

# Minimising animal tests through education and training

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# Overview

- PETA International Science Consortium Ltd.
- Education and training
  - Training sessions
  - Outreach
  - Webinars
- REACH webinar series



# PETA International Science Consortium



# Education and Training

- Training sessions
- Outreach: [pisc ltd.org.uk](http://pisc ltd.org.uk)
- Webinars

# Training Sessions



# Outreach

## FACTSHEETS

### IN VITRO METHODS FOR SERIOUS EYE DAMAGE AND EYE IRRITATION

**START here if you suspect your test substance causes serious eye damage**

Use of existing data and QSAR modelling

**TOP-DOWN APPROACH:** start with use of the method/chemicals below themselves

OECD 104/105 AOP  
OECD 104/105  
OECD 104/105  
OECD 104/105

STOP testing, test substance should be labelled as GHS Cat 1

STOP testing, test substance should be labelled as GHS Cat 2

**START here if you suspect your test substance is not an irritant**

Use of existing data and QSAR modelling

**BOTTOM-UP APPROACH:** start with use of methods similar to those below

OECD 104/105 AOP  
OECD 104/105  
OECD 104/105  
OECD 104/105

STOP testing, test substance should be labelled as GHS Cat 1

STOP testing, test substance should be labelled as GHS Cat 2

STOP testing, test substance should be labelled as GHS Cat 3

STOP testing, test substance should be labelled as GHS Cat 4

#### References

If the substance is not classified as GHS Cat 1 or GHS Cat 2, the substance is likely to be GHS Cat 3. To avoid over-testing, additional testing may be conducted with *in vitro* methods that allow classification of Cat 3 chemicals in a weight-of-evidence approach. *In vitro* methods such as the EOP, Pao-COR or similar ones, which address uncertainties in the absence of severity, may be suitable. For more information on tested testing strategies for serious eye damage and eye irritation, please see Wood & al., 2014. Alternative methods for regulatory toxicology – a state-of-the-art review. European Union Reference Laboratory for Research on Toxicological Testing and Scott & al., 2015. A proposed eye irritation testing strategy to reduce and replace *in vivo* studies using bottom-up and top-down approaches. Toxicology in Water 14 (1-3).

METHOD	PRINCIPLE OF THE TEST	APPLICABILITY DOMAIN	GHS CATEGORISATION
OECD 104/105. Bovine Corneal Opacity and Permeability (BCOP) Test Method for Identifying Chemicals Causing Serious Eye Damage and 10 Chemicals for Identifying Chemicals Causing Eye Irritation or Serious Eye Damage	Test substance is directly applied to cow eyes obtained as by-products from abattoirs. Corneal opacity measured qualitatively as the amount of light transmission through the cornea and permeability measured quantitatively as the amount of sodium fluorescein dye that passes across the full thickness of the cornea; an measured. Optimal test methodology can be conducted for additional information.	Applicable to solids, liquids (including semi-solids), creams and emulsions.	For the identification of substances causing serious eye damage 10/60 Cat 1 and substances not requiring classification for eye irritation or serious eye damage. OECD 104/105 testing video available at <a href="http://www.youtube.com/watch?v=Zp9z6D9E">www.youtube.com/watch?v=Zp9z6D9E</a>
OECD 104/105. Rabbit Cornea Eye (RCE) Test Method for Identifying Chemicals Causing Serious Eye Damage and 10 Chemicals for Identifying Chemicals Causing Eye Irritation or Serious Eye Damage	Test substance is directly applied to chicken eyes obtained as by-products from abattoirs. Corneal swelling, opacity and fluorescence retention are assessed.	Applicable to solids (except for solids or solids in water), liquids, emulsions and gels.	For the identification of substances causing serious eye damage 10/60 Cat 1 and substances not requiring classification for eye irritation or serious eye damage.
OECD 104/105. Fluorescent Leakage (FLU) Test Method for Identifying Chemicals Causing Serious Eye Damage and 10 Chemicals for Identifying Chemicals Causing Eye Irritation or Serious Eye Damage	Epithelial monolayer Mad-Darby cornea lining MCEC3 cells are cultured in permeable inserts. The test chemical is applied for 3 minutes and then removed, next, the non-toxic highly fluorescent sodium-fluorescein dye is added, and the amount of dye that passes through the cell layer is measured. Spectrophotometric light scatter predicts toxicity.	Applicable to water-soluble chemicals or mixtures. Limitations for substances of high viscosity substances (especially oils) is overcome by increasing the number of wash steps. Not applicable to strong acids and bases, self-heating or highly volatile substances.	For the identification of substances causing serious eye damage 10/60 Cat 1.
OECD 104/105. In Vitro Eye Irritation (IIEI) Assay	Measures cell viability (MTT assay) of 3RGC Ocularles (derived from Rabbit Cornea) corneal epithelial cells or 3RGC and others. Reagents are generally obtained from human eyes in 1 to 2 minutes or rabbit eyes in 3 to 4 minutes. The test requires a 5-minute exposure.	Applicable to test chemicals that are soluble in saline, DMEM or normal oil.	For the identification of substances causing serious eye damage 10/60 Cat 1 and substances not requiring classification for eye irritation or serious eye damage.
OECD 104/105. Bovine Corneal Opacity and Permeability (BCOP) Test Method for Identifying Chemicals Causing Serious Eye Damage and 10 Chemicals for Identifying Chemicals Causing Eye Irritation or Serious Eye Damage (eg. Epithelial)	Measures Epithelial Permeability is reconstructed from primary bovine cells, which have been cultured for several days to form a stratified, highly differentiated equine epithelium morphologically similar to that found in the bovine cornea. The test substance is applied to the cornea, and cell viability (MTT assay) is used to predict toxicity.	Applicable to substances and mixtures as well as solids, liquids, semi-solids and emulsions.	For the identification of substances causing serious eye damage 10/60 Cat 1 and substances that are not classified.
OECD 104/105. Bovine Corneal Opacity and Permeability (BCOP) Test Method for Identifying Chemicals Causing Serious Eye Damage and 10 Chemicals for Identifying Chemicals Causing Eye Irritation or Serious Eye Damage	A sub-confluent monolayer of human U2OS cells is exposed to increasing concentrations of the test chemical. The cellular metabolic rate – measured by pH change in the medium conditioned – is used to predict toxicity.	Applicable to water-soluble chemicals (substances and mixtures) as well as water-soluble solids, gases, chemicals and emulsions that maintain uniformity during the analysis time.	For the identification of substances causing serious eye damage 10/60 Cat 1 and substances that are not classified.

