Replacing Animal Testing for Assessing Consumer Safety

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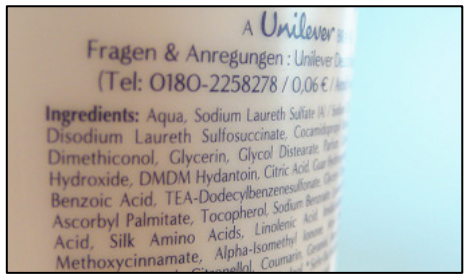
Non-animal Test Methods

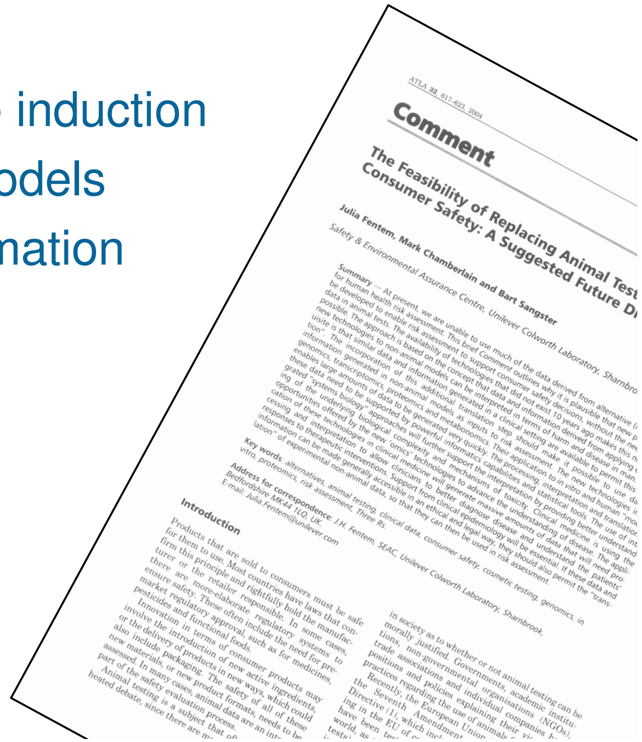
- **Past:**
  - hazard focus
  - emphasis on tests for classification and labelling (‘positives/negatives’)
  - direct replacement of a specific animal test

- **Present:**
  - focus on non-animal approaches for consumer safety **risk assessment**
  - data required for **safety decision** should be driver
  - non-animal testing strategy capable of characterising the key hazard parameters modulated by chemicals of interest



Conceptual approach

- What new information do I need to make a risk assessment decision without animal testing?
  - Risk = Hazard x Exposure
  - Exposure
    - Developing our new risk assessment approaches around exposure information (e.g. new applications of TTC)
  - Hazard
    - Understanding the pathways of human disease induction
    - New non-animal (*in vitro, in silico*) predictive models
    - New ways to interpret, weight & integrate information
    - Evaluating the usefulness of new technologies
    - Maximising the use of historical animal data


ATLA 32, 617–623, 2004

Comment

The Feasibility of Replacing Animal Test... Consumer Safety: A Suggested Future D...

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Introduction

Conceptual approach – in practice, the last 6 years

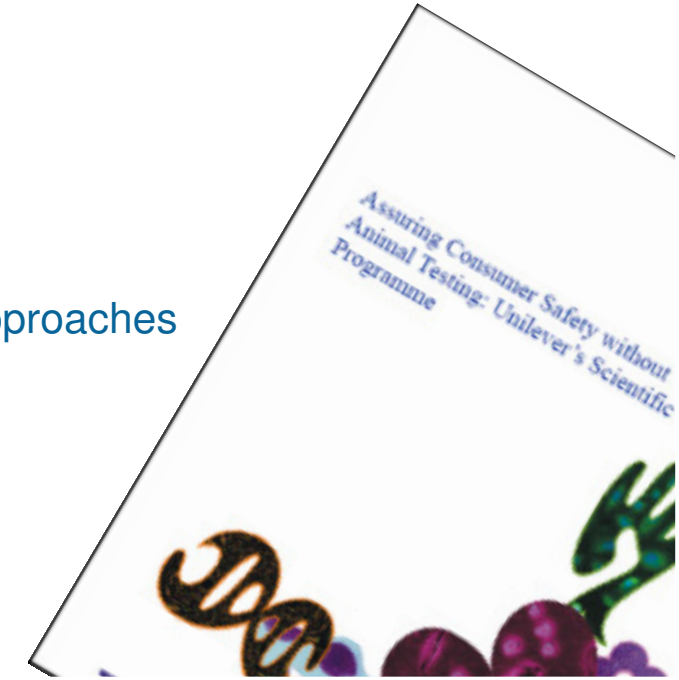
- Three scientific areas of interest - the same long term goal
    - Risk-based safety decisions without new animal data

