

Latest developments in PBT testing approaches for the Regulatory Assessment of Offshore Chemicals

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OVERVIEW

- **Background**
- **Persistence (P)**
- **Bioaccumulation (B)**
- **Toxicity (T)**
- **Conclusions**

BACKGROUND

- Cefas registers offshore chemicals for UK and Netherlands
- OSPAR Harmonised Mandatory Control System (HMCS) used since 2000
- Test protocols from 1990s
- Environmental risk assessment science is evolving
- Identify which developments could affect the regulatory framework in the future

ASIDE: OSPAR and REACH

- Offshore sector subject to both regulatory frameworks
- Discussion is from OSPAR viewpoint but will refer to REACH where appropriate
- OSPAR has stated goal of harmonisation where possible
- OSPAR will address this issue after 2018 REACH registration deadline

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BIODEGRADABILITY



Problems with Ready Tests

“...these standard tests, particularly the ready biodegradation tests, have a high potential to produce conflicting results and false negatives. Therefore new tests are required that can accurately assess rates of degradation”

ECETOC, 2009: Technical Report No.108

- Designed to identify ready biodegradability
- Used to highlight persistent substances
- Tests not ideal for this purpose
- Causes of inconsistencies researched

Cefic ECO11 project

- Part of Cefic Long Range Initiative (LRI)

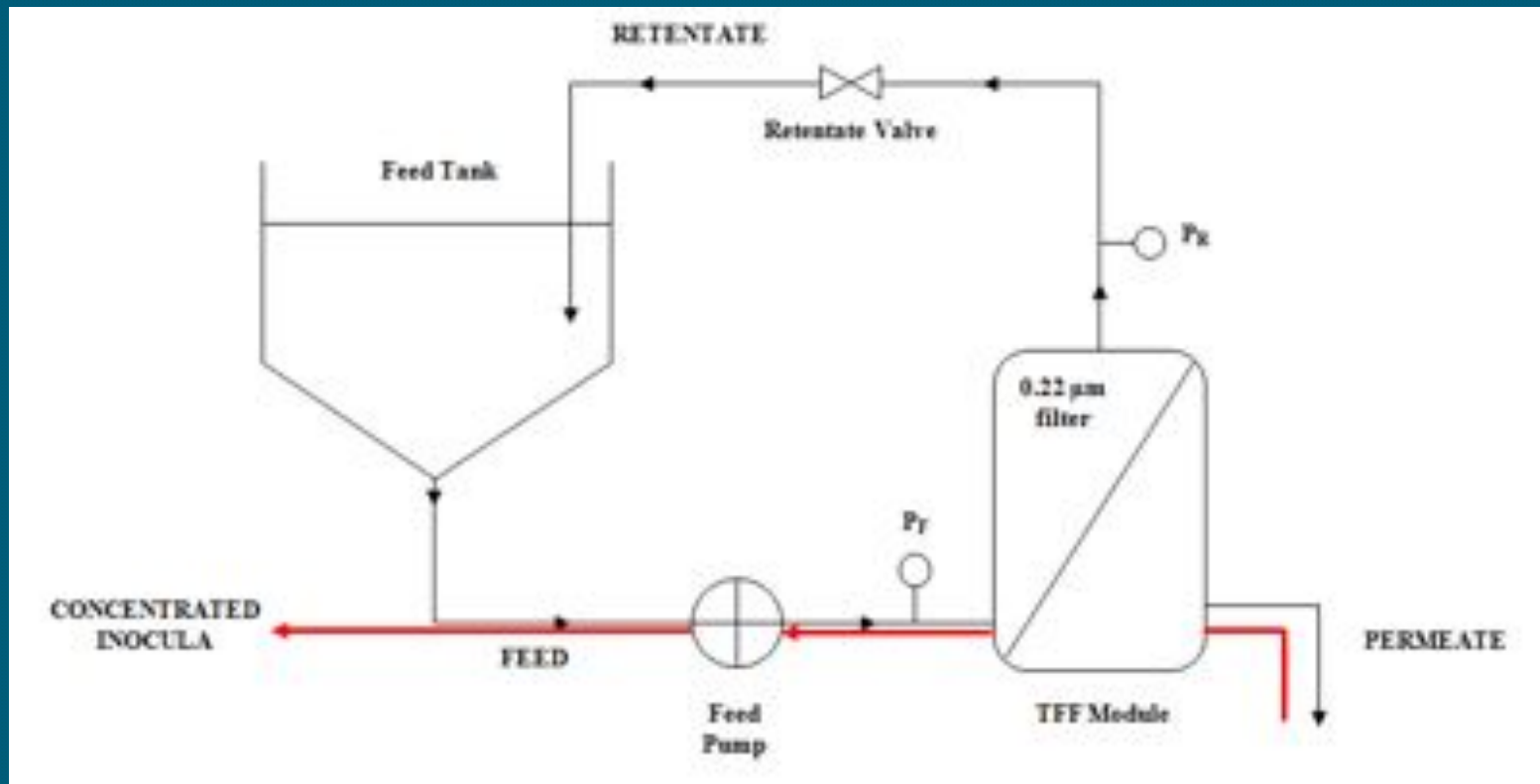
Objectives included:

