How Well QSARs Predict Aquatic Toxicity of REACH Registered Substances?

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Introduction

- All standard information requirements can be adapted by a reliable and relevant QSAR adaptation (Annex XI, Section 1.3)
- But do QSARs always produce same information on all the effects measured in a e.g. OECD TG 210 study (hatching and survival, abnormal appearance, abnormal behaviour, weight, length)?
- Purpose of this study was to assess whether the use of QSARs would estimate hazards differently to standard experimental studies and whether any differences would affect regulatory decision-making.

Information on aquatic toxicity required under REACH

- 1-10 ton/a: Annex VII, Section 9.1
  - Short-term toxicity testing on aquatic invertebrates, and
  - Growth inhibition study on aquatic plants
- 10-100 ton/a: Annex VIII, Section 9.1
  - Short-term toxicity testing on fish,
- 100-1000 ton/a: Annex IX, Section 9.1.
  - Long-term toxicity testing on aquatic invertebrates (OECD 211)
  - **Long-term toxicity testing on fish (OECD 210)**

Materials and methods

1. We collected newly conducted reliable long-term fish studies (OECD TG 210) resulting from ECHA evaluation decisions
   - 89 experimental studies received (+23 not yet evaluated), out of which 49 were for monoconstituents;
2. We predicted chronic fish toxicity (NOEC) by QSAR models **ECOSAR v2.2, VEGA v.1.2.0 (IRFMN v1.0.1)** and **iSafeRat® Desktop version 4.2** for the same (monoconstituent) substances;
3. Experimental NOEC values (most sensitive) were compared to the predictions;
4. We assessed whether any deviation between experimental data and prediction would affect regulatory risk management by exceeding the specific thresholds
   - Chronic 1 or 2 classification (assuming non-rapid degradation),
   - T criterion under PBT assessment.

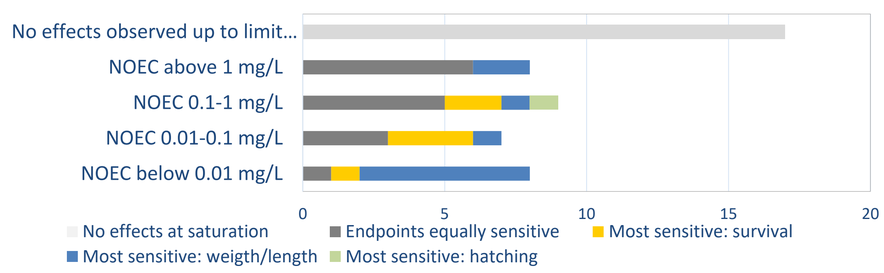
How reliability of the QSAR model was considered?

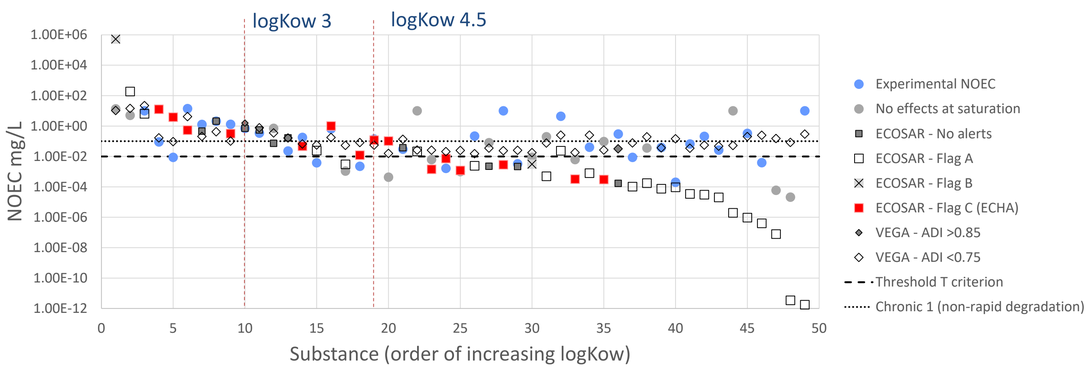
ECOSAR Flag A (model alerts the user)	ECOSAR Flag B (user guide indicates a warning)	ECOSAR Flag C (ECHA additional warning)
SAT SaturateSolublity (Effect level exceeds WS by factor 10) ACR AcuteToChronicRatios (empirically derived class-specific ratio) KOW1 LogKowCutOff (endpoint-specific) MW DomainOfApplicability (MW > 1000)	MET inorganics, inorganic salts and metals including organometals HYD hydrolytically unstable or highly reactive chemicals SALT (complex) salts - SMILES is changed to neutral species automatically Kow or MW or FRAG (fragment) or FLU (perfluorinated substance) or CNC (imidazole ring, quaternary nitrogen, nitrogen heterocycles other than pyridine) out of domain ION ionized at pH 4-9	R2 of ECOSAR class is < 0.6 N (number) of substances used in the training set of the class is < 5 ION ionizable substances; > 90 % pH range 4 - 9 (percepta output) KOW input fragment not present in KOWWIN training set

