

# **Prediction of Specific Target Organ Toxicity after Repeated Exposure (STOT-RE) Using NAMs**

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# With Grateful Thanks to

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 CEFIC LRI AIMT8



# Specific Target Organ Toxicant – Repeated Exposure (STOT-RE)

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- STOT-RE Classifications
  - Category 1:  $\text{NOEL} \leq 10 \text{ mg/kg bw/d}$
  - Category 2:  $10 < \text{NOEL} \leq 100 \text{ mg/kg bw/d}$
  - Not Classified

***... And Can We Predict It?***

# Specific Target Organ Toxicant – Repeated Exposure (STOT-RE) ... *And Can We Predict It?*

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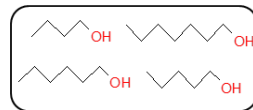
- Assumed to be driven by organ level toxicity
- Much recent focus on the liver
- Possibility to extend the AOP concept to the regulatory context
- Can NAM data support these classifications?



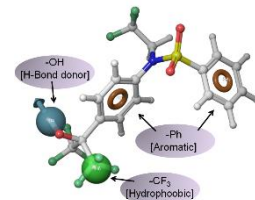
# Aim of This Study



**NAMs**



**Grouping**



***In Silico***

**Can We Predict / Gain  
Insights on Regulatory  
Classifications?**

**STOT-RE**



# Data: Compounds to Assess

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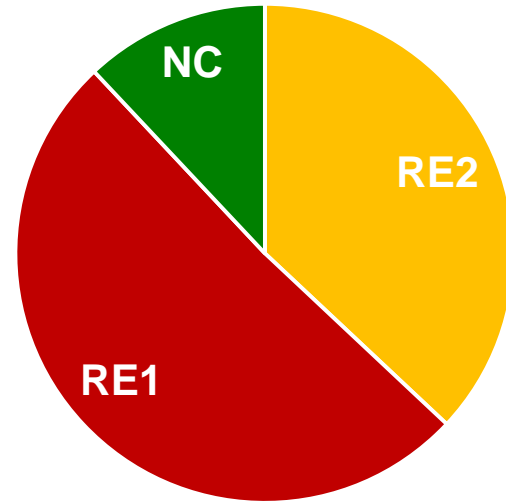
 **COSMOS**

