

GENOTOXIC SUBSTANCES IN PRINTED PAPER AND BOARD FOOD CONTACT MATERIALS

A prioritisation strategy based on non-animal methods

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Dr. Birgit Mertens
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UNIVERSITEIT
BRUSSEL

EFSA, Parma, 10-11 July 2018





PAPER AND BOARD

+

PRINTING INKS



Printed paper and board

Widely & Frequently used

Major cause of contamination by FCM

Thousands of non (recently) safety-evaluated substances



PAPER AND BOARD

+

PRINTING INKS



Printed paper and board

Widely & Frequently used

Major cause of contamination by FCM

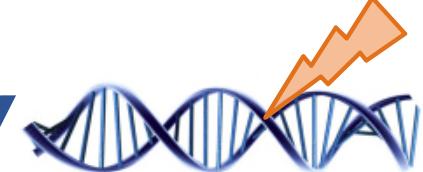
Thousands of non (recently) safety-evaluated substances



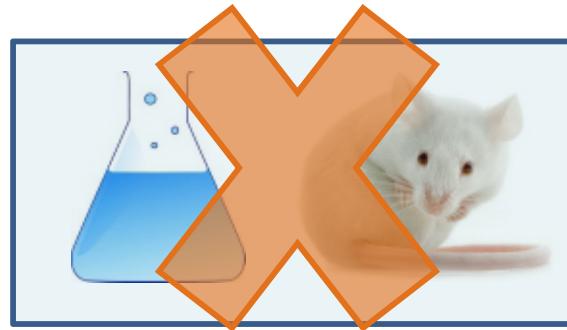
AIM:

PRIORITISATION STRATEGY

Genotoxicity



METHODOLOGY



**Step 1:
Database
compilation**



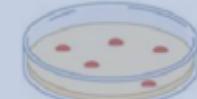
**Step 2:
In silico
prediction**



**Step 3:
Literature
review**



**Step 4:
In vitro
testing**



NON-ANIMAL METHODS

Step 1



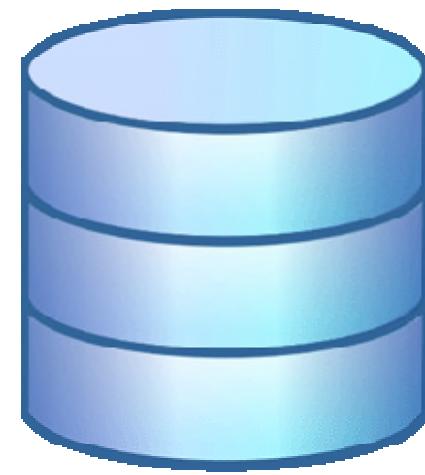
Step 2



Step 3



Step 4



STEP 1: DATABASE COMPIRATION

Van Bossuyt M, Van Hoeck E, Vanhaecke T, Rogiers V & Mertens B* (2016)*

Printed paper and board food contact materials as a potential source of food contamination.