- Determine the influence of inocula source, concentration and diversity on biodegradability test outcome
- Assess the relative bias of different methods for increasing cell concentration

ECO 11 outcomes

- Inocula source, concentration and diversity **ALL** affect biodegradability test outcome
- Low inocula concentrations result in variable inclusion of competent degraders
- Use of **larger test volumes** promotes inclusion of competent degraders
- Concentration of large volumes of sample possible by **TANGENTIAL FLOW FILTRATION**

Tangential Flow Filtration schematic



- **Simple equipment: set-up costs <£5k possible**

THE IMPROVED OECD 306

- Method enhancements studied under Eco 11 programme proposed for OECD 306
- Tangential Flow Fractionation used to concentrate sea water
- Simulates effect of larger sample size:
 - Increased cell diversity
 - Greater probability of competent degraders
- Cefas approached in 2013 to consider acceptability under OSPAR
- OSPAR recommended workshop to discuss

Biodegradability Workshop

17-18th February 2015

- Organised by Cefas in association with ECETOC
- Industry, Regulatory bodies and Test Labs representing 7 countries
- Theory presentations
- Discussion sessions
- Practical demonstration of TFF methodology



Workshop Outcomes

- Support for enhanced methodology established
- Ring Test encouraged
- Participation of 12 laboratories possible

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But....

- Test Protocol details to be finalised
- Test substances to be agreed
- Funding the project remains under discussion

Next Steps

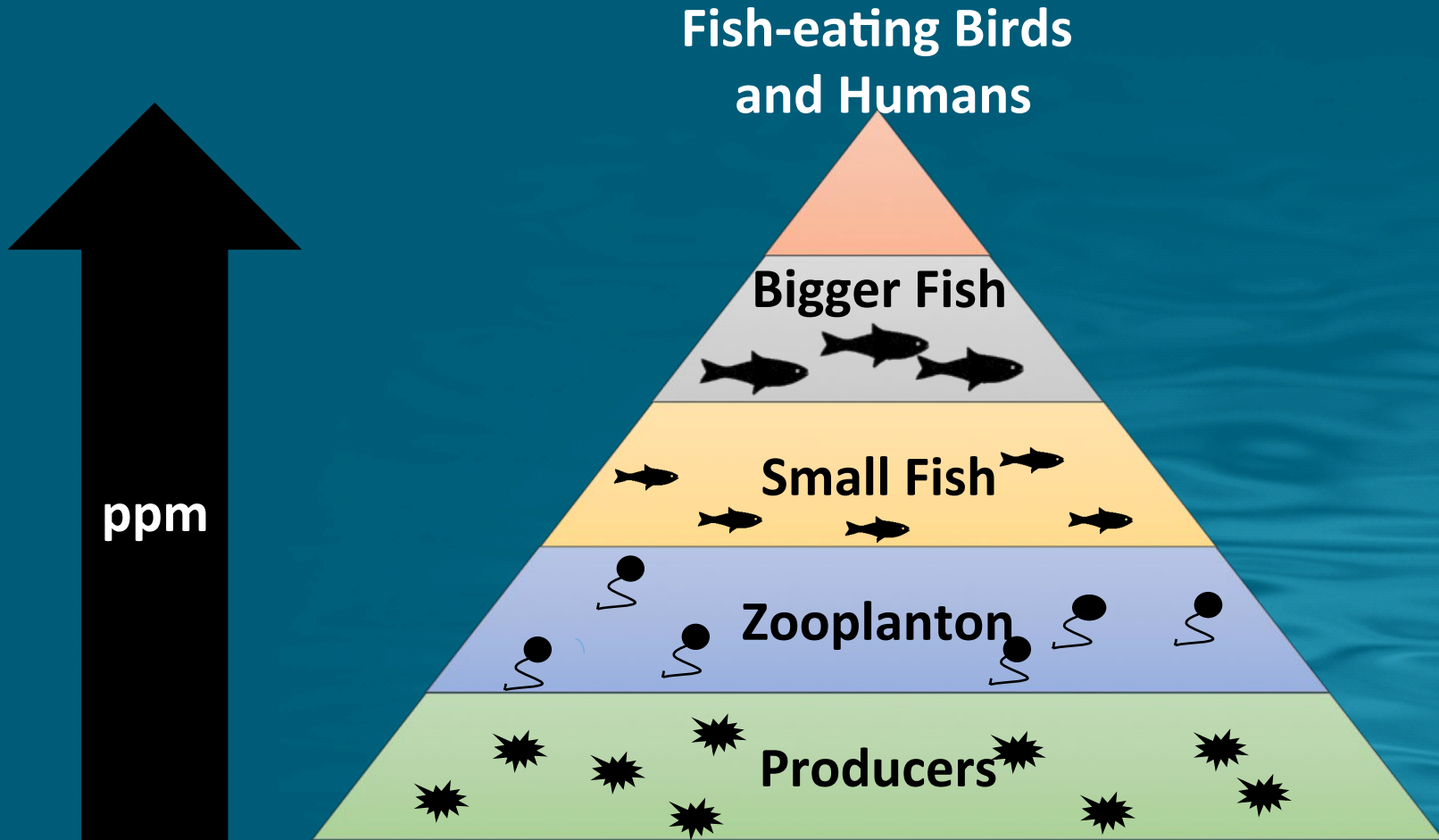
- Ring Test expected to be 13 month project