## Award for Adverse Outcome Pathway Development Provided to New Contributors to the AOP Wiki

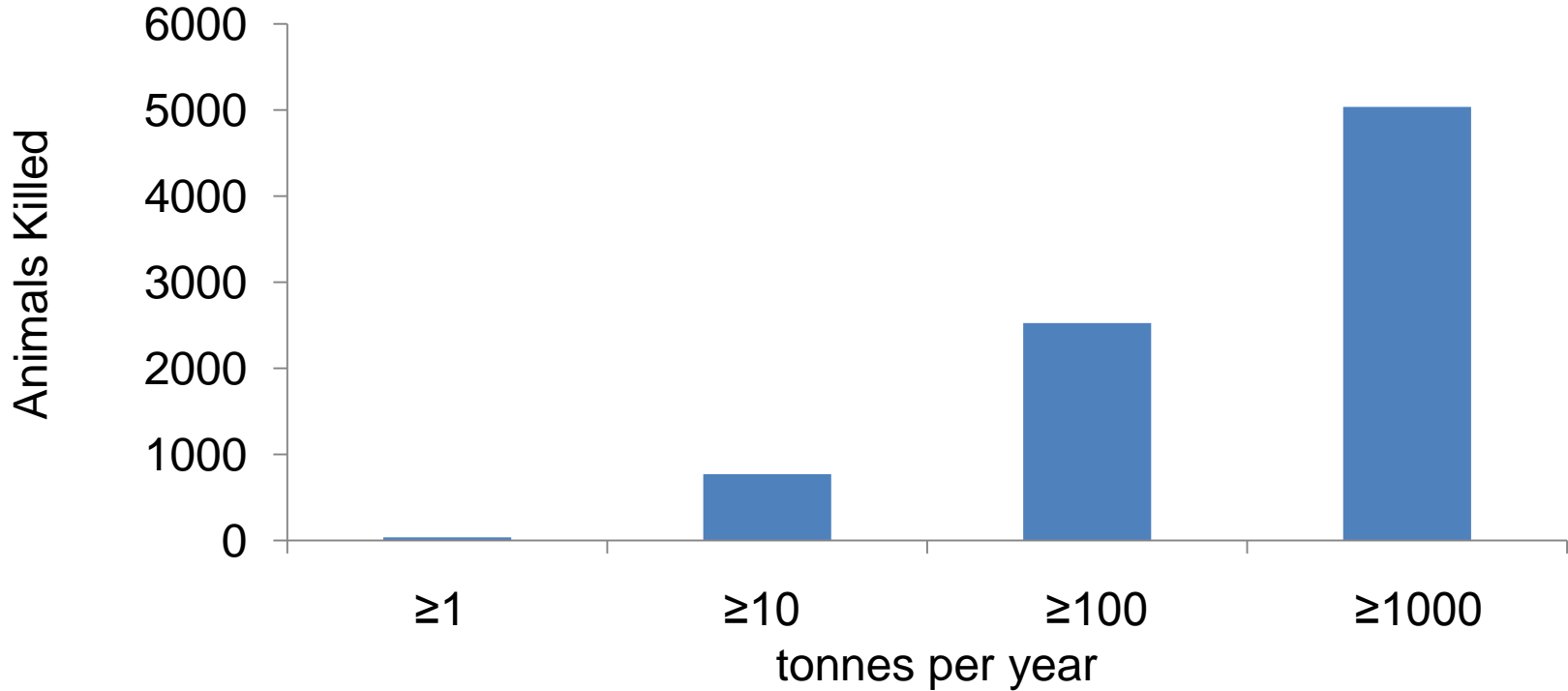
With the advent of emerging technologies and the adoption of a new vision for the science of toxicity testing, modern toxicology is moving away from the animal tests scientists have traditionally relied upon to identify chemical hazards. Tests using animals are often considered to be “black box” studies that determine whether a chemical is toxic to animals, but they do not necessarily reveal the mechanism by which the substance caused toxicity or whether it would cause similar toxicity in humans. In recent years, however, non-animal testing strategies have been developed that are geared towards probing the specific mechanism of chemical toxicity. These testing strategies can be based on Adverse Outcome Pathways (AOPs), which are a conceptual framework describing a sequential chain of causally linked events at different levels of biological organisation that lead to an adverse health or ecotoxicological effect.