*Regulatory Toxicology and Pharmacology 81: 10-19. *Equal contribution*

Step 1



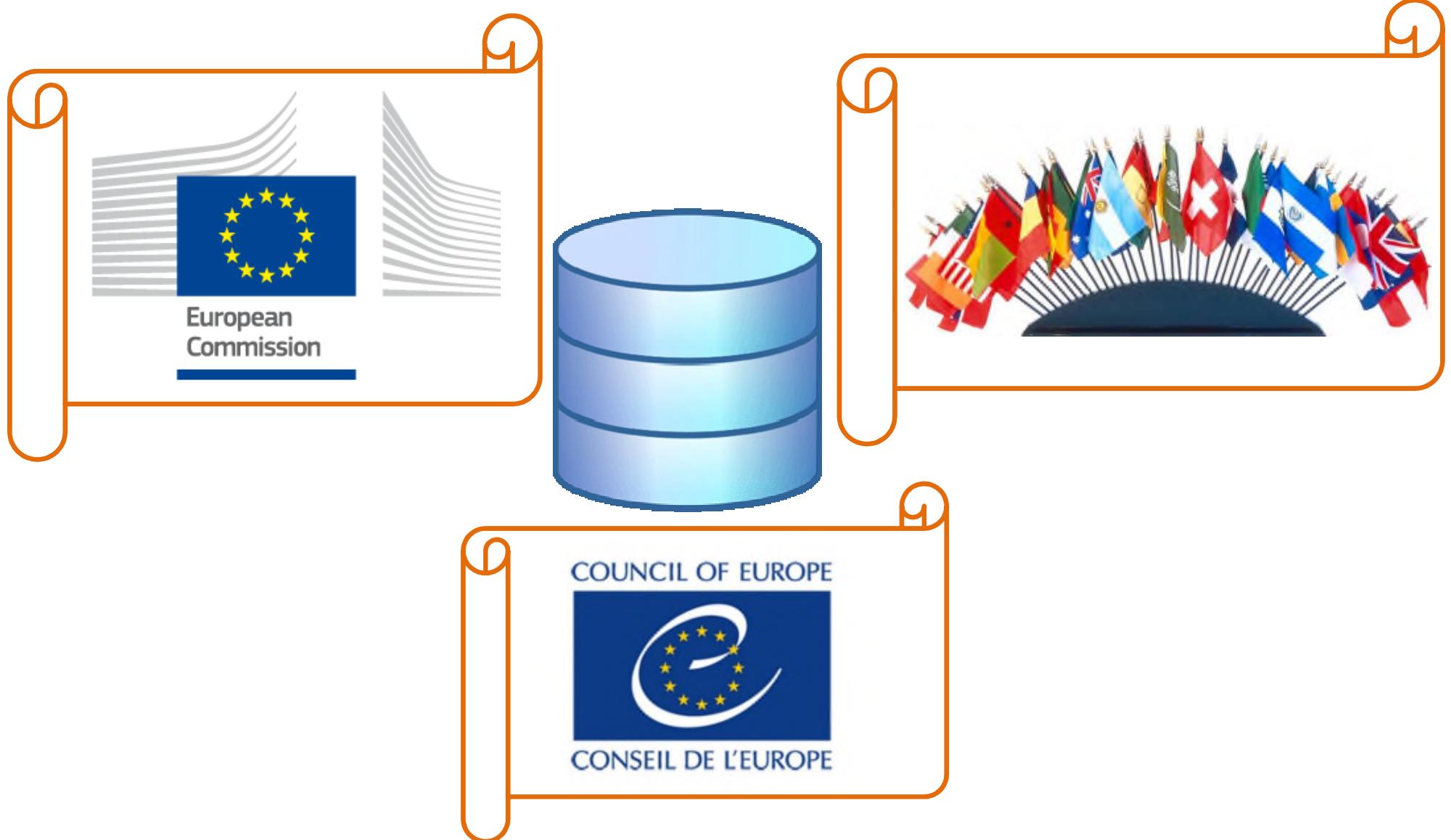
Step 2



Step 3



Step 4



Step 1



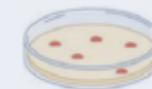
Step 2



Step 3



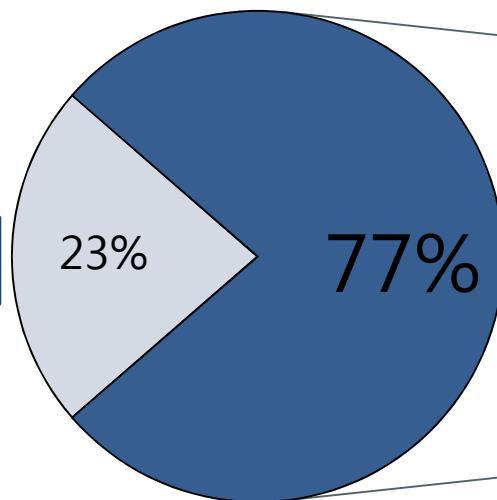
Step 4



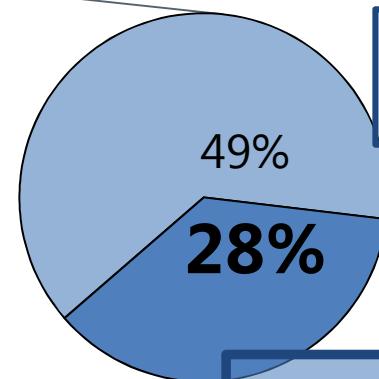
$$\Sigma = 6073$$



Evaluated



Non-evaluated



Other (polymers,
mixtures, metals, etc.)