**HESS DB**

 **RepDose**

**ToxRefDB**

90 day rat oral NOELs for  
1160 compounds



# Data: NAMs

- Freely available US EPA ToxCast data

**Toxicity Forecaster (ToxCast™)**

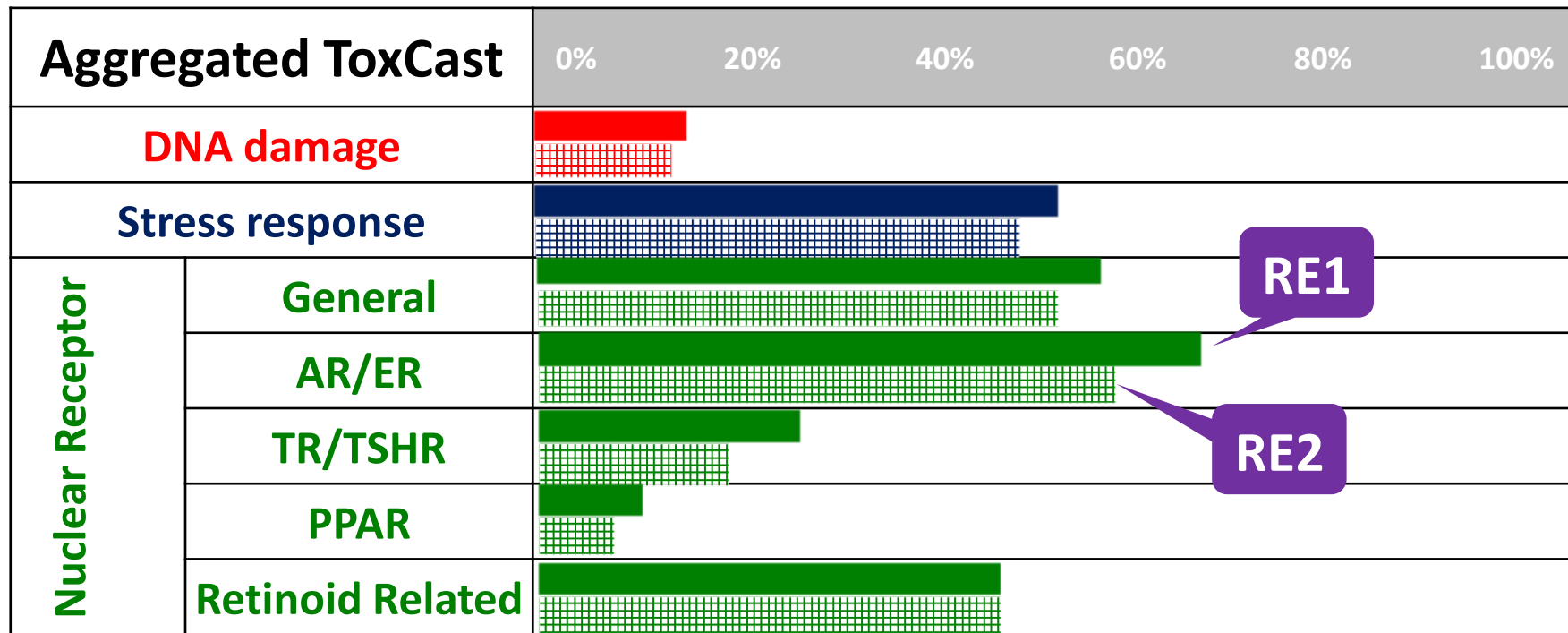
- Results aggregated to enable easier analysis

With thanks to:



Aggregated ToxCast		Coverage
DNA damage		60%
Stress response		54%
Nuclear Receptor	General	53%
	AR/ER	54%
	TR/TSHR	53%
	PPAR	51%
	Retinoid Related	46%
Cytotoxicity		21%

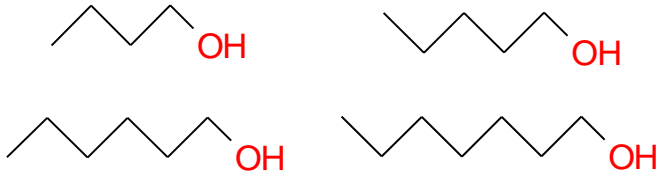
# Relationships of NAM Data to STOT-RE Classifications