When complete:

- Results to be reported to OSPAR (OIC)
- The revised 306 to be submitted for OECD approval
- Caution required! Revised method is not a Ready Test
- Need to retain unmodified method as option

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BIOACCUMULATION



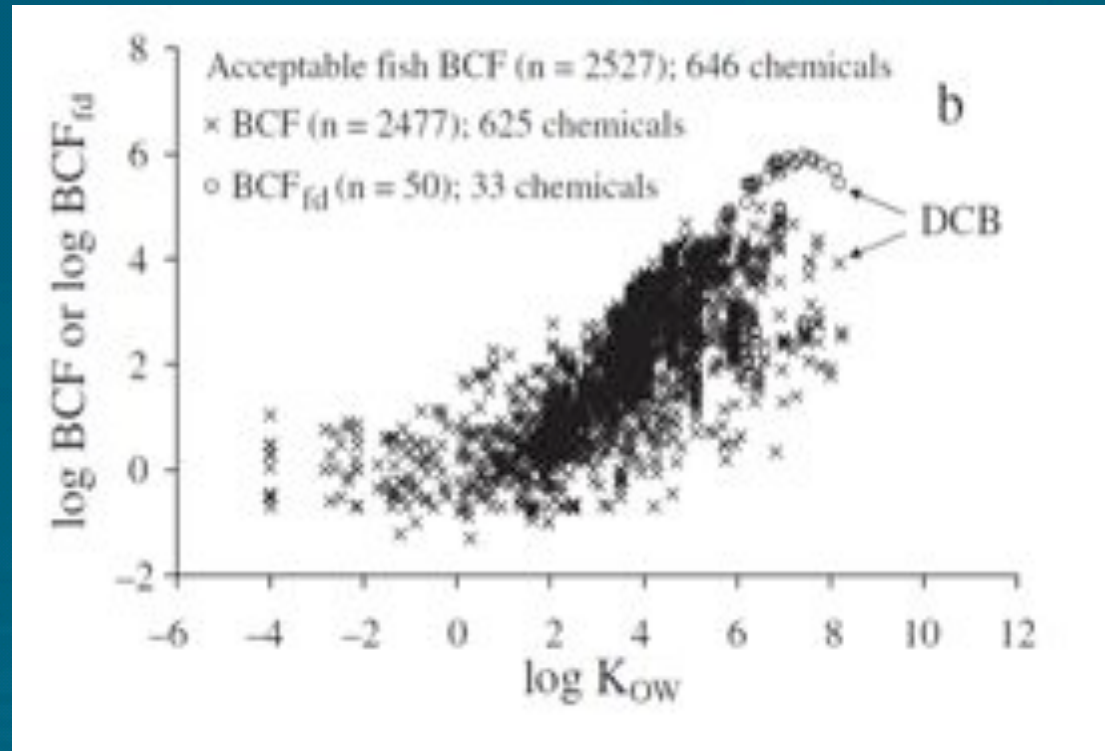
Parameters Currently Used

- **BIOCONCENTRATION FACTOR (BCF)**
- Costly to determine
- Ethically objectionable (fish)

- **OCTANOL / WATER PARTITION COEFFICIENT ($\text{Log}P_{ow}$ or $\text{Log}K_{ow}$)**
- Easy to determine
- Has other applications
- Accuracy = ?

LogP_{ow} vs LogBCF

For certain types of substances, BCF is significantly less than LogP_{ow} predicts



J.A. Arnot and F.A.P.C. Gobas, Environ. Rev. 14: 257-297 (2006)

Problems with LogP_{ow}

- LogP_{ow} assesses bioaccumulation potential
- Assumes simple partitioning process
- Not applicable where partitioning does not occur (surfactants)
- Applicability breaks down where other processes become significant
- BCF then preferable....

Problems with BCF

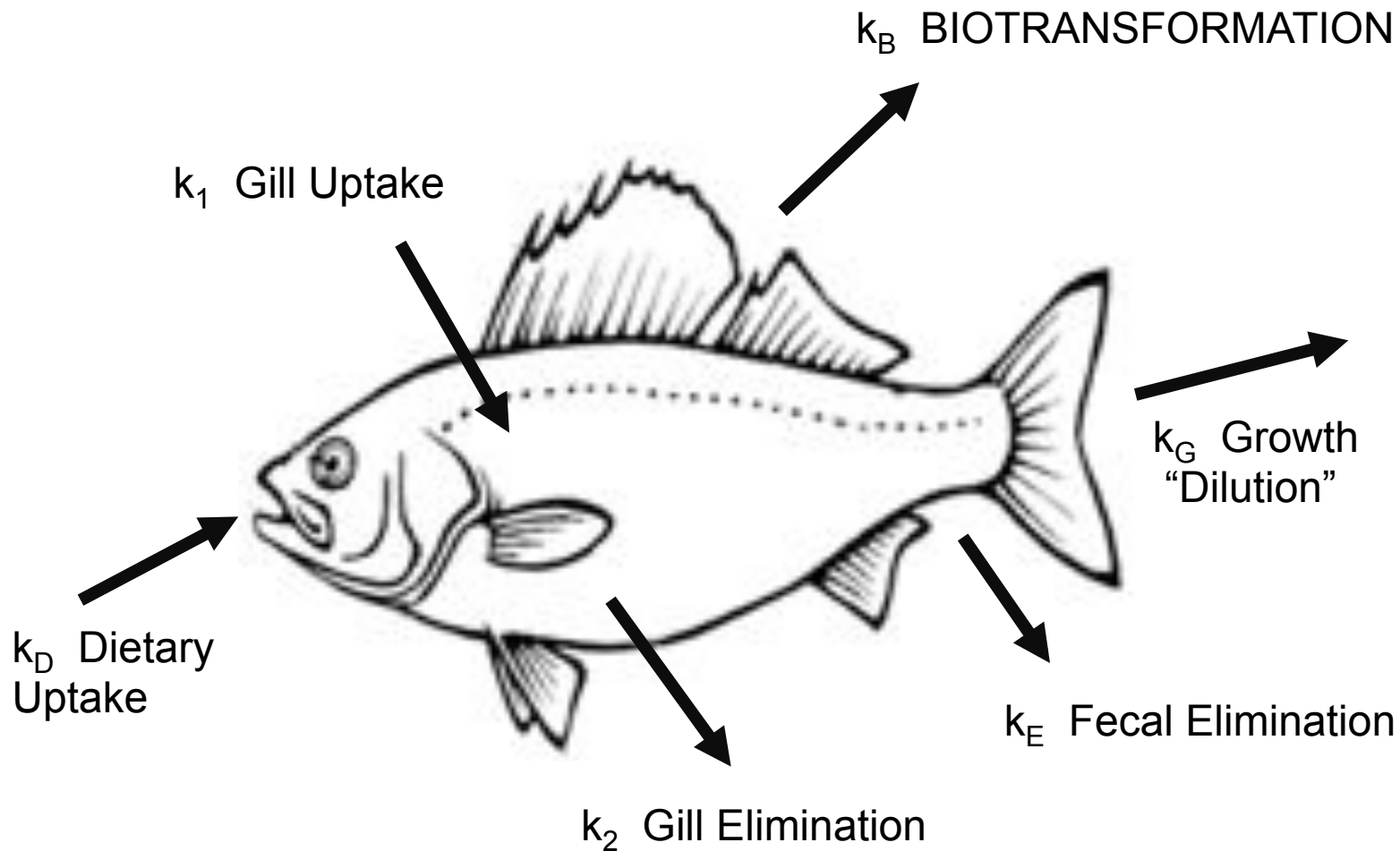
- BCF does not always provide most reliable indication of bioaccumulation
- Neglects dietary uptake.....
 -this can be significant for very hydrophobic substances
- Neither LogP_{ow} nor BCF are ideal parameters to assess bioaccumulation

New approaches

- 2014 Helsinki Bioaccumulation Workshop discussed alternative metrics
 - **Bioaccumulation factor, BAF** (steady state ratio of chemical concentration in an organism and its ambient environment including dietary exposure),
 - **Biomagnification factor, BMF** (steady state ratio of chemical concentration between an organism and its food),
 - **Trophic magnification factor, TMF** (the average biomagnification factor in a food web that includes multiple trophic levels)
 - **The elimination half-life ($t_{1/2}$)**

k_B biotransformation rate constant

- A common element to all of these new metrics is k_B , the biotransformation rate constant
 - Reflects rate of metabolism
 - Can be determined from laboratory measurements or QSAR model predictions
- High k_B will offset high $\text{Log}P_{ow}$
- Proposed as new prescreening criterion, alongside $\text{Log}P_{ow}$



Arnot, 2015: Bioaccumulation Assessment: Developing Frameworks and Finding Common Ground. Presentation at SETAC Meeting, Barcelona, 5th May 2015

Future Direction

- Bioaccumulation science continues to evolve
- It has now outgrown the simple partitioning approach and parameters used to measure it.
- Helsinki workshop concluded greater use of Weight of Evidence (WoE) approaches to address bioaccumulation assessment can be anticipated in the future
- Acceptability of WoE already established

Weight of Evidence in Bioaccumulation Assessment

- REACH Guidance available
- Allowed in HMCS since 2008
- But few submissions made, and fewer satisfactory
- Problems are a lack of:
 - References to publicly-available literature documenting bioaccumulation characteristics of the relevant substance
 - Adherence to REACH Guidance on reporting Weight of Evidence
 - Discussion of the bioaccumulation characteristics of metabolites, where applicable

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ECOTOXICITY



ECOTOXICITY

In particular for human toxicity, information shall be generated whenever possible by means other than vertebrate animal tests, through the use of alternative methods, for example, in vitro methods or

qualitative or quantitative structure-activity relationship models or from information from structurally related substances (grouping or read-across).

REACH Article 13, EC 1907/2006

- Main factor for change remains drive to avoid vertebrate testing
- Non-testing methods enshrined into REACH
- 3 areas highlighted:
 - *In vitro* methods
 - QSARs
 - Grouping and Read-across approaches

In vitro methods

- The ability of cell systems to predict whole organism toxicity
- Includes foetal or embryonic forms of vertebrate species
- Not easy to predict whole organism toxicity from cell measurements
- Potential to under-estimate *in vivo* toxicities (up to 1000x)
- Limited regulatory use so far for REACH (ECHA 2014)

In vitro methods

- Underestimates avoidable through suitable precautions
- Results with Rainbow trout gill cell line show promise
- Fish embryo tests more advanced
 - Good correlation with acute toxicity data
 - OECD 236 approved in 2013

QSARs

- Quantitative Structure Activity Relationships
- The prediction of a substance's ecotoxicological characteristics from its physicochemical properties
- Extensive use for LogP_{ow}
- Toxicological application more complex
- Understanding of toxic mode of action key to success.....

QSARs

-regulatory acceptance also essential
- Requires mechanistic transparency
- Requires quality data sources

However....

- Suitable platforms now available which are mechanistically transparent and freely available.



toxRead

what's toxRead | how to use | developers | downloads | contacts

toxRead

- is a software to assist user in making reproducible read across evaluations.
- shows the similar chemicals, structural alerts and relevant features in numerous institutions

VEGA | TRY THE NEW TOXREAD | DOWNLOAD VEGA | QSAR REGULATION & RESEARCH | ABOUT QSAR READ ACROSS | COMMUNITY

ToxBank | caleidos | ORCHESTRA | ANTORES | VEGA | CRS | caleidos

originated from:

Chemicals with associated experimental values

Libraries of chemicals with associated experimental values were checked and originated from the LIFE projects

Structural alerts and algorithms of relevant features

Structural alerts derive and relevant features libraries originated from the used within VEGA, ToxTree, and other tools developed within the LIFE

ANTORES | VEGA | CRS | caleidos | PRASIL

OECD.org | Data

OECD

BETTER POLICIES FOR BETTER LIVES

OECD Home | About | Countries

OECD Home - Chemical safety and toxicity - Assessment of chemicals

Testing of chemicals

Assessment of chemicals

Risk management of chemicals

The OECD QSAR Toolbox

To increase the regulatory acceptance of (Q)SAR methods, the OECD is developing a QSAR toolbox to make (Q)SAR technology readily accessible, transparent, and less demanding in terms of infrastructure costs.

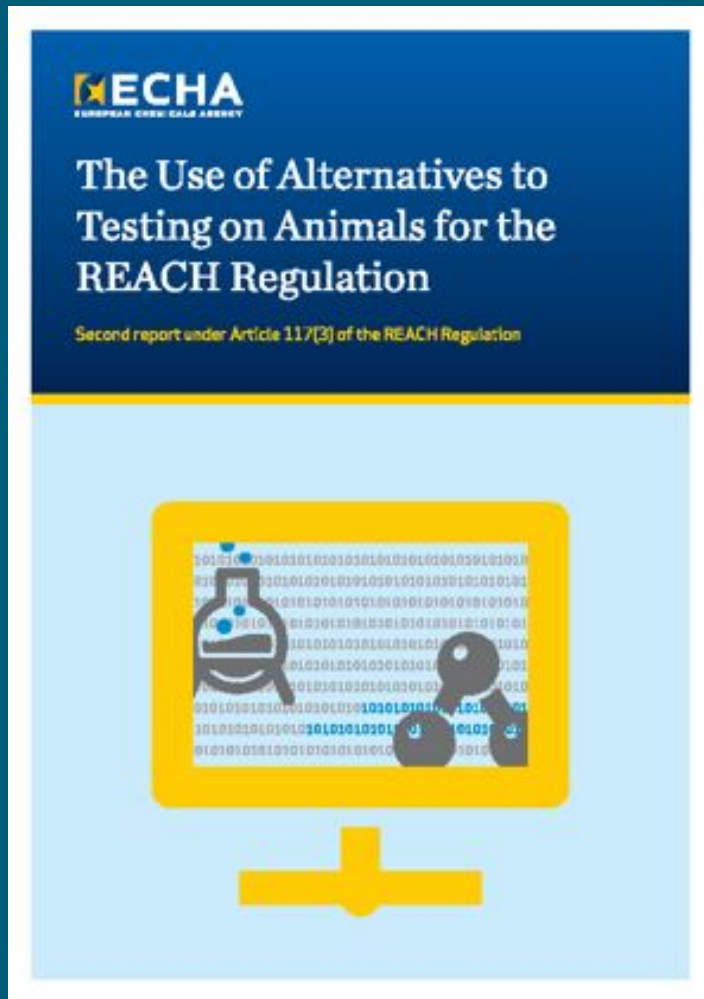
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QSARs

Words of Warning:

- Data quality may not be formally guaranteed (OECD Toolbox)
- Most models based on LogP_{ow} – CAUTION!
Not applicable to surfactants
- A QSAR is not a substitute for ecotoxicological knowledge!

Regulatory use of QSARs



- Accepted for OSPAR HMCS registration
- Usage described in OSPAR Agreement 2012-05 (HOCNF Guidelines)
- Usage in REACH registrations monitored every four years

Use of QSARs in short-term fish toxicity studies [ECHA 2014].

Tonnage band	QSAR usage, from 2010 data			QSAR usage, from 2014 data		
	Total reports	Reports based on QSAR	(% of total reports)	Total reports	Reports based on QSAR	(% of total reports)
100-1000	6942	147	2.1	8917	330	3.7
>1000	1405	18	1.3	6104	120	2.0
Non-phase-in substances	143	3	2.1	362	12	3.3

- **Limited application to date**
- **But usage is increasing**

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- Ecotoxicological alternatives to acute fish test also permitted under the HMCS, but few validated methods so low take-up

CONCLUSIONS

New approaches to the assessment of persistence, bioaccumulation and toxicity in progress, but there are challenges in each area:

- WoE to assess bioaccumulation is acceptable under the OSPAR HMCS, but so far under-used
- Ecotoxicological alternatives to acute fish test also permitted under the HMCS, but few validated methods so low take-up
- OSPAR Protocols may require modification to accept new approaches to persistence assessment

Thank you !