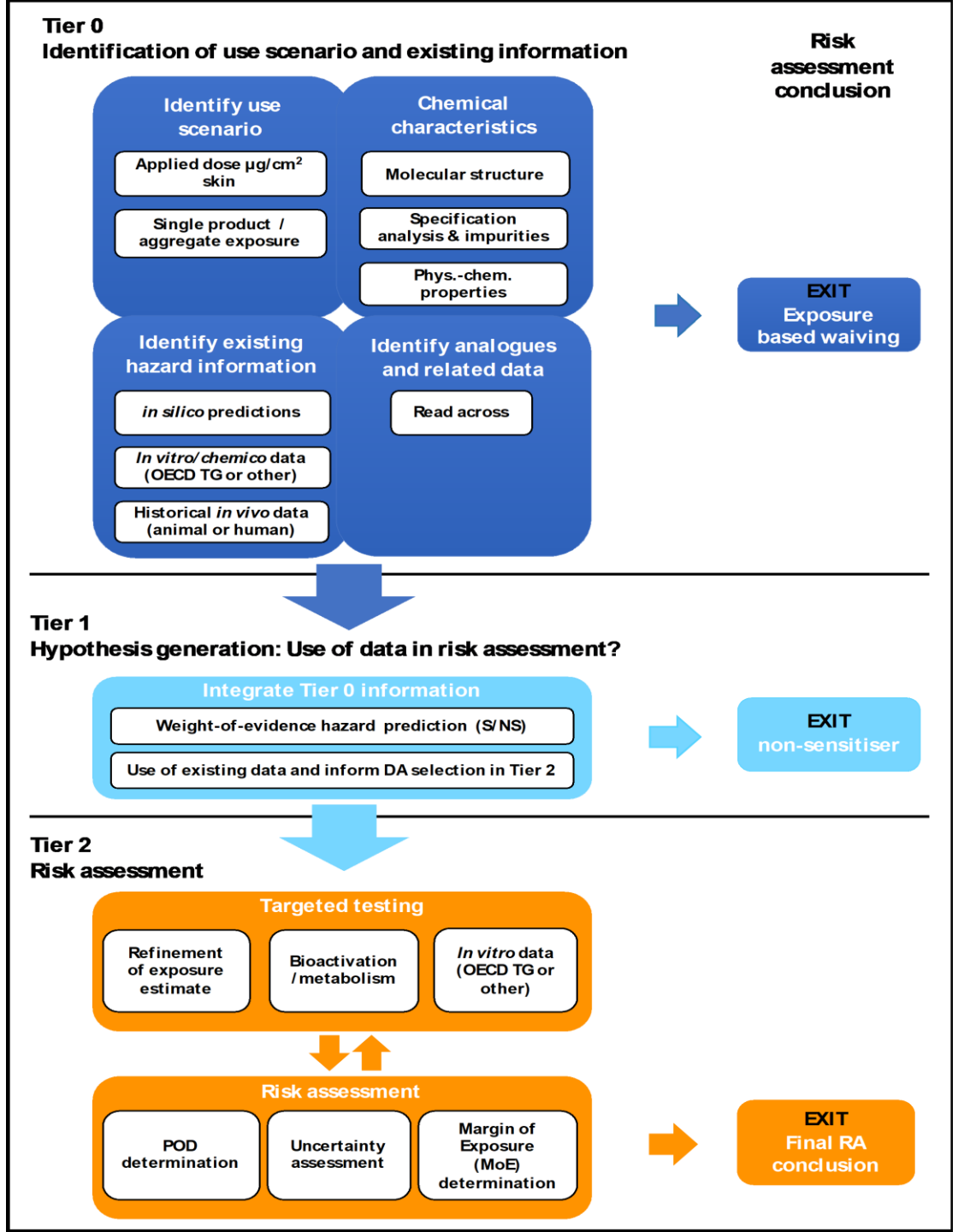
http://www.oecd.org/chemicalsafety/risk-assessment/iata-integrated-approaches-to-testing-and-assessment.htm

Next Generation Risk Assessment (NGRA) framework



https://health.ec.europa.eu/latest-updates/sccs-notes-guidance-testing-cosmetic-ingredients-and-their-safety-evaluation-12th-revision-2023-05-16_en

Gilmour et al, ALTEX, 2023 doi: 10.14573/altex.221116

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Skin sensitisation NGRA framework case studies

NGRA case studies conducted and published over the last years

- eg. coumarin, geraniol, lactic acid, propyl paraben, resorcinol etc.
- Different consumer use scenarios explored
- Case study workshops (SCCS, EPAA etc.)

Case study: MDBGN

- Consistent data with clear risk decision making

- **Framework NGRA**
- **NoG SCCS**

Case study: Geraniol

- Consistent NAM info
- Slight differences in DA outcomes

- **OECD IATA case study**

What did we learn?

- NAM/DA data to be included in a weight of evidence
- Tiered approach
- Not one approach fits all
- Different from QRA approach
- New areas of uncertainty determined

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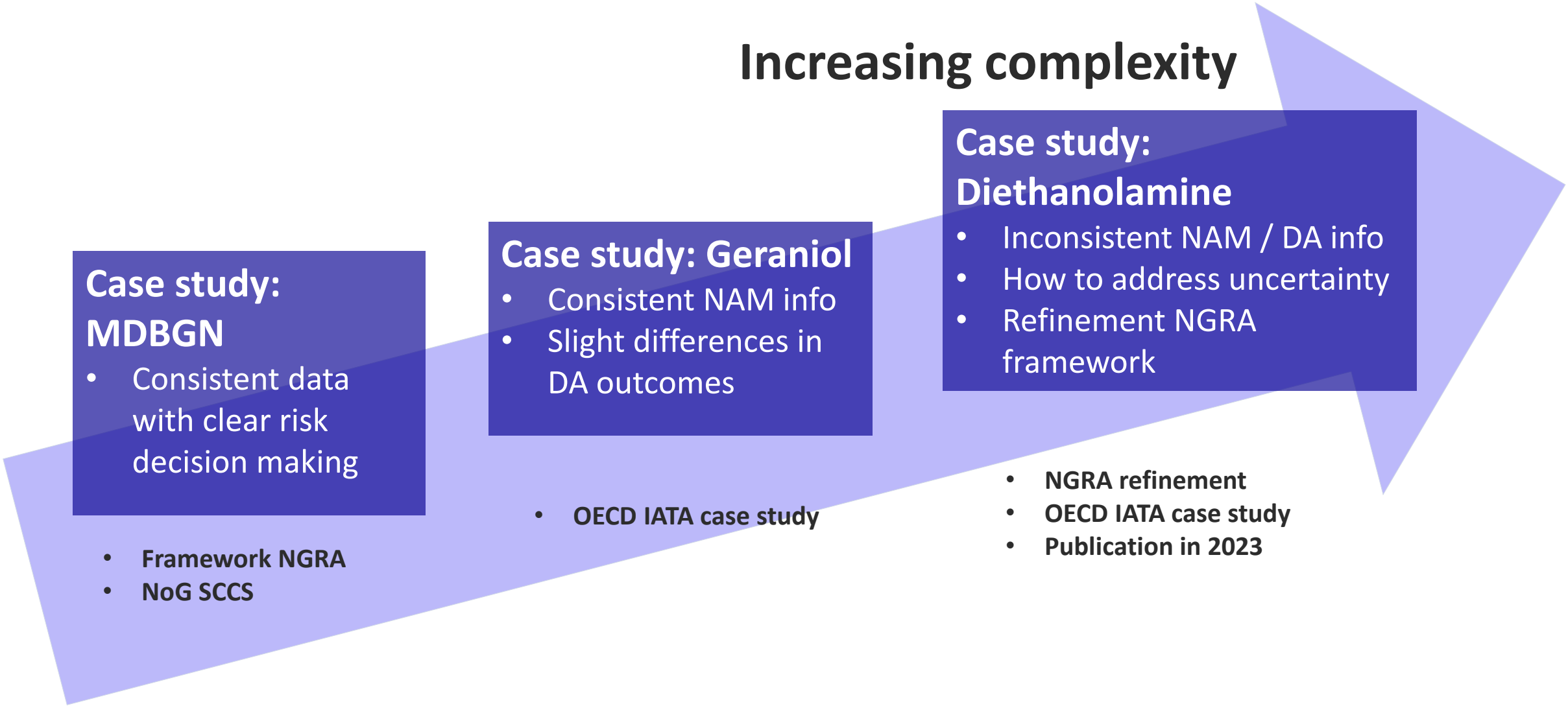
Skin sensitisation NGRA framework case studies



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NGRA case study scope

The aim of this case study was to explore the impact of inconsistent NAM information on the final risk assessment outcome for hypothetical (not representing real consumer exposures) exposure scenarios. The use of read across, including the use of analogue data, was considered out of scope to allow focus on how to deal with the inconsistent data in absence of analogues.

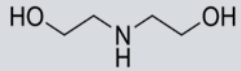
NGRA Tier 0 : Identify use scenarios

Exposure scenario was hypothetical to conduct consumer risk assessments to assess the potential risk induction of skin sensitisation; using the selected DA to derive a Point of Departure (POD) and to explore how to better address uncertainty in the risk assessment process.

- Rinse-off: exposure from use of **0.8% DEA in a shampoo** was calculated to be 0.6 µg/cm^2
- Leave-on: exposure from use of **0.8% DEA in a deodorant** was calculated to be 60 µg/cm^2
- Exposure-based waiving not applicable
- Aggregate exposure not considered
- Read –Across not considered

Product	Product applied (g/day)	Use level (%)	Skin retention	Skin surface (cm^2)	Consumer Exposure Level (µg/cm^2/d)
Shampoo	0,11	0.8	0,01	1 440	0,6
Deodorant (non-spray)	1.5	0.8	1	200	60

NGRA Tier 0 - steps 2&3:
Identify molecular structure, phys chem properties & existing information

Name	Diethanolamine
CAS number	111-42-2
SMILES	C(CO)NCCO
Structural formula	
Physicochemical properties	Molecular weight: 105.14 Da LogP: -1.43 LogS: 0.98 LogVP: -3.55 Boiling pt. [°C]: 268.8 Melting pt. [°C]: 28 Volatility[1]: semi-volatile pH: 10.3 LogD @ pH 7: -3.38 H2O solubility @ pH 7: 3 g/L Plasma protein binding (% bound): 11.3

No indication of applicability domain issues for *in vitro* / *in silico* NAMs based upon phys chem information.

Name	Diethanolamine	AOP KE addressed
Mechanistic domain based on expert review	Pro-Schiff base	KE-1
TIMES-SS (v2.30.1.11)	Parent: Non-sensitiser Metabolite: Non-sensitiser	KE-1
TOXTREE (v2.6.13) Skin sensitisation reactivity domains Protein binding alerts	No alert Schiff base formation	KE-1
OECD Toolbox (TB) (v4.4): https://qsartoolbox.org OASIS protein binding alerts for skin sensitisation Skin sensitisation automated workflow for DASS	No alert Negative (no analogues identified)	KE-1
DEREK 6.01 (Nexus 2.2.2)	Positive (Equivocal)[2]	KE-1
DPRA	Negative/minimal (Cys depl: 5.9% and Lys depl: 2.2%)	KE-1
KeratinoSens™	Negative (EC1.5: >2000 µM, EC3: >2000 µM, Imax: 1, IC50%: >2000 µM)	KE-2
U-SENS™	Positive (CD86 EC150: 26.9 µg/mL, CV70: >200 µg/ml)	KE-3
h-CLAT	Positive (CD86 EC150: 1242.5 µg/mL, CD54 EC200: 1280.9 µg/mL, CV75: 2277 µg/mL)	KE-3
Dermal penetration rate (from Brain et al. 2005; Kraeling et al. 2004)	Minimal <3%	

NOT an exhaustive list (what was collected for this case study)

➢ **TIER 0 : NO EXIT**

Exposure based waiving not applicable to both exposure scenarios

NGRA Tier 1: Hypothesis generation
How will the data be used in risk assessment?

The available NAM information (Tier 0) demonstrate inconsistent outcomes with respect to sensitisation potential of DEA.

- Two of the four *in silico* tools applied predicted no reactivity or skin sensitisation potential (TIMES-SS and OECD TB). Derek Nexus predicted that DEA being a skin sensitiser and ToxTree reported that DEA could form a Schiff base after activation.
- DPRA and KeratinoSens™ gave negative results while U-SENS™ and h-CLAT were positive according to the prediction models specified in the respective OECD TG.
- Due to the possibility that DEA could be a pro-hapten, the DPRA and KeratinoSens™ data need to be considered with caution.

➢ **TIER 1 : NO EXIT**

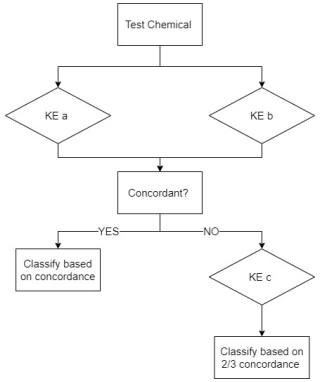
A weight of evidence assessment demonstrated that it is not possible to reach the conclusion with high certainty that DEA is a non-sensitiser

NGRA Tier 1: Hypothesis generation
7 Defined Approaches (DA) applied in Case Study for DEA

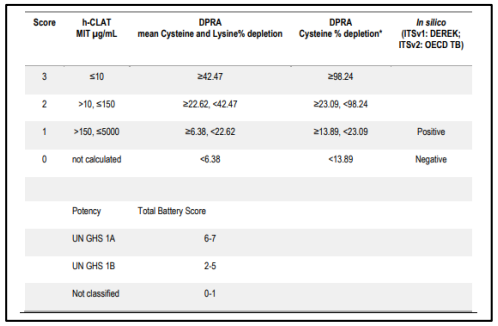
- DA were considered individually (no need to use more than one DA for NGRA)
- DA potency predictions and risk outcomes were compared

Hazard	Potency (GHS 1A/ 1B)	Potency grouping	Continuous PoD values

OECD (2021), Guideline No. 497



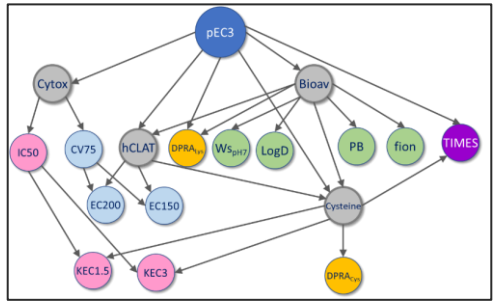
2 out of 3



Score	h-CLAT MIT µg/mL	DPRA mean Cysteine and Lysine% depletion	DPRA Cysteine % depletion*	*In silico* (ITSv1: DEREK; ITSv2: OECD TB)
3	≤10	≥42.47	≥98.24	
2	>10, ≤150	≥22.62, <42.47	≥23.09, <98.24	
1	>150, ≤5000	≥6.38, <22.62	≥13.89, <23.09	Positive
0	not calculated	<6.38	<13.89	Negative

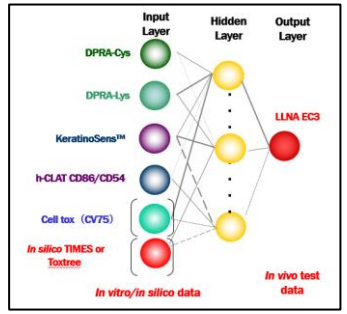
Potency	Total Battery Score
UN GHS 1A	6-7
UN GHS 1B	2-5
Not classified	0-1

ITSv1/v2



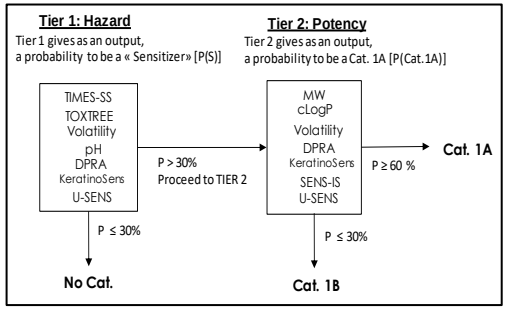
BN-ITS

(Jaworska et al. 2015, Kern et al. 2023)



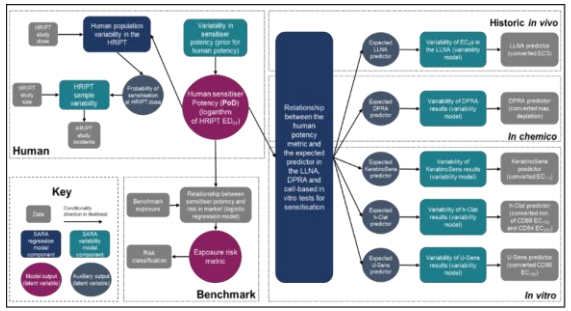
2x ANN EC3

(Hirota et al 2015, 2018)



Sequential Testing Strategy

(Tourneix et al. 2019)



SARA

(Reynolds et al. 2019, Gilmour et al 2022)

Tier 1: Hypothesis generation - 7 Defined Approaches (DA) outcomes

Defined Approach	DA prediction for DEA
ITSv1 DA	GHS Cat. 1B **skin sensitiser** (ITS score of 2)
ITSv2 DA	Inconclusive
ANN (TIMES-SS)	**Weak sensitiser** (EC3 value: 81.5%)
ANN (Toxtree)	**Weak sensitiser** (EC3 value: 59.1%)
Sequential testing strategy (STS)	Tier 1: Non-sensitiser (13% probability to be a sensitiser) Due to NS in Tier 1 potency prediction: Tier 2 not applicable
BN ITS	High probability (> 99%) to be a non-sensitiser (Bayes Factor: >30, strong evidence)
SARA	**Human sensitiser** potency ED01 = 13000 µg/cm^2 (95th % confidence interval 530 – 370000 µg/cm^2) SARA risk metric (Probability exposure is low risk) = 0.5

=> TIER 1 : NO EXIT
Non-sensitiser cannot be concluded with sufficient certainty

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Tier 2: Risk assessment based on 7 Defined Approaches (DA)

Defined Approach	DA prediction for DEA
ITSv1 DA	GHS Cat. 1B **skin sensitiser** (ITS score of 2).
ITSv2 DA	Inconclusive
ANN (TIMES-SS)	**Weak sensitiser** (EC3 value: 81.5%).
ANN (Toxtree)	**Weak sensitiser** (EC3 value: 59.1%).
Sequential testing strategy (STS)	Tier 1: Non-sensitiser (13% probability to be a sensitiser) Due to NS in Tier 1 potency prediction: Tier 2 not applicable.
BN ITS	High probability (> 99%) to be a non-sensitiser (Bayes Factor: >30, strong evidence).
SARA	**Human sensitiser** potency ED01 = 13000 µg/cm^2 (95th % confidence interval 530 – 370000 µg/cm^2) SARA risk metric (Probability exposure is low risk) = 0.5

TIER 2 :

- Convert to PoD for risk assessment
- Different NAM outcomes introduced uncertainty
- Therefore, a DA prediction of non-sensitiser was also converted into a PoD with the aim to further increase confidence in the risk assessment.

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NGRA Tier 2: Risk assessment – point of departure

0.8% in SHAMPOO	ITSv1	ITSv2	ANN (TIMES)	ANN (Toxtree)	STS	BN-ITS	SARA
	DA output						
DA ouput	Cat. 1B	Inconclusive	EC3=81.5 %	EC3=59.1 %	NS P(S)= 13%	NS P(NS)=99% BF (>30%)	ED_{01}=13000 µg/cm^2 (530–370000) µg/cm^2
PoD (µg/cm^2)	> 500	> 500	14 775	20 375	25 000	25 000	13 000

EC3 (%) is converted to µg/cm2 Using a standardised approach (Robinson *et al*. 2000, Griem *et al*. 2003)

NGRA Tier 2: Risk assessment – uncertainty assessment

0.8% in SHAMPOO	ITSv1	ITSv2	ANN (TIMES)	ANN (Toxtree)	STS	BN-ITS	SARA
	DA output						
DA ouput	Cat. 1B	Inconclusive	EC3=81.5 %	EC3=59.1 %	NS P(S)= 13%	NS P(NS)=99% BF (>30%)	ED_{01}=13000 µg/cm^2 (530–370000) µg/cm^2
PoD (µg/cm^2)	> 500	> 500	14 775	20 375	25 000	25 000	13 000
	Calculate MoE for 0.8% in SHAMPOO						
Consumer exposure level (µg/cm^2)	0,6	0,6	0,6	0,6	0,6	0,6	0,6
MoE (PoD/CEL) P(low risk)*SARA ONLY	> 833	> 833	33 958	24 625	41 667	41 667	24 000

EC3 (%) is converted to µg/cm2 Using a standardised approach (Robinson *et al*. 2000, Griem *et al*. 2003)

MoE = PoD / CEL

NGRA Tier 2: Risk assessment – uncertainty assessment

0.8% in SHAMPOO	ITSv1	ITSv2	ANN (TIMES)	ANN (Toxtree)	STS	BN-ITS	SARA
DA output							
DA ouput	Cat. 1B	Inconclusive	EC3=81.5 %	EC3=59.1 %	NS P(S)= 13%	NS P(NS)=99% BF (>30%)	ED_{01}=13000 µg/cm² (530–370000) µg/cm²
PoD (µg/cm²)	> 500	> 500	14 775	20 375	25 000	25 000	13 000
Calculate MoE for 0.8% in SHAMPOO							
Consumer exposure level (µg/cm²)	0,6	0,6	0,6	0,6	0,6	0,6	0,6
MoE (PoD/CEL) P(low risk)*SARA ONLY	> 833	> 833	33 958	24 625	41 667	41 667	24 000
Weight of evidence assessment / Characterise uncertainty							
WoE : confidence in NAM	Moderate						
WoE : Conservatism in transformation of DA outcome to PoD	Unknown		Low		High	High	Low
WoE: MoE certainty P(low risk)*SARA ONLY	Low	Low	High	High	High	High	Low P (low risk) = 0.5
Risk assessment outcome	**Safe**						

EC3 (%) is converted to µg/cm2 Using a standardised approach (Robinson *et al*. 2000, Griem *et al*. 2003)

MoE = PoD / CEL

Confidence in NAM
Conservatism in DA → PoD
Size of MoE

Shampoo (0,8%)

SAFE use, regardless of PoD determination based on individual DA

NGRA Tier 2: Risk assessment – uncertainty assessment

0.8% DEODORANT	ITSv1	ITSv2	ANN (TIMES)	ANN (Toxtree)	STS	BN-ITS	SARA
	DA output						
DA ouput	Cat. 1B	Inconclusive	EC3=81.5 %	EC3=59.1 %	NS P(S)= 13%	NS P(NS)=99% BF (>30%)	ED_{01}=13000 µg/cm^2 (530–370000) µg/cm^2
PoD (µg/cm^2)	> 500	> 500	14 775	20 375	25 000	25 000	13 000
	Calculate MoE for 0.8% in NON-SPRAY DEODORANT						
Consumer exposure level (µg/cm^2)	60	60	60	60	60	60	60
MoE (PoD/CEL) P(low risk)*SARA ONLY	> 8	> 8	246	340	416	416	217 (8.8-617)

NGRA Tier 2: Risk assessment – uncertainty assessment

0.8% in deodorant	ITSv1	ITSv2	ANN (TIMES)	ANN (Toxtree	STS	BN-ITS	SARA
DA output							
DA ouput	Cat. 1B	Inconclusive	EC3=81.5 %	EC3=59.1 %	NS P(S)= 13%	NS P(NS)=99% BF (>30%)	ED_{01}=13000 µg/cm^2 (530–370000) µg/cm^2
PoD (µg/cm^2)	> 500	> 500	14 775	20 375	25 000	25 000	13 000
Calculate MoE for 0.8% in NON-SPRAY DEODORANT							
Consumer exposure level (µg/cm^2)	60	60	60	60	60	60	60
MoE (PoD/CEL)	>8	>8	246	340	416	416	217 (8.8-617)
Weight of evidence assessment / Characterise uncertainty							
WoE : confidence in NAM	Moderate						
WoE : Conservatism in transformation of DA outcome to PoD	Unknown		Low		High	High	Low
WoE: MoE certainty **P(low risk)*SARA ONLY**	Low	Low	High	High	High	High	Low P (low risk) = 0.5
Risk assessment outcome	UNSAFE	UNSAFE	SAFE	SAFE	SAFE	SAFE	UNSAFE

Deodorant (0,8%)

SAFE/UNSAFE use, regardless of PoD determination based on individual DA

Case Study Conclusions

- DEA suitable case study molecule due to inconsistencies in the existing NAM information.
- Information from NAMs can be applied within a WoE (following the NGRA framework) to reach a conclusion on consumer risk.
- Information regarding the e.g. reaction chemistry is a critical element to understand the applicability domain of the NAM.
- DEA was predicted to be a pro-hapten which introduced uncertainty in the use of some NAM information within DA and decision making.
- In order to reach a decision on safety using NAM we have calculated a MoE and then evaluated possible areas of uncertainty
  - applicability domain of NAM and the impact this could have on DA outcome
  - relative conservatism in deriving a PoD from the DA outcome.
- Whilst the inconsistencies in the NAM information led to differences in the DA outputs, there was less impact on the risk assessment outcomes:
  - 4 of the 7 applied DA resulted in a conclusion of safe (STS, BN-ITS and the two ANN versions)
  - 3 resulted in a conclusion of un-safe (ITSv1, ITSv2, SARA)

To be continued

- More case studies & stakeholder exchanges (e.g. read-across to be addressed)

- Sources of uncertainty

    - NAM applicability, *in silico* tool selection & versions,
    - Conservatism in DA outcome transformation to PoD
    - MoE-approach to uncertainty assessment was introduced

…

Acknowledgements

ICCS Skin Sensitisation Working Group

Thank you for your attention

OBRIGADA !

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