VEGA:
ADI >8.85: high reliability
ADI < 0.75: low reliability

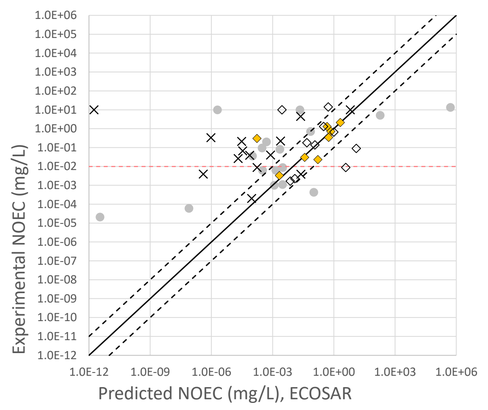
Results

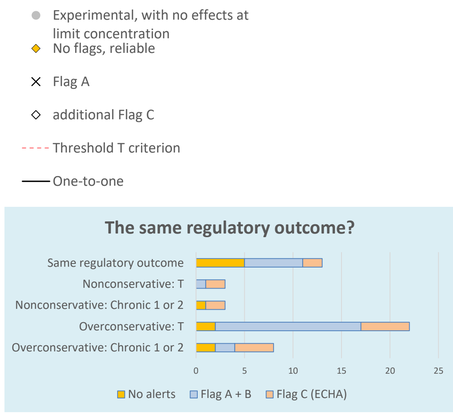
What effect endpoint drives the NOEC (substances sorted according to the lowest NOEC)?



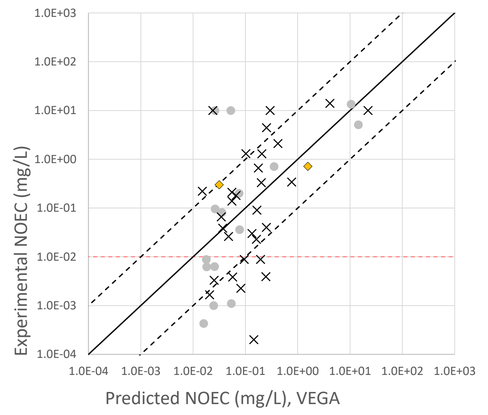


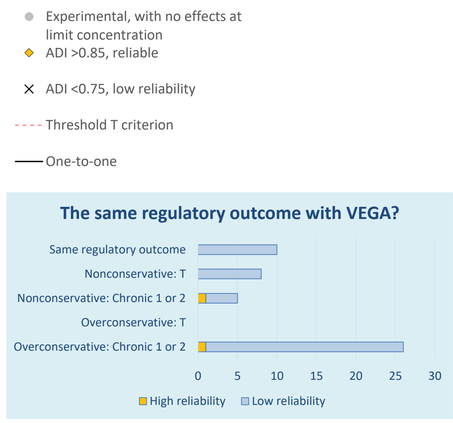
ECOSAR



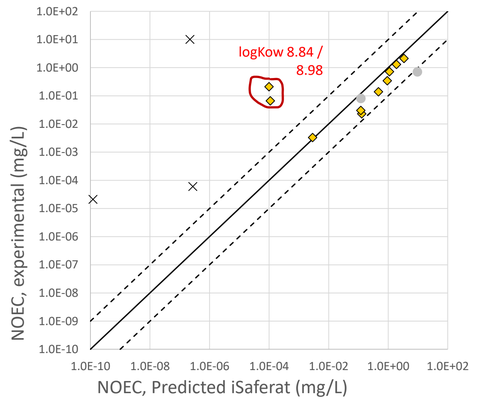


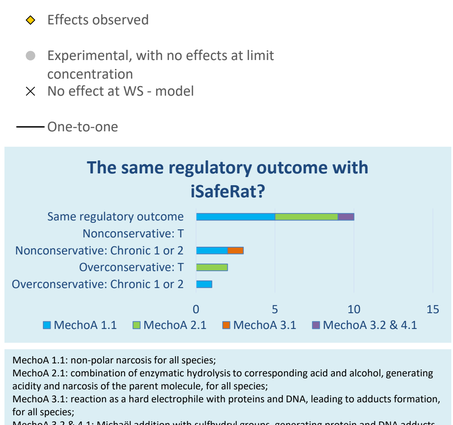
VEGA





iSafeRat





MechoA 1.1: non-polar narcosis for all species;
MechoA 2.1: combination of enzymatic hydrolysis to corresponding acid and alcohol, generating acidity and narcosis of the parent molecule, for all species;
MechoA 3.1: reaction as a hard electrophile with proteins and DNA, leading to adducts formation, for all species;
MechoA 3.2 & 4.1: Michaël addition with sulfhydryl groups, generating protein and DNA adducts for all species & rapid metabolism to pyruvate, which is incorporated into Krebs cycle.

Conclusions

1) Further work is needed in model development for them to be used in regulatory hazard assessment instead of experimental long-term fish studies such as OECD TG 210:

- Improvements needed to widen the applicability domain. The models we tested could produce predictions for:
  - **ECOSAR:** 18/49 substances (no warning flags + ECHA warnings); **ECOSAR:** 8/49 (no warning flags);
  - **VEGA**: 2/49 substances (high reliability);
  - **iSafeRat**: 16/49 substances.
- Different regulatory outcome with (Q)SAR
  - **ECOSAR:** No flags but still 50% mismatch for regulatory outcome ('Esters' , 'Amides')
  - **ECOSAR:** Flag C (ECHA) predictions had 82% mismatch. Flags based on N, R2, need to be considered (e.g. 'Imides', 'Carbonyl Ureas', 'Aliphatic Amines', 'Benzodioxoles')
  - **iSafeRat** provides a better match: 10/16 the same regulatory outcome and 0/16 nonconservative T.

2) The OECD TG 210 study may not capture the long-term effects properly for superhydrophobic substances (there are issues with maintaining the substance in the solutions etc)
- ECOSAR and iSafeRat for such substances are often overconservative but are more correct?

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