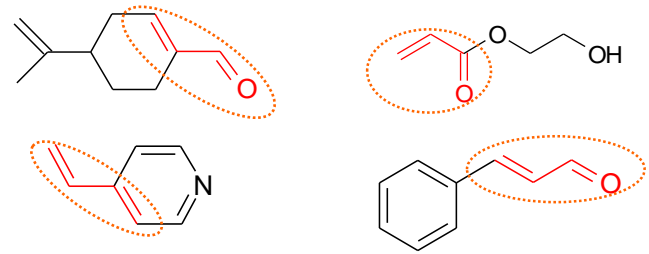


# Grouping Similar Molecules for Read-Across

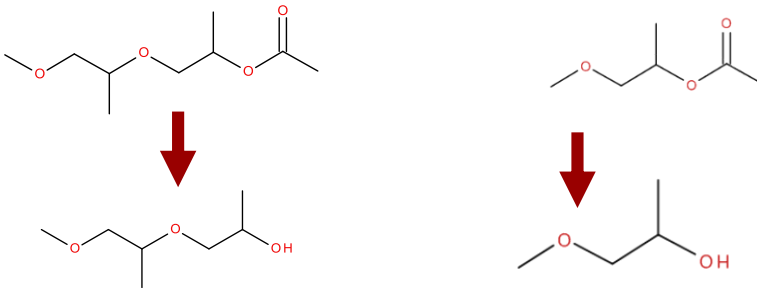
## Structural



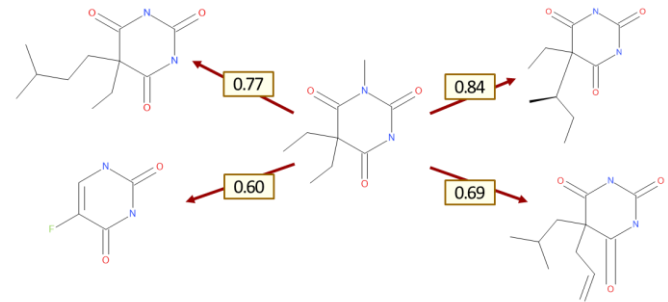
## Mechanistic / Mode of Action



## Common Metabolites / Degradants

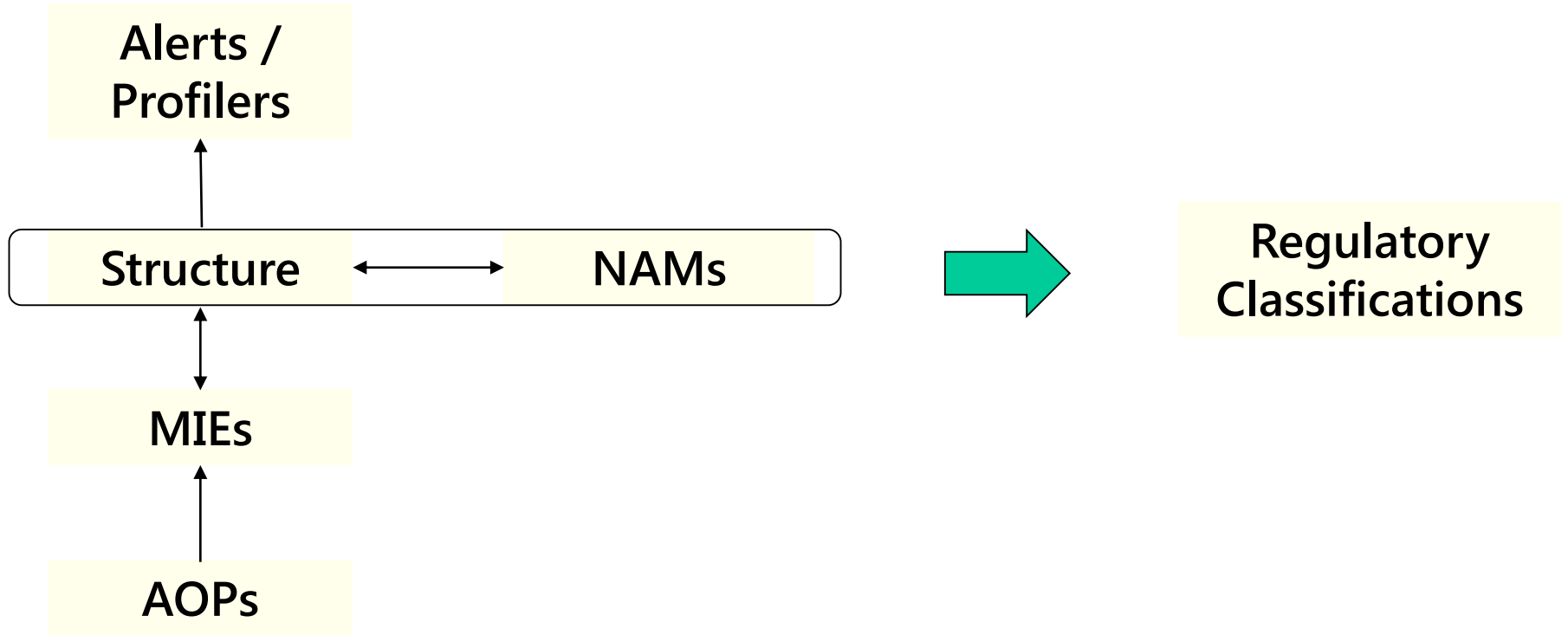


## Molecular Similarity



# Use of Grouping to Support Classifications

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# Use of Grouping to Support Classifications

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Manual grouping based on  
chemical and toxicological  
expertise works best!