Single
substances

#1723

Step 1



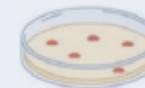
Step 2



Step 3



Step 4



STEP 2: *IN SILICO* PREDICTION

1. Van Bossuyt M, Van Hoeck E, Raitano G, Manganelli S, Braeken E, Ates G, Vanhaecke T, Van Miert S, Benfenati E, Mertens B* & Rogiers V* (2017)
(Q)SAR tools for priority setting: a case study with printed paper and board food contact material substances.
Food and Chemical Toxicology 102: 109-119.
2. Van Bossuyt M, Van Hoeck E, Raitano G, Vanhaecke T, Benfenati E, Mertens B* & Rogiers V* (2018)
Performance of in silico models for mutagenicity prediction of food contact materials. Toxicological Sciences 163(2): 632-638

*Equal contribution

Step 1



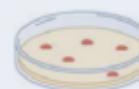
Step 2



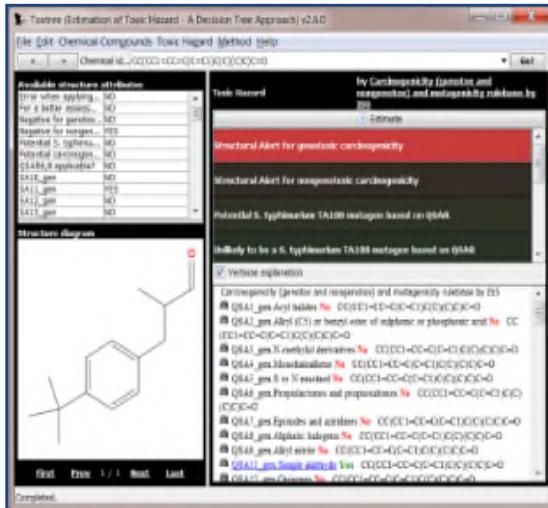
Step 3



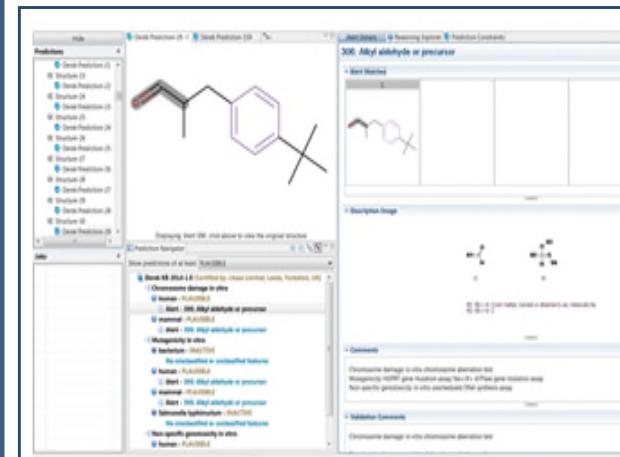
Step 4



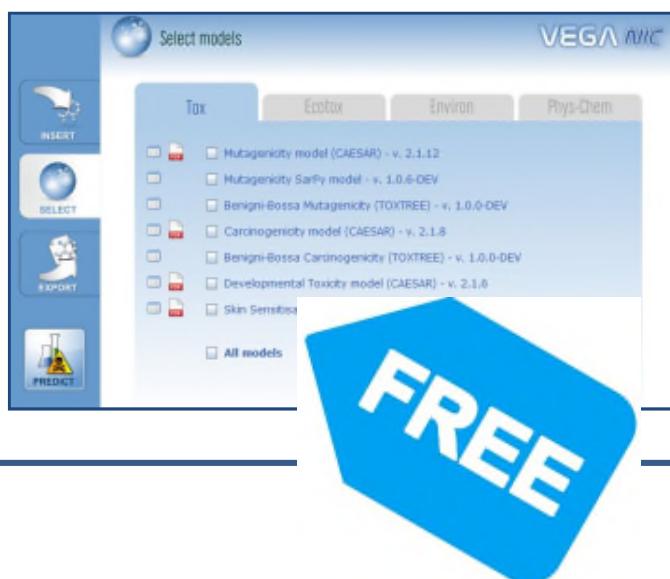
Toxtree



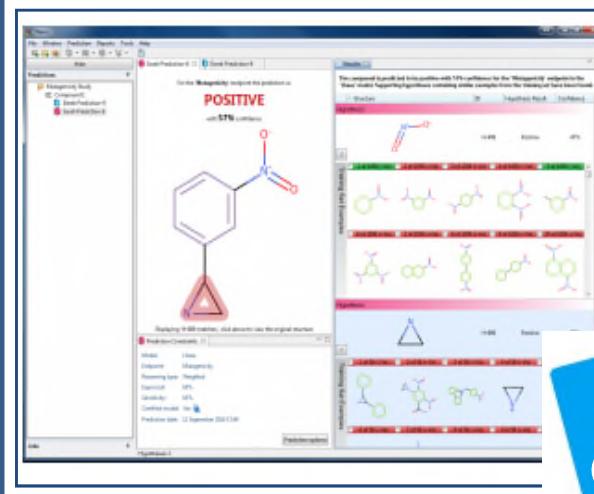
Derek Nexus



VEGA



Sarah Nexus



€£\$

Step 1



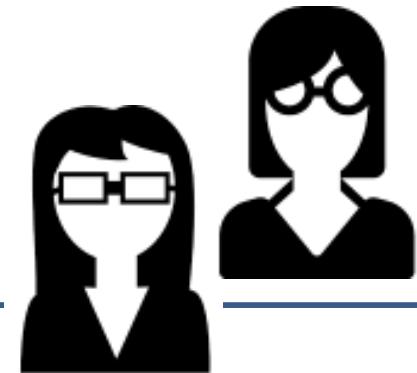
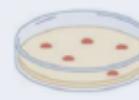
Step 2



Step 3



Step 4



Toxtree

The screenshot shows the Toxtree software interface. On the left, there's a sidebar with 'Available structures attributes' and a 'Structures diagram' showing a chemical structure of 4-(4-methylphenyl)butan-2-one. The main panel displays 'Toxic Hazard' results, including 'By Carcinogenicity (genotoxic and non-genotoxic) and mutagenicity reference by QSR' and 'Structural Alert for genotoxic carcinogenicity' and 'Structural Alert for non-genotoxic carcinogenicity'. At the bottom, there's a list of QSRs (Quantitative Structure-Activity Relationships) such as QSR1_gen-toxicity-teratogenicity, QSR2_gen-Mutagenicity_in vitro, etc.