- **Skin Allergy**
    - Assessment of model performance both in isolation and within context of a new risk assessment framework
    - Assessment and prioritisation of current knowledge gaps
- **Cancer**
    - Fundamental understanding of skin cancer & chemical carcinogenicity
    - New models, technologies and tools for future weight-of-evidence / mode-of-action Risk Assessments
- **General Toxicity**
    - What questions are we asking?
    - Which models and technologies can help to answer these questions?
        - Lung toxicity as a case study

Conceptual approach – in practice, the last 6 years

- Internal Unilever (SEAC) research
- External research
  - Unilever-sponsored academic research
  - Evaluating new approaches with contract research organisations
  - Initiating bespoke research with biotechnology companies

- External scientific partnerships
  - Involvement with EU-funded projects
  - Participation in cross-industry collaborative research
    - Colipa
  - Working with other scientific groups on alternative approaches
    - UK NC3Rs
    - EPAA
    - US ‘Human Toxicology Project’ consortium

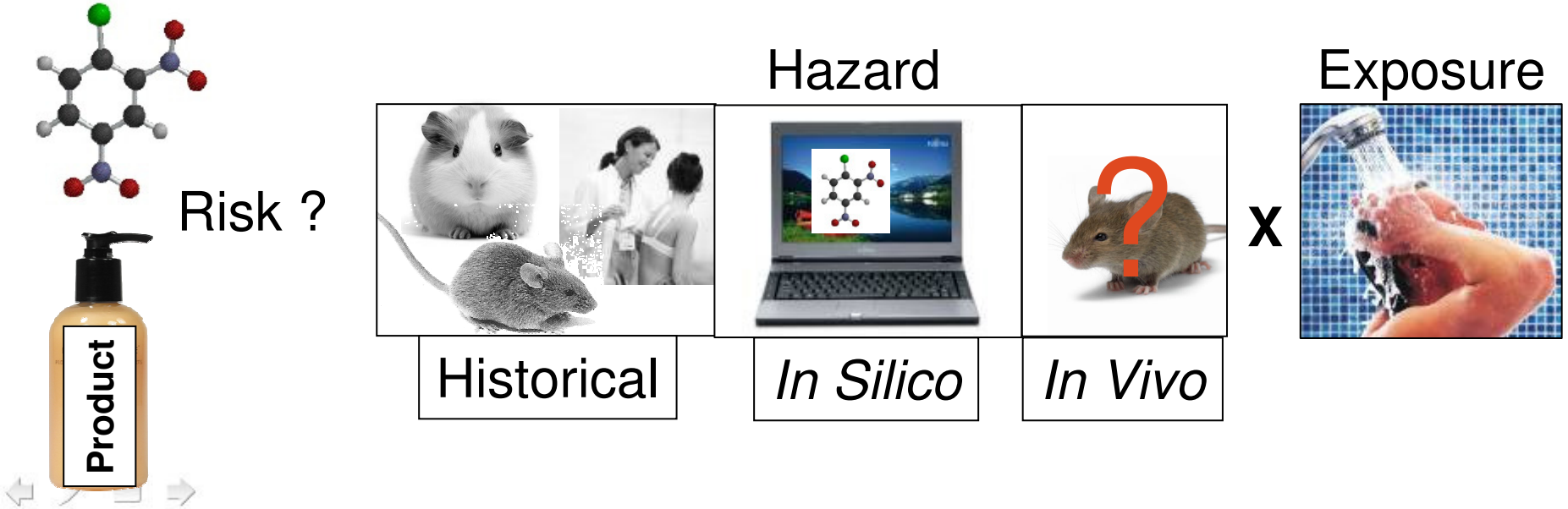


Skin Allergy case study