# Significant Groupings With STOT-RE Data

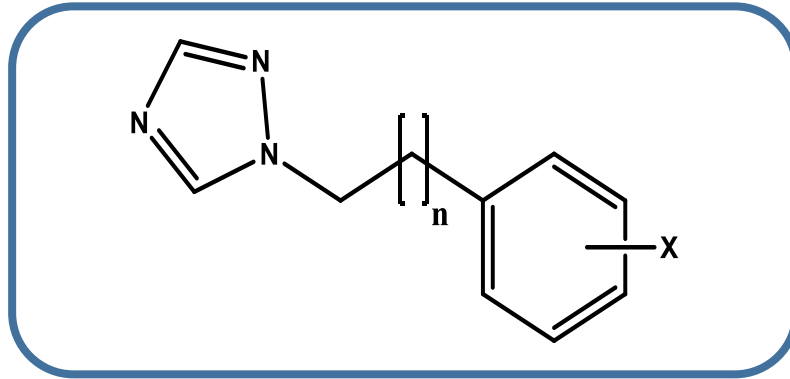
Grouping	No of Compounds
Nitrobenzene/aniline	90
Azole antifungal	21
Pyrethroid	20
Haloalkane	19
Stannane	13
Alkylated phenol	12
Glycol (ethylene)	11
Chlorophenoxy	10

Grouping	No of Compounds
Perfluorinated	10
Haloalkene	9
Benzoquinone	8
Glycol (propylene)	7
Phthalate ester	7
Thiourea	5
Haloacetic acid	4

- How Big?
- How to Define?

# Azole Antifungals

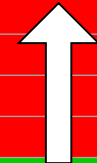
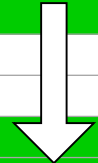
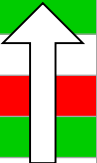
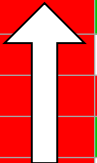
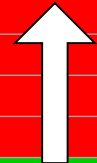
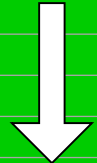
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- **Hepatotoxicity** shared across group
- CYP51A1 forms putative target
- Steroidogenic dysfunction, notably in production of cholesterol