As part of a collaborative effort between the European Commission's Joint Research Centre, the US Environmental Protection Agency, and the Organisation for Economic Co-operation and Development, an [AOP Wiki](#) has been created to provide an interactive and virtual platform for AOP development and to promote international consensus on the developed AOPs. Working with the organisers of the AOP Wiki, the PETA International Science Consortium Ltd. (PISC) is launching a data challenge to encourage new contributors to add to existing entries in the AOP Wiki using available data.

Launch of the AOP Award at the European Commission's Joint Research Centre (JRC) booth at the 51<sup>st</sup> annual EUROTOX Congress held in Porto, Portugal on 13-16 September, 2015.



# Animal Testing for REACH



# Webinar Series: Leading Experts

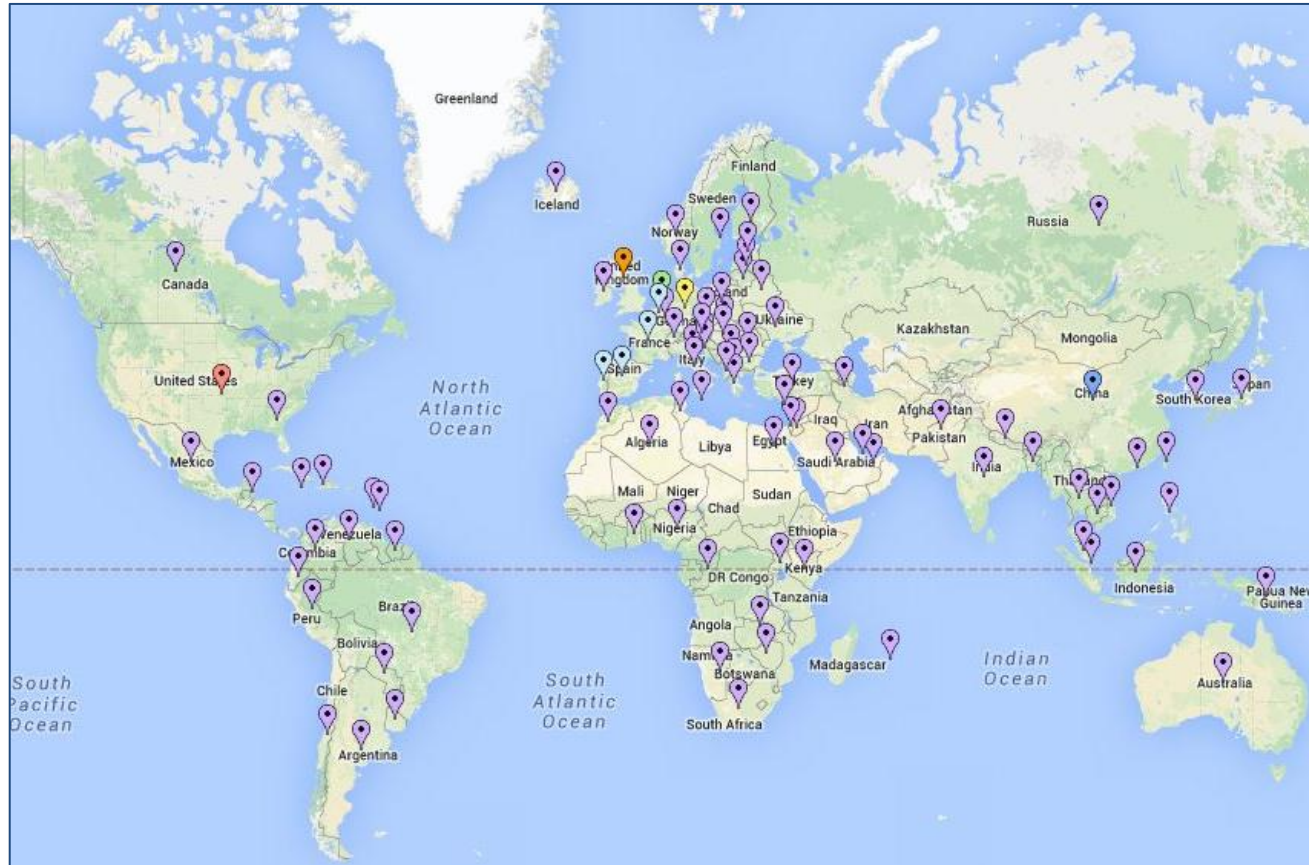




# Webinars

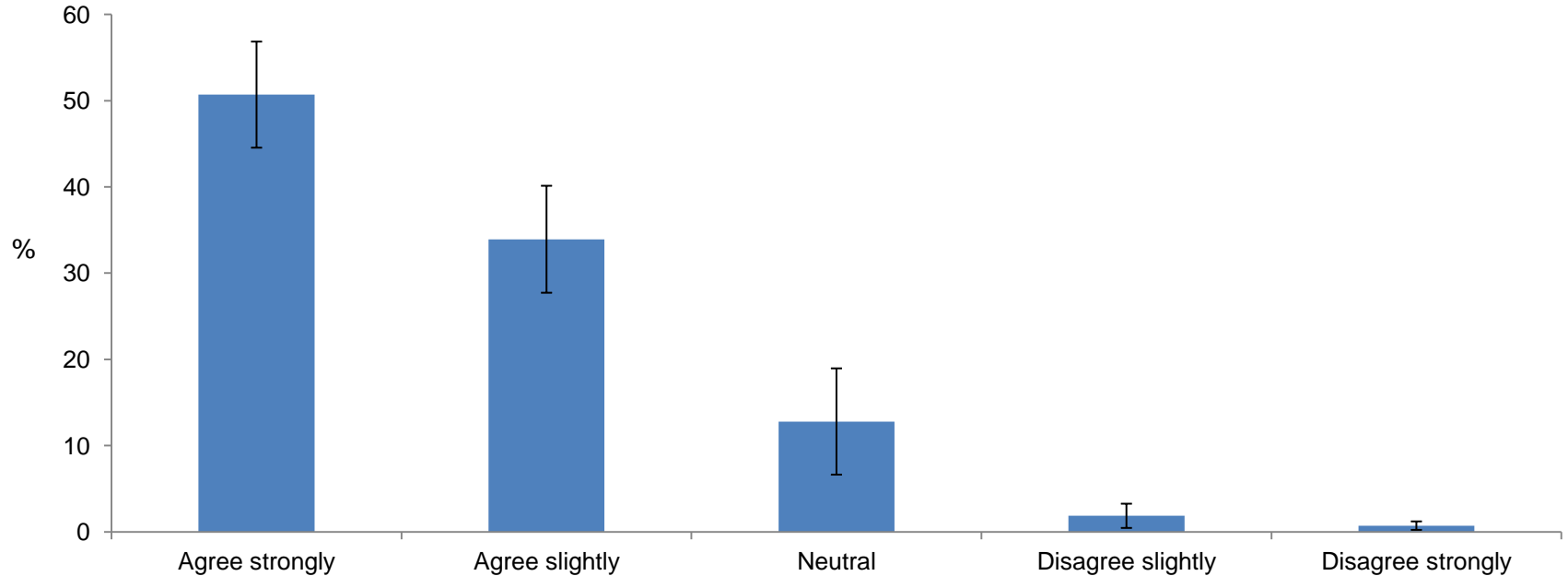