QUALITATIVE

The screenshot shows the Derek Nexus software interface. It features a 'Prediction' list on the left with items like 'Derek Predictor 1', 'Derek Predictor 2', etc. The main area shows a chemical structure of 4-(4-methylphenyl)butan-2-one and a 'Description Design' section with various toxicity predictions and comments, such as 'Dermatitis - 4-(4-methylphenyl)butan-2-one' and 'Chromosome damage in vitro chromosome aberration test'.

Derek
Nexus

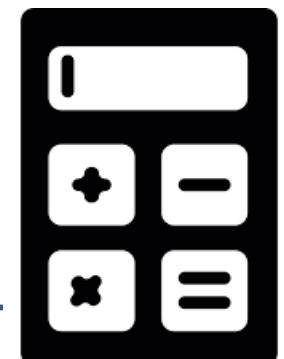
VEGA

The screenshot shows the VEGA software interface. On the left, there are buttons for 'INSERT', 'SELECT', 'EXPORT', and 'PREDICT'. The main area is titled 'Select models' and shows tabs for 'Tox', 'Ecotoxic', 'Environ', and 'Phys.Chem'. Under the 'Tox' tab, several models are listed with checkboxes: Mutagenicity model, Mutagenicity SarPy, Benigni-Bossa Mutagenicity, Carcinogenicity model, Benigni-Bossa Carcinogenicity (TOXFREE) - v. 1.0.0-DEV, Developmental Toxicity model (CAESAR) - v. 2.1.0, Skin Sensitisation model (CAESAR) - v. 2.1.5, and All models.

QUANTITATIVE

The screenshot shows the Sarah Nexus software interface. It displays a 'Prediction' result for 'Derek Predictor 1' showing 'POSITIVE' with 'STM criteria'. Below it, there's a 'Results' section with a 'Reaction' table and a 'Chemical Reactions' section showing various reaction schemes and structures.

Sarah
Nexus



Step 1



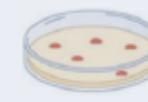
Step 2



Step 3



Step 4



Combination of gene mutation prediction results

POSITIVE IN ALL 4

#106

POSITIVE IN 3

#128

POSITIVE IN 2

#94

POSITIVE IN 1

#204

NO ALERT IN ANY TOOL

#1191

Step 1



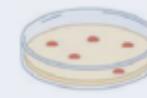
Step 2



Step 3



Step 4



POSITIVE IN ALL 4

#106

**HIGHEST
PRIORITY**

POSITIVE IN 3

POSITIVE IN 2

POSITIVE IN 1

NO ALERT IN ANY TOOL



**LOWEST
PRIORITY**

Step 1



Step 2



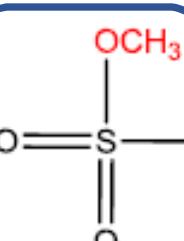
Step 3



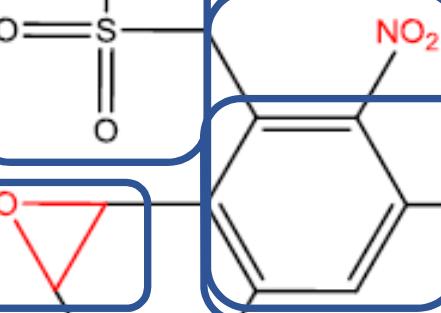
Step 4



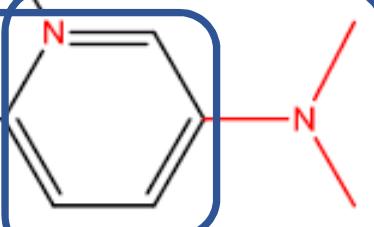
1) Sulphonic acid alkyl ester



2) Aromatic nitro group

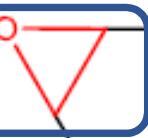


3) Aromatic azo group

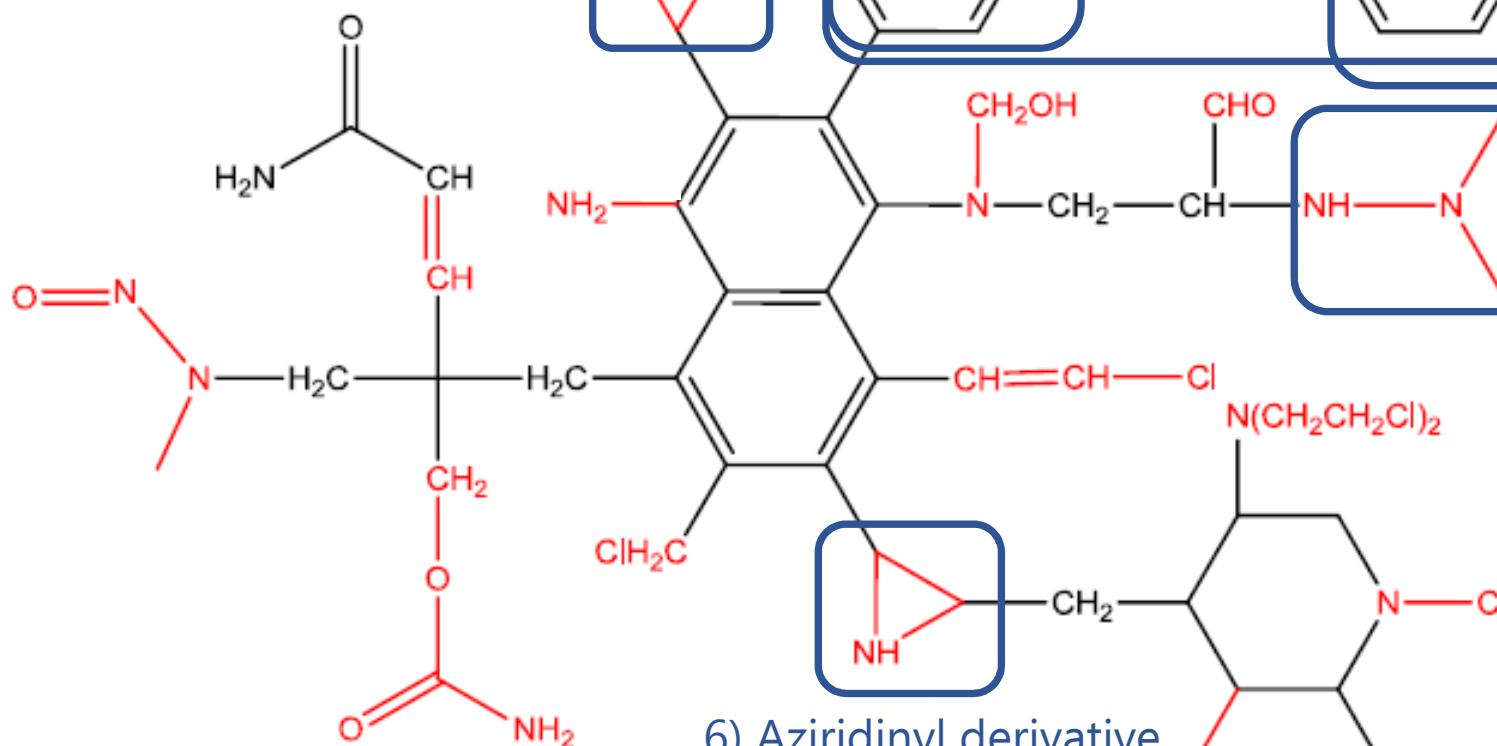


4) Aromatic alkyl amino group

7) Epoxide

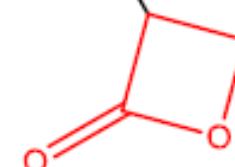


5) Alkyl hydrazine



6) Aziridinyl derivative

Ashby-Tennant
polycarcinogen



Step 1



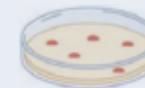
Step 2



Step 3



Step 4



STEP 3: LITERATURE REVIEW

*Van Bossuyt M, Van Hoeck E, Vanhaecke T, Rogiers V * & Mertens B**

***Prioritising substances of emerging concern for in-depth safety evaluation based on their genotoxic potential:
the example of printed paper and board food contact materials.***

Submitted to Toxicology Letters.

Step 1



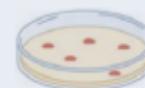
Step 2



Step 3



Step 4



PRIORITY SUBSTANCES (#106)



Official evaluation available

Genotoxic *in vivo*

Inconclusive

Not genotoxic *in vivo*

NO official evaluation available

Data collection from existing databases



Step 1



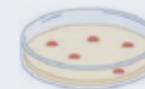
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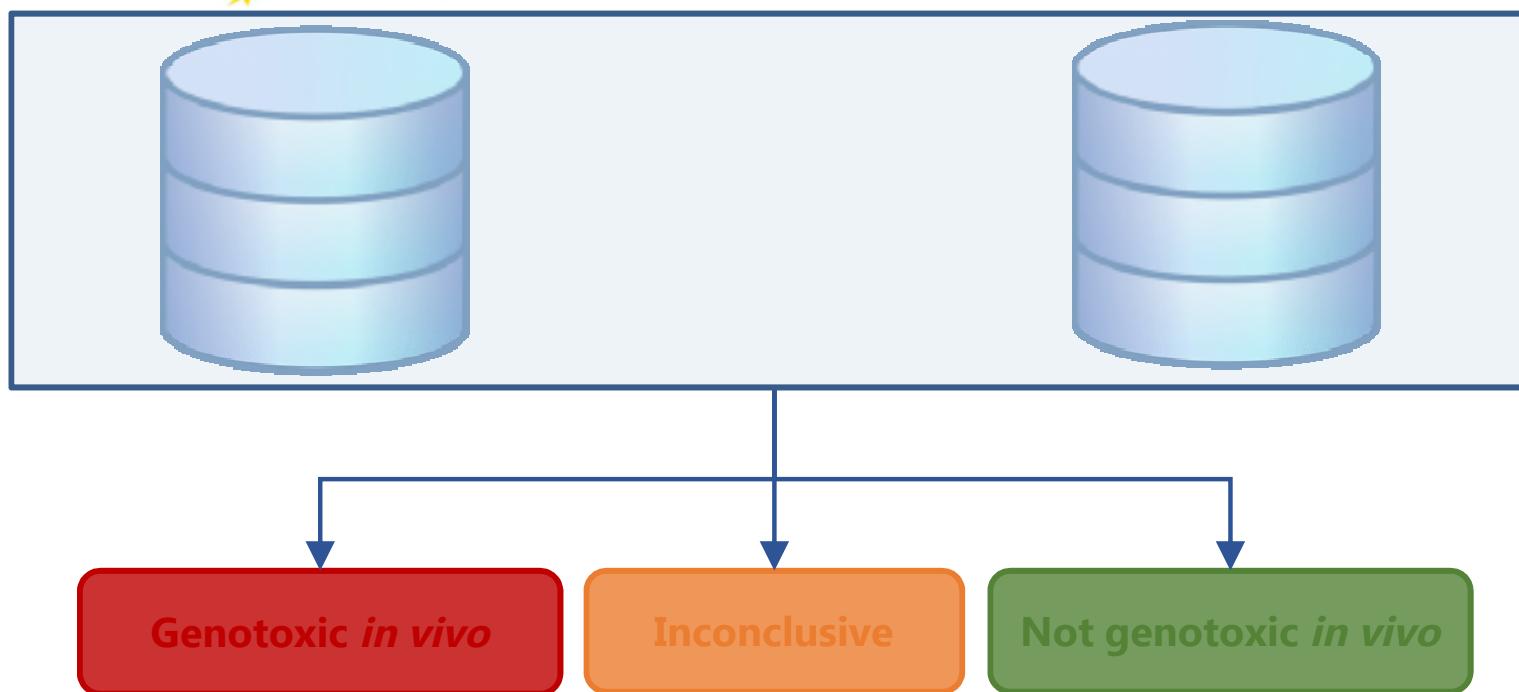
Step 3



Step 4



European Union Reference Laboratory
for Alternatives to Animal Testing



Step 1



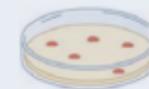
Step 2



Step 3



Step 4



**Gene mutation data
are lacking!**

Step 1



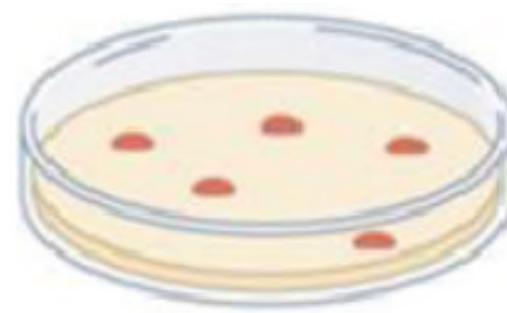
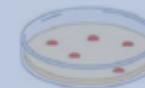
Step 2



Step 3



Step 4



STEP 4: IN VITRO TESTING

*Van Bossuyt M, Van Hoeck E, Vanhaecke T, Rogiers V * & Mertens B**

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Submitted to Toxicology Letters.

Step 1



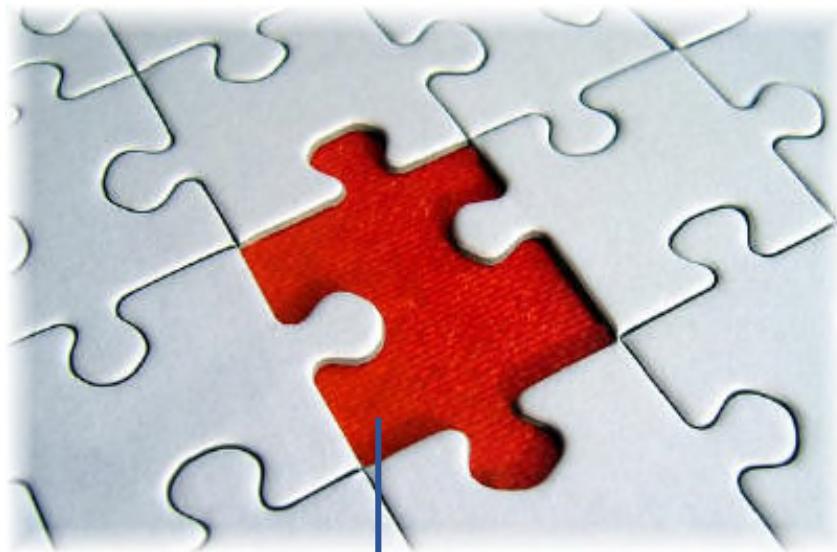
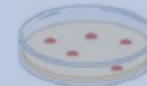
Step 2



Step 3



Step 4



**Gene mutation data
are lacking!**



Step 1



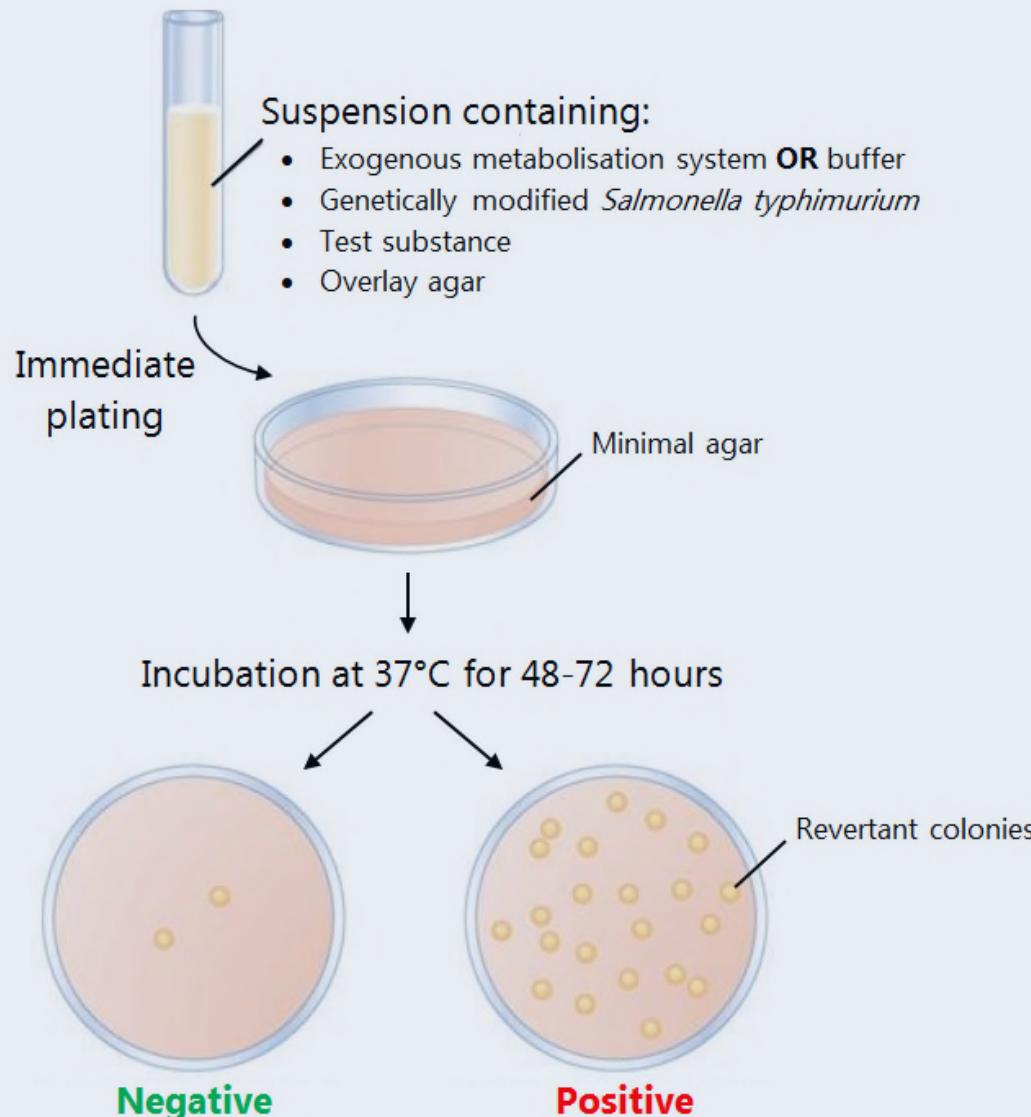
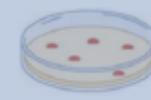
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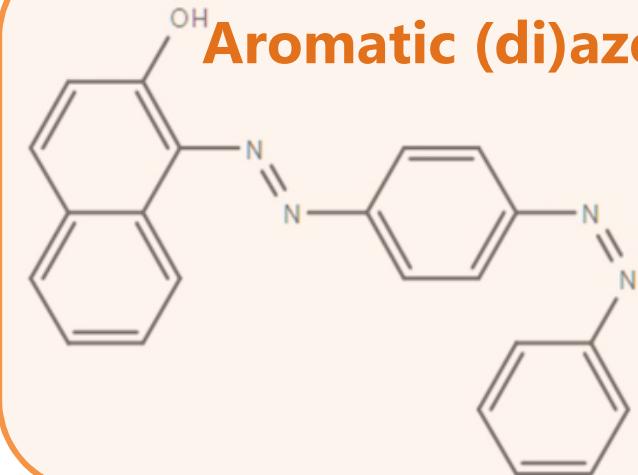
Step 3



Step 4



Aromatic (di)azo



1

Metabolisation system

2

Additional cofactors

3

Pre-incubation 30'

Step 1



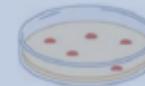
Step 2



Step 3



Step 4



Negative in NON-OFFICALLY VERIFIED gene mutation test



CONCLUSION

Step 1:
Database
compilation

#123



Step 2:
In silico
prediction

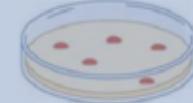
#106



Step 3:
Literature
review



Step 4:
In vitro
testing



Future perspectives

- For a full safety evaluation, **additional aspects** need to be investigated



FCM-related

- Actual use
- Type of food
- Conditions of use
- ...



TOX-related

- Other genotoxic endpoint
- Other toxicological endpoints
- ...

- This prioritisation strategy can be extended to **other substance types/groups**



THANK YOU FOR YOUR ATTENTION!