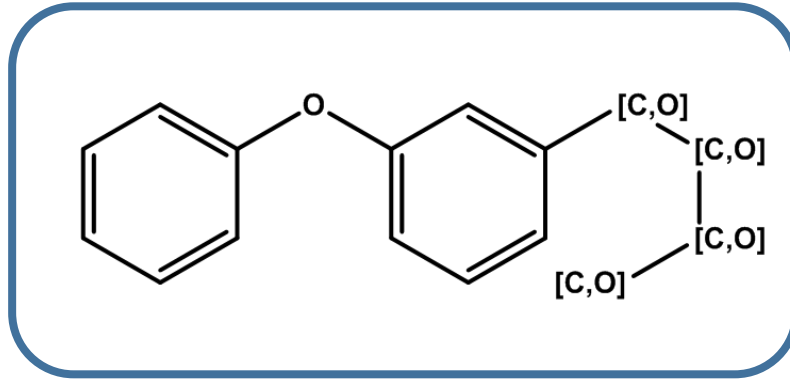
# Azole Antifungals – Role of NAMs

	STOT RE	Organ/ systemic	Liver Effect	Cytotox	DNA damage	Stress response	Nuclear Receptor				Retinoid Related
							General	AR/ ER	TR/ TSHR	PPAR	
Bromuconazole	1	Yes	Yes								
Climbazole	1	Yes	Yes								
Cyproconazole	1	Yes	Yes								
Difenoconazole	1	Yes	Yes								
Diniconazole	1	Yes	Yes								
Fenbuconazole	1	Yes	Yes								
Flusilazole	1	Yes	Yes								
Hexaconazole	1	Yes	Yes								
Posaconazole	1	Yes - NS									
Propiconazole	1	Yes	Yes								
Triadimenol	1	Yes	Yes								
Triflumizole	1	Yes	Yes								
Triticonazole	1	Yes	Yes								
Imazalil	2										
Ipconazole	2	NS									
Ketoconazole	2	NS									
Myclobutanil	2	Yes	Yes								
Penconazole	2	Yes	Yes								
Voriconazole	2	Yes	Yes								
Paclobutrazol		NS									
Tetraconazole		NS									



# Pyrethroids

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- **Hepatotoxicity** and **nephrotoxicity** appear most prominent
- Endocrine disrupting capacity suspected
- Androgen and oestrogen receptor activation noted through Tox21

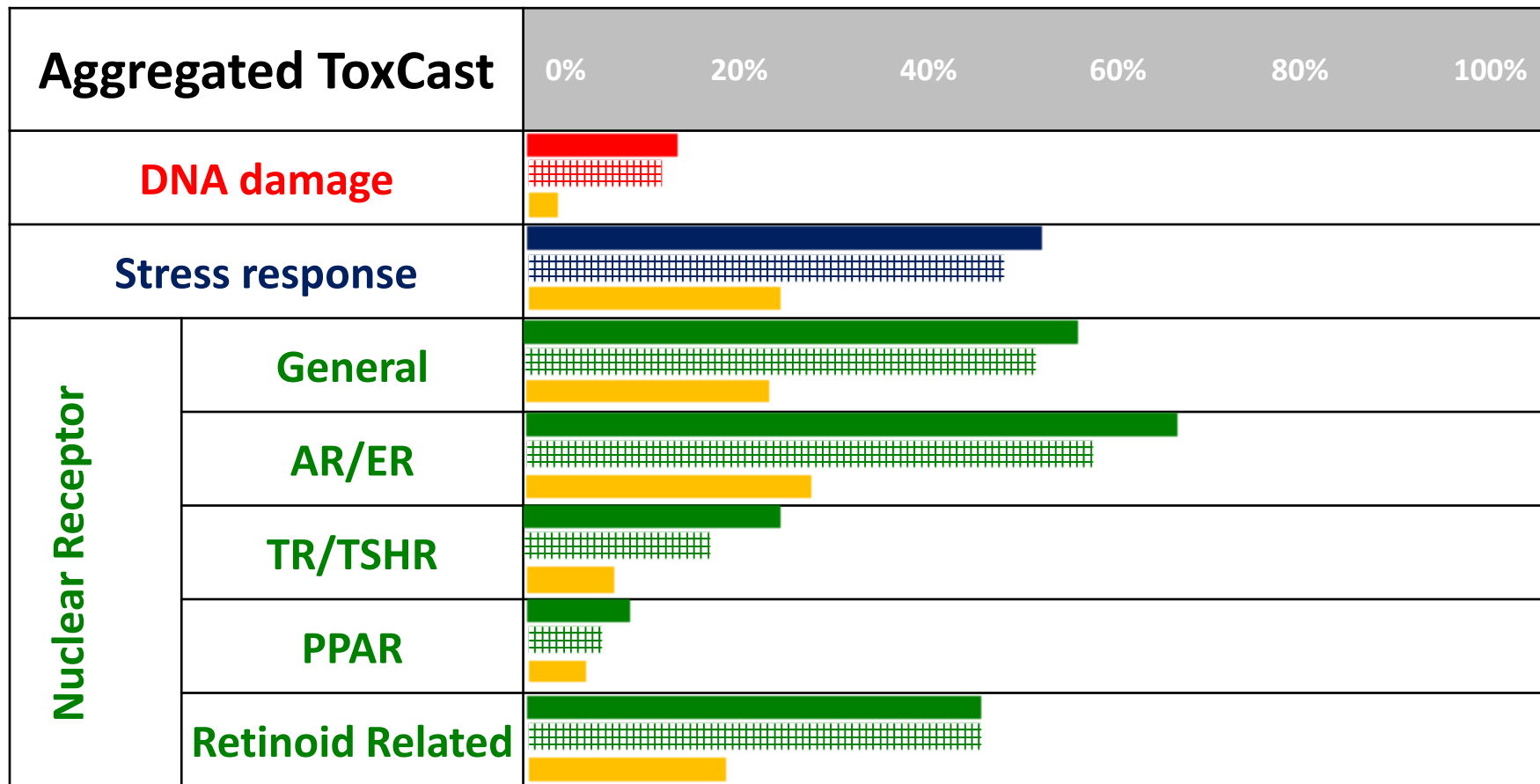
# Pyrethroids

	STOT RE	Organ/ systemic	Liver Effect	Kidney Effect	Cytotox	DNA damage	Stress response	Nuclear Receptor				
								General	PPAR	Retinold Related	TR/TSHR	AR/ER
Cyclopropane...	1	Yes		Yes								
Cyfluthrin	1	Yes	Yes									
Cyhalofop-butyl	1	Yes	Yes		Red	Green	Green		Green	Green	Red	Red
Cyhalothrin	1	NS				Green	Red	Red		Red		Red
Deltamethrin	1	NS				Green		Red				
Diclofop-methyl	1	Yes	Yes			Green						
Esfenvalerate	1	Yes	Yes			Green			Green			Green
Etofenprox	1	Yes	Yes	Yes		Green		Red		Green		Red
Fluazifop-butyl	1	Yes	Yes	Yes	Red	Green	Green	Red		Green		Red
Fluazifop-P-butyl	1	Yes		Yes	Red	Green	Red	Red		Green		Red
Fluvalinate	1	Yes				Green	Red	Red	Green		Green	Red
Lactofen	1	Yes	Yes	Yes	Red	Green	Red	Red	Red	Red	Red	Red
3-phenoxybenzyl...	2	Yes-NS				Green	Green	Red	Red	Green		Green
a-Cypermethrin	2	Yes-NS										
Fenoxycarb	2	NS			Red	Green	Red	Red	Red	Red		Red
Fenpropathrin	2	Yes		Yes	Red	Green		Red	Green	Red		Red
Flucythrinate	2											
Phenothrin	2	Yes	Yes			Green	Green	Red	Green	Green	Green	
Pyriproxyfen	2	Yes	Yes	Yes	Red	Green	Red	Red		Red		Red
I-Cyhalothrin		NS				Green	Red	Red	Green	Red	Red	Red





# Non-Classified Compounds Have Fewer Hits





# Unspecific Toxicity

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- Prediction of unspecific effects and related regulatory classification remains elusive
  - Not well defined or identified
- Does no hits in ToxCast indicate non-specific toxicity?
  - Are all modes of action covered?
  - Does unspecific toxicity = Non Classified?

# Conclusions

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## Can We Predict Regulatory Classifications?

- Well curated STOT-RE / NAM data sets
- ToxCast data make sense, but not wholly predictive
  - Hits RE1 > RE2
- Low / no toxicity associated with low number of hits

## Can We Gain Insight on Regulatory Classifications?

- Organ level effects indicative of STOT-RE classifications
- NAMs can be related to AOPs / MoA
- Significant groupings possible, albeit limited
- Tox21 assays support grouping and possibly read-across (and vice versa)