WEBINAR 1	OECD QSAR TOOLBOX AND READ-ACROSS	Dr. Grace Patlewicz, formerly of DuPont Prof. Mark Cronin, Liverpool John Moores University
WEBINAR 2	SKIN IRRITATION AND CORROSION	Dr. Gertrude-Emilia Costin, Institute for In Vitro Sciences Dr. Costanza Rovida, REACH Mastery and CAAT Europe
WEBINAR 3	SERIOUS EYE DAMAGE AND EYE IRRITATION	Dr. Kim Norman, Institute for In Vitro Sciences Dr. João Barroso, EURL ECVAM
WEBINAR 4	SKIN SENSITISATION	Dr. Susanne Kolle, BASF SE Dr. Silvia Casati, EURL ECVAM
WEBINAR 5	ALTERNATIVE APPROACHES TO MAMMALIAN ACUTE SYSTEMIC TOXICITY TESTING	Dr. Pilar Prieto, EURL ECVAM Dr. Lawrence Milchak, 3M
WEBINAR 6	(ZEBRA)FISH EMBRYO ACUTE TOXICITY TEST TO PREDICT SHORT-TERM TOXICITY TO FISH (AND BEYOND)	Dr. Marlies Halder, EURL ECVAM Prof. Thomas Braunbeck, University of Heidelberg Dr. Scott Belanger, Procter & Gamble
WEBINAR 7	THE REGULATORY PROCESSES INVOLVED IN ACCEPTANCE OF NON-ANIMAL TESTS	Dr. Derek Knight, ECHA Ms. Karin Kilian, European Commission

# International Audience



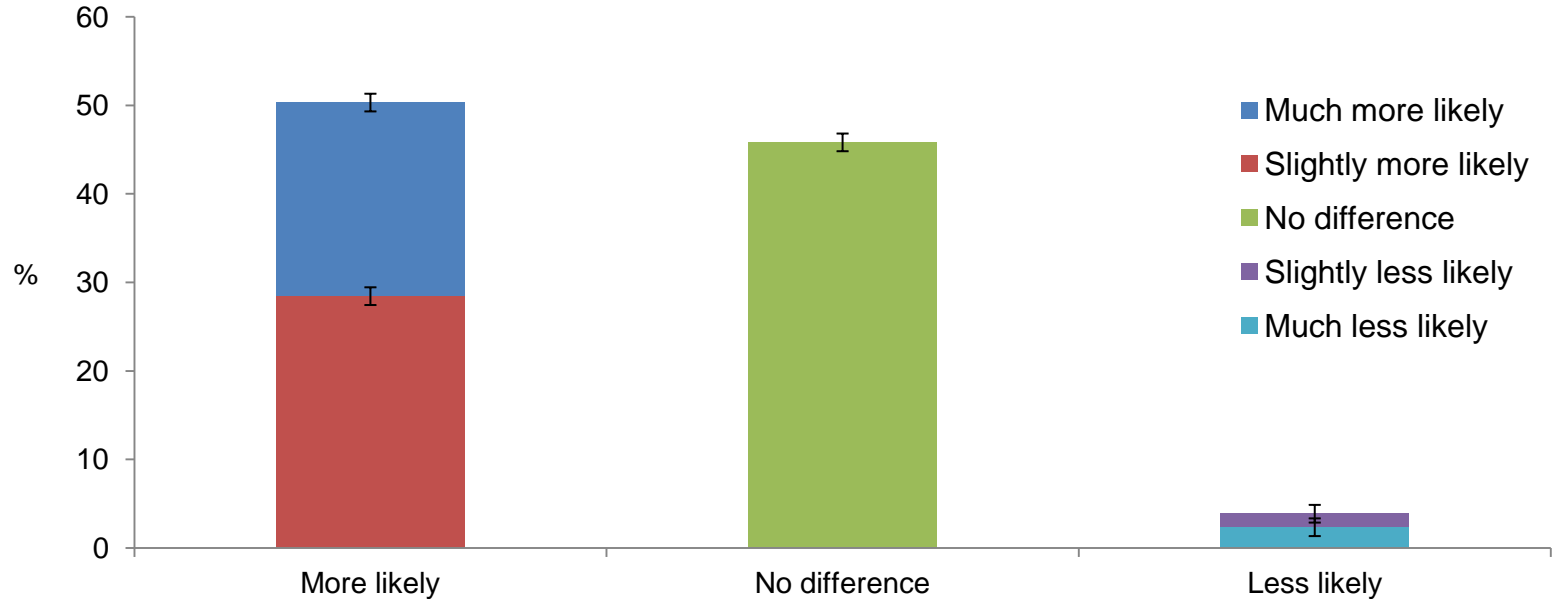
# Awareness of Testing Strategies

Q: “I gained some useful guidance on how to incorporate non-testing/non-animal methods into a testing strategy”.



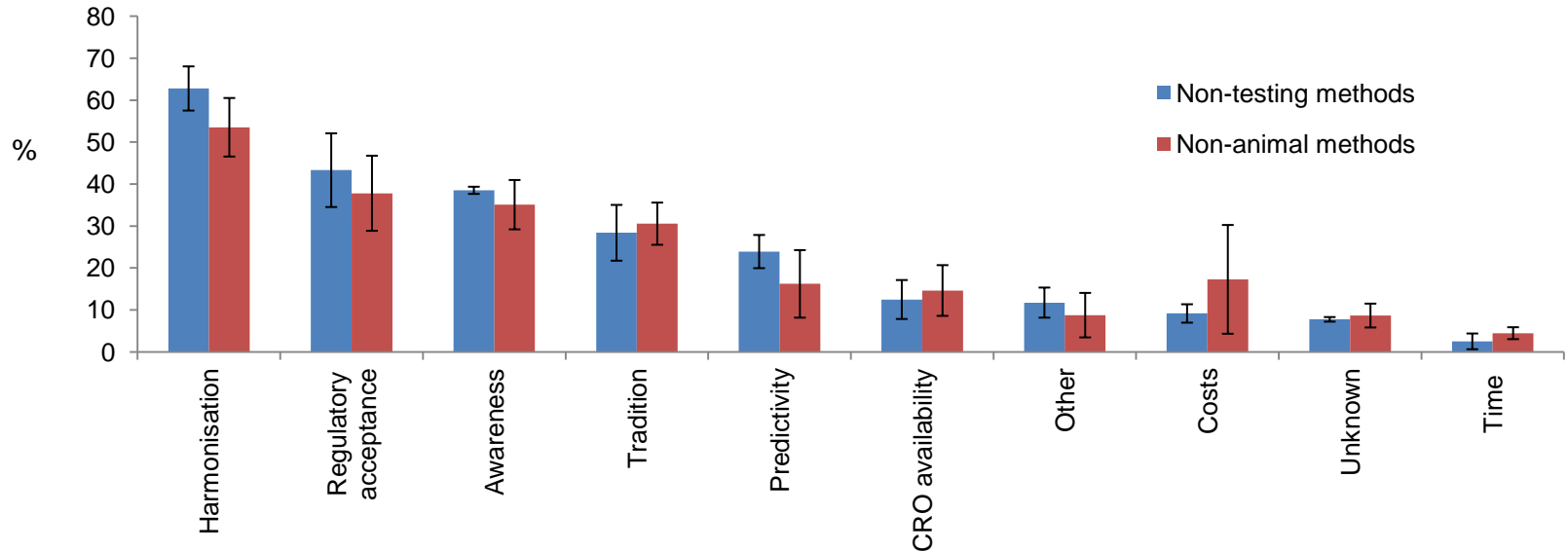
# Likely Use of Non-Testing Methods

Q: “As a result of attending this webinar, are you more or less likely to use non-testing methods for REACH 2018”?



# Barriers to Uptake of Non-Testing and Non-Animal Methods

Q: “What do you think are the main barriers to the uptake of a) non-testing and b) non-animal testing methods for REACH”?



# Conclusions

- The Science Consortium uses a multifaceted approach to education and training, including:
  - Training sessions
  - Outreach
  - Webinars
- REACH webinar series
  - The Science Consortium teamed up with Chemical Watch and leading experts
  - Feedback indicates that the webinars were effective in increasing the intent of companies to use of non-animal methods.



# Acknowledgements



# Thank you!

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