BN-ITS Evaluation of RCPL dataset

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IDEA wokshop

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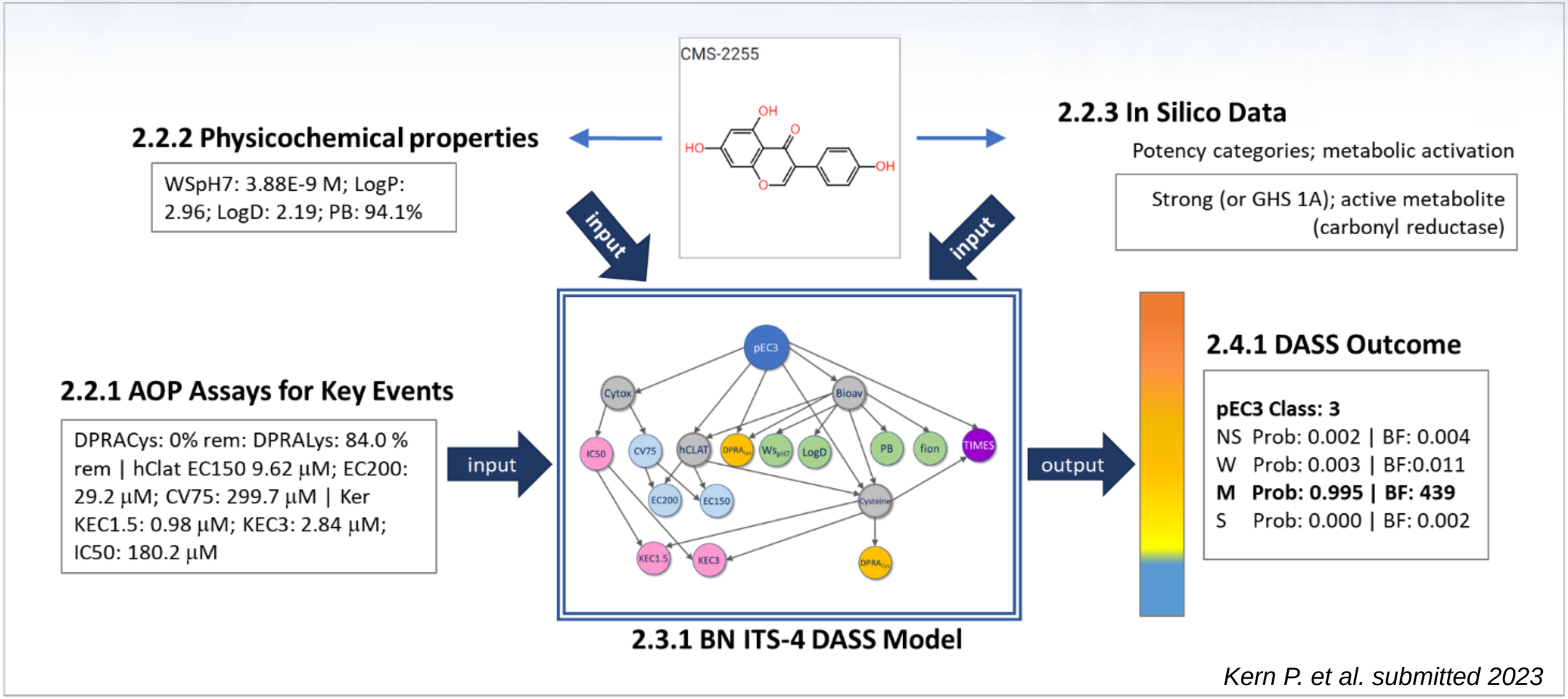
Bayesian Network ITS



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Disclaimer: Dataset and Category prediction

- 257 chemicals with LLNA data
- BN-ITS predicts ONLY potency categories based on LLNA
- Conversion to a POD can be done, but how is a question of data/ information need. Not an automated process.

pEC3 Class	Cut-offs	Corresponding potency categories
1	pEC3 ≤ -1.90 (EC3% ≥ 50)	Non-sensitizers
2	-1.90 < pEC3 ≤ -1.13 (10 ≤ EC3% < 50)	Weak sensitizer
3	-1.13< pEC3 ≤ -0.20 (1 ≤ EC3% < 10)	Moderate-Strong sensitizer
4	-0.20 < pEC3 (EC3% < 1)	Strong sensitizer

BN-ITS input variables

Input type	Endpoint	Unit
Bioavailability/ physicochemical properties (ACD/Labs or OPERA)	1. Ws: Water solubility at pH=7 2. LogD: Distribution coefficient at pH=7* 3. LogP: Octanol-water distribution coefficient* 4. Plasma protein binding in % 5. Fraction ionized (calculated in R-code)*	M (mol/L) dimensionless dimensionless dimensionless
In silico data (TIMES-SS or ToxGPS)	Data given in 3 classes (Non-sensitizer (1), weak (2) or moderate/strong (3). TIMES-SS: most potent results among parent and generated metabolites. ToxGPS: results from reactivity classes including metabolic potential.	dimensionless classes (NS, W, S)
KE 1: DPRACys, DPRALys	% cysteine- (Cys), and lysine- (Lys) peptide remaining in the DPRA	% remaining peptide
KE 2: KEC1.5, KEC3, IC50	• 1.5-fold (KEC1.5) and 3-fold (KEC3) Induction of Nrf2‡-dependent luciferase activity in the KeratinoSens™ assay • 50% reduction in cell viability in the KeratinoSens™ assay	µM (µmol/L)
KE 3: EC150, EC200, CV75	• 150% induction of the cell surface activation marker CD86 in the h-CLAT; • 200% induction of the cell surface activation marker CD54 in the h-CLAT; • 25% reduction in cell viability in the h-CLAT.	µM (µmol/L)

Predictivity versus LLNA

- BN-ITS predicts LLNA based potency classes
- Overall- good correlation for the RCPL set with the LLNA data
- 8 chemicals underpredicted
- Need to review predictions in more detail
- Consider Bayes factors for decision making
- Conversion to POD in μg/cm^2
  - Data discordancies?
  - Analog information?

Name	CAS #	LLNA group	Prediction	BN prediction	Lower Bayes Factor
Methyl 2-octynoate (Methyl heptine carbonate)	111-12-6	4	C3	Moderate	X
1,4-Phenylenediamine (PPD)	106-50-3	4	C3	Moderate	X
Benzaldehyde	100-52-7	1	C1	Non-sensitizer	
Cinnamic aldehyde	104-55-2	3	C4	Strong or extreme	X
p-Mentha-1,8-dien-7-al (Perillaldehyde)	2111-75-3	3	C4	Strong or extreme	X
Phenylacetaldehyde	122-78-1	3	C3	Moderate	
Allyl phenoxyacetate	7493-74-5	3	C1	Non-sensitizer	
Methyl 2-nonynoate (Methyl octine carbonate)	111-80-8	3	C3	Moderate	
Benzyl Alcohol	100-51-6	1	C2	Weak	
1,1,3-Trimethyl-2-formylcyclohexa-2,4-diene (Safranal)	116-26-7	3	C3	Moderate	
Vanillin	121-33-5	1	C1	Non-sensitizer	
Isoeugenol	97-54-1	3	C3	Moderate	
2,4-dinitrochlorobenzene (DCNB)	97-00-7	4	C4	Strong or extreme	
Coumarin	91-64-5	1	C1	Non-sensitizer	
Benzyl salicylate	118-58-1	3	C2	Weak	X
Benzyl bromide	100-39-0	4	C4	Strong or extreme	X
Citral	5392-40-5	3	C3	Moderate	
Glutaraldehyde	111-30-8	4	C3	Moderate	X
Methyl salicylate	119-36-8	1	C1	Non-sensitizer	
3-Propylidenephthalide	17369-59-4	3	C3	Moderate	X
Hexyl cinnamic aldehyde	101-86-0	3	C3	Moderate	
Geraniol	106-24-1	2	C3	Moderate	
Isomethylionone (α-)	127-51-5	2	C2	Weak	X
Lyral (HICC)	31906-04-4	2	C2	Weak	
Benzyl benzoate	120-51-4	2	C2	Weak	
Eugenol	97-53-0	2	C2	Weak	
5-chloro-2-methyl-4-isothiazolin-one (CMIT)	26172-55-4	4	C4	Strong or extreme	
1,4-Dihydroquinone	123-31-9	4	C3	Moderate	
Carvone	6485-40-1	2	C3	Moderate	X
trans-2-Hexenal	6728-26-3	3	C3	Moderate	X
Cinnamic alcohol	104-54-1	2	C3	Moderate	
Hydroxycitronellal	107-75-5	2	C1	Non-sensitizer	
4-Hydroxy-2,5-dimethyl-3(2H)-furanone (Furaneol)	3658-77-3	3	C1	weak	

Predictivity versus PV

- Overall, still good match- some differences
- But that's expected…
- Comparison is done by grouping PV values
- 8 underpredictions
- Differences due to Human versus LLNA data driving PV
- Conversion to POD and then compare

Name	CAS #	PV value (ug/cm2)	BN prediction
Methyl 2-octynoate (Methyl heptine carbonate)	111-12-6	125	Moderate
1,4-Phenylenediamine (PPD)	106-50-3	3.9	Moderate
Benzaldehyde	100-52-7	4094	Non-sensitizer
Cinnamic aldehyde	104-55-2	885	Strong or extreme
p-Mentha-1,8-dien-7-al (Perillaldehyde)	2111-75-3	2175	Strong or extreme
Phenylacetaldehyde	122-78-1	750	Moderate
Allyl phenoxyacetate	7493-74-5	775	Non-sensitizer
Methyl 2-nonynoate (Methyl octine carbonate)	111-80-8	109	Moderate
Benzyl Alcohol	100-51-6	>25000	Weak
1,1,3-Trimethyl-2-formylcyclohexa-2,4-diene (Safranal)	116-26-7	106	Moderate
Vanillin	121-33-5	No PV derived - very weak/nonsensitiser	Non-sensitizer
Isoeugenol	97-54-1	325	Moderate
2,4-dinitrochlorobenzene (DCNB)	97-00-7	3.4	Strong or extreme
Coumarin	91-64-5	11792	Non-sensitizer
Benzyl salicylate	118-58-1	17715	Weak
Benzyl bromide	100-39-0	50	Strong or extreme
Citral	5392-40-5	1450	Moderate
Glutaraldehyde	111-30-8	20	Moderate
Methyl salicylate	119-36-8	No PV derived- very weak/nonsensitiser	Non-sensitizer
3-Propylidenephthalide	17369-59-4	925	Moderate
Hexyl cinnamic aldehyde	101-86-0	23620	Moderate
Geraniol	106-24-1	9197	Moderate
Isomethylionone (α-)	127-51-5	>25000	Weak
Lyral (HICC)	31906-04-4	4275	Weak
Benzyl benzoate	120-51-4	>25000	Weak
Eugenol	97-53-0	7357	Weak
5-chloro-2-methyl-4-isothiazolin-one (CMIT)	26172-55-4	2.3	Strong or extreme
1,4-Dihydroquinone	123-31-9	47.5	Moderate
Carvone	6485-40-1	17573	Moderate
trans-2-Hexenal	6728-26-3	39.3	Moderate
Cinnamic alcohol	104-54-1	5775	Moderate
Hydroxycitronellal	107-75-5	5275	Non-sensitizer
4-Hydroxy-2,5-dimethyl-3(2H)-furanone (Furaneol)	3658-77-3	1181	weak

Potency Ranking

- Differences can be due to some category boundaries e.g. for weak/ NS, as well as strong/ moderate.

Name	CAS #	RCPL Potency ug/cm2	BN prediction	LLNA group
5-chloro-2-methyl-4-isothiazolin-one (CMIT)	26172-55-4	2.3	Strong or extreme	4
2,4-dinitrochlorobenzene (DCNB)	97-00-7	3.4	Strong or extreme	4
1,4-Phenylenediamine (PPD)	106-50-3	3.9	Moderate	4
Glutaraldehyde	111-30-8	20	Moderate	4
trans-2-Hexenal	6728-26-3	39.3	Moderate	3
1,4-Dihydroquinone	123-31-9	47.5	Moderate	4
Benzyl bromide	100-39-0	50	Strong or extreme	4
1,1,3-Trimethyl-2-formylcyclohexa-2,4-diene (Safranal)	116-26-7	106	Moderate	3
Methyl 2-nonynoate (Methyl octine carbonate)	111-80-8	109	Moderate	3
Methyl 2-octynoate (Methyl heptine carbonate)	111-12-6	125	Moderate	4
Isoeugenol	97-54-1	325	Moderate	3
Phenylacetaldehyde	122-78-1	750	Moderate	3
Allyl phenoxyacetate	7493-74-5	775	Non-sensitizer	3
Cinnamic aldehyde	104-55-2	885	Strong or extreme	3
3-Propylidenephthalide	17369-59-4	925	Moderate	3
4-Hydroxy-2,5-dimethyl-3(2H)-furanone (Furaneol)	3658-77-3	1181	weak	3
Citral	5392-40-5	1450	Moderate	3
p-Mentha-1,8-dien-7-al (Perillaldehyde)	2111-75-3	2175	Strong or extreme	3
Benzaldehyde	100-52-7	4094	Non-sensitizer	1
Lyral (HICC)	31906-04-4	4275	Weak	2
Hydroxycitronellal	107-75-5	5275	Non-sensitizer	2
Cinnamic alcohol	104-54-1	5775	Moderate	2
Eugenol	97-53-0	7357	Weak	2
Geraniol	106-24-1	9197	Moderate	2
Coumarin	91-64-5	11792	Non-sensitizer	1
Carvone	6485-40-1	17573	Moderate	2
Benzyl salicylate	118-58-1	17715	Weak	3
Hexyl cinnamic aldehyde	101-86-0	23620	Moderate	3
Benzyl Alcohol	100-51-6	>25000	Weak	1
Isomethylionone (α-)	127-51-5	>25000	Weak	2
Benzyl benzoate	120-51-4	>25000	Weak	2
Vanillin	121-33-5	NS	Non-sensitizer	1
Methyl salicylate	119-36-8	NS	Non-sensitizer	1

Conversion of Category Prediction into POD – some approaches

Potency Category	LLNA EC3 % ranges (ECETOC)	LLNA EC3 % conversion to µg/cm² (based on LLNA dosing*)	Default NESIL µg/cm² to be used as POD
Extreme (Potent)	< 0.1	< 25	1
Strong	≥ 0.1 - < 1	≥25 - <250	10
Moderate	≥ 1 - < 10	≥250 - <2500	100
Weak	≥ 10 - ≤ 100	≥2500 - <25000	1000
GHS 1A	< 2%	< 500	< 500
GHS 1B	> 2%	> 500	> 500

- Conversion of LLNA categories or EC3% to µg/cm² using a conversion factor (1% = 250 µg/cm²)
- GHS categorization models limited in deriving POD (only < or > 500 µg/cm²)
- Some methods (Bayesian) allow confidence evaluation around predicted value

Conversion to POD categories

- When converting to POD, using categories, only a few are underpredicted.
- Default POD conservative for most

Name	CAS #	PV value (ug/cm2)	Default POD (ug/cm2)	POD based LLNA conversion categories
Methyl 2-octynoate (Methyl heptine carbonate)	111-12-6	125	100	250-2500
1,4-Phenylenediamine (PPD)	106-50-3	3.9	100	250-2500
Benzaldehyde	100-52-7	4094	>10000	>12500
Cinnamic aldehyde	104-55-2	885	1 or 10	< 250
p-Mentha-1,8-dien-7-al (Perillaldehyde)	2111-75-3	2175	1 or 10	< 250
Phenylacetaldehyde	122-78-1	750	100	250-2500
Allyl phenoxyacetate	7493-74-5	775	>10000	>12500
Methyl 2-nonynoate (Methyl octine carbonate)	111-80-8	109	100	250-2500
Benzyl Alcohol	100-51-6	>25000	1000	2500 - 12500
1,1,3-Trimethyl-2-formylcyclohexa-2,4-diene (Safranal)	116-26-7	106	100	250-2500
Vanillin	121-33-5	No PV derived - very weak/nonsensitiser	>10000	>12500
Isoeugenol	97-54-1	325	100	250-2500
2,4-dinitrochlorobenzene (DCNB)	97-00-7	3.4	1 or 10	< 250
Coumarin	91-64-5	11792	>10000	>12500
Benzyl salicylate	118-58-1	17715	1000	2500 - 12500
Benzyl bromide	100-39-0	50	1 or 10	< 250
Citral	5392-40-5	1450	100	250-2500
Glutaraldehyde	111-30-8	20	100	250-2500
Methyl salicylate	119-36-8	No PV derived- very weak/nonsensitiser	>1000	>12500
3-Propylidenephthalide	17369-59-4	925	100	250-2500
Hexyl cinnamic aldehyde	101-86-0	23620	100	250-2500
Geraniol	106-24-1	9197	100	250-2500
Isomethylionone (α-)	127-51-5	>25000	1000	2500 - 12500
Lyral (HICC)	31906-04-4	4275	1000	2500 - 12500
Benzyl benzoate	120-51-4	>25000	1000	2500 - 12500
Eugenol	97-53-0	7357	1000	2500 - 12500
5-chloro-2-methyl-4-isothiazolin-one (CMIT)	26172-55-4	2.3	1 or 10	< 250
1,4-Dihydroquinone	123-31-9	47.5	100	250-2500
Carvone	6485-40-1	17573	100	250-2500
trans-2-Hexenal	6728-26-3	39.3	100	250-2500
Cinnamic alcohol	104-54-1	5775	100	250-2500
Hydroxycitronellal	107-75-5	5275	>10000	>12500
4-Hydroxy-2,5-dimethyl-3(2H)-furanone (Furaneol)	3658-77-3	1181	1000	2500 - 12500

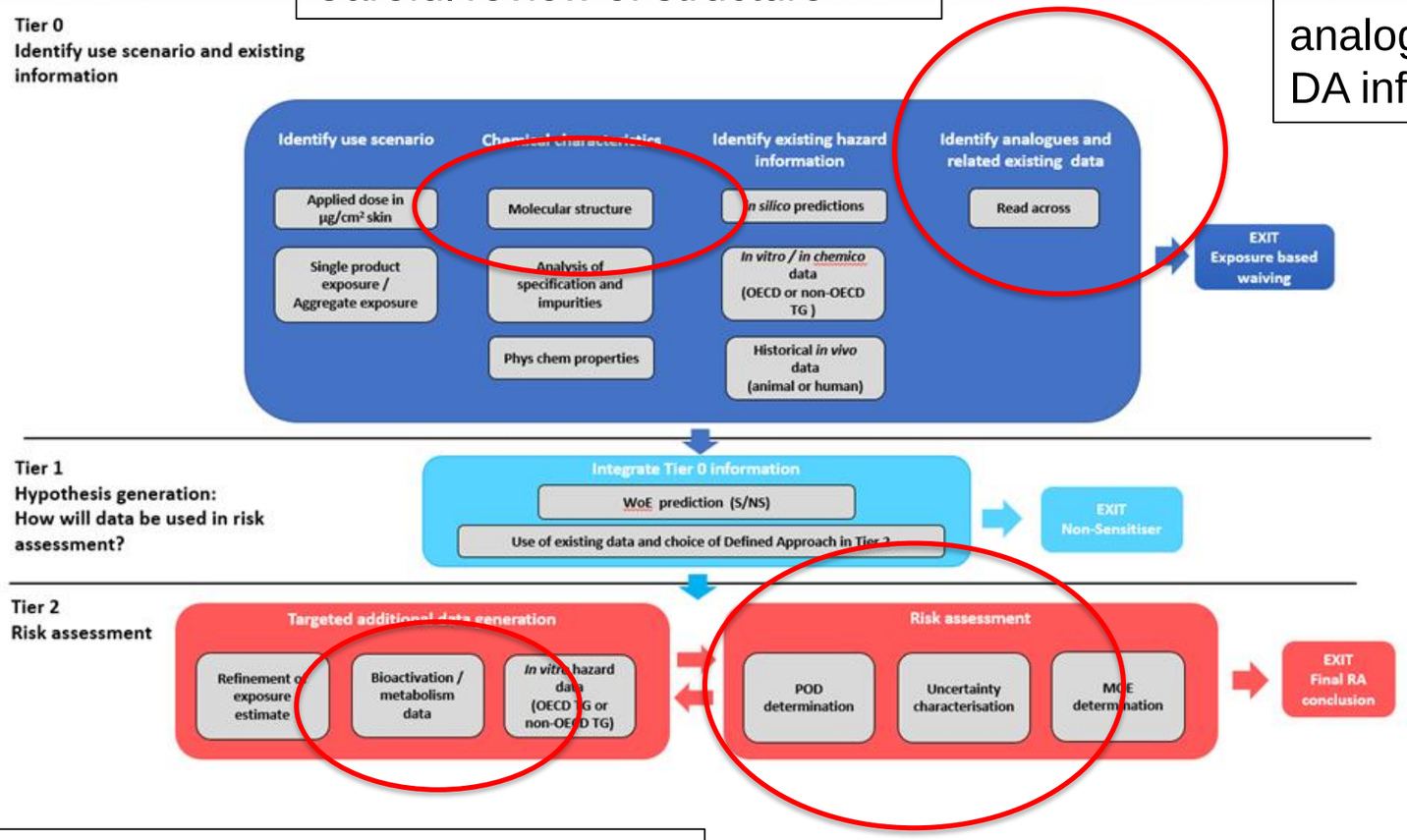
How do we use the BN-ITS results?

- We never use the BN-ITS results in isolation
- Some considerations prior to decision making
  - Data completeness and consistancy (input data)
  - BN-ITS prediction and BF
  - Structure review- indication for pro/ pre hapten and potential for false negative results in NAM information
  - Structure review – new/ unknown structural features?
  - Prediction of potential more reactive metabolites
  - Review suitable analog materials and compare hazard data (also NAM, if available)
  - Analog data can influence POD in both directions
  - Decide on a conversion to POD in context of an NGRA case
  - POD set using a weight of evidence of all available data
  - Build in different levels of conservatism in POD, depending on application and data availability

Use of BN-ITS in NGRA to derive POD

Careful review of structure

Review of suitable analogs to support NAM/ DA information



Consider potential skin metabolism and formation of reactive metabolites

Predict potency category and convert to POD, considering uncertainty of all data and NGRA question to be answered

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More refined POD

- Possibility to use %tile probability to set POD.
- Default POD in line (or even more conservative) than 70th %tile.
- 90th %tile very conservative.
- Applied when needed

Name	CAS #	PV value (ug/cm2)	Default POD	50th	70th	80th	90th
Methyl 2-octynoate (Methyl heptine carbonate)	111-12-6	125	100	608.3	331.2	133.5	4.3
1,4-Phenylenediamine (PPD)	106-50-3	3.9	100	366.4	215.3	51.2	2.0
Benzaldehyde	100-52-7	4094	>10000	28235.6	17310.0	13359.7	10612.0
Cinnamic aldehyde	104-55-2	885	1 or 10	220.3	12.5	2.5	0.5
p-Mentha-1,8-dien-7-al (Perillaldehyde)	2111-75-3	2175	1 or 10	106.3	7.0	1.9	0.5
Phenylacetaldehyde	122-78-1	750	100	537.9	341.0	251.6	221.7
Allyl phenoxyacetate	7493-74-5	775	>10000	48281.0	28759.1	22196.1	17130.7
Methyl 2-nonynoate (Methyl octine carbonate)	111-80-8	109	100	812.6	541.9	442.5	361.3
Benzyl Alcohol	100-51-6	>25000	1000	4231.4	2517.6	1988.3	1570.3
1,1,3-Trimethyl-2-formylcyclohexa-2,4-diene (Safranal)	116-26-7	106	100	725.7	483.9	395.1	322.6
Vanillin	121-33-5	No PV derived - very weak/nonsensitiser	>10000	39334.3	23430.0	18083.1	14363.9
Isoeugenol	97-54-1	325	100	793.2	528.9	431.9	352.7
2,4-dinitrochlorobenzene (DCNB)	97-00-7	3.4	1 or 10	25.9	3.1	1.1	0.4
Coumarin	91-64-5	11792	>10000	41187.8	25987.8	20642.8	16397.2
Benzyl salicylate	118-58-1	17715	1000	3015.7	1159.8	735.2	466.0
Benzyl bromide	100-39-0	50	1 or 10	221.6	10.8	2.6	0.5
Citral	5392-40-5	1450	100	735.4	490.3	390.4	318.8
Glutaraldehyde	111-30-8	20	100	330.8	194.3	31.7	1.3
Methyl salicylate	119-36-8	No PV derived- very weak/nonsensitiser	>1000	43378.2	27056.5	21491.8	17071.5
3-Propylidenephthalide	17369-59-4	925	100	687.1	424.7	329.6	13.4
Hexyl cinnamic aldehyde	101-86-0	23620	100	1279.7	790.9	613.9	488.8
Geraniol	106-24-1	9197	100	624.1	299.4	39.9	1.9
Isomethylionone (α-)	127-51-5	>25000	1000	4918.3	2926.3	1654.1	813.9
Lyral (HICC)	31906-04-4	4275	1000	5509.5	3356.3	2284.9	918.0
Benzyl benzoate	120-51-4	>25000	1000	6558.8	4288.7	3387.1	2804.3
Eugenol	97-53-0	7357	1000	4727.4	3091.2	2441.3	1189.5
5-chloro-2-methyl-4-isothiazolin-one (CMIT)	26172-55-4	2.3	1 or 10	21.1	2.5	0.8	0.4
1,4-Dihydroquinone	123-31-9	47.5	100	531.9	345.8	282.4	236.5
Carvone	6485-40-1	17573	100	911.4	549.2	437.2	339.4
trans-2-Hexenal	6728-26-3	39.3	100	387.1	195.4	31.0	1.5
Cinnamic alcohol	104-54-1	5775	100	600.7	362.0	281.0	63.5
Hydroxycitronellal	107-75-5	5275	>10000	45836.3	28100.2	22320.8	17227.0
4-Hydroxy-2,5-dimethyl-3(2H)-furanone (Furaneol)	3658-77-3	1181	1000	4351.6	2650.9	2093.6	1392.0

Conclusions

- BN-ITS allows prediction of potency categories based on LLNA
- Bayesian DA enable experimental data variability to be modelled and uncertainty in POD & risk metrics can be factured into decision making
- Comparison with PV can only be done easily by ranking/ grouping.
- Non matching predictions often due to differences in human/ LLNA data and categorization
- Potency categories can be converted to POD (based on BN prediction only) and compared with PV, but usually POD are set based on a WoE considering more information in context of an NGRA.
- New areas of uncertainties need to be considered.
- Approach can differ depending on case study for which data are used: hazard , NGRA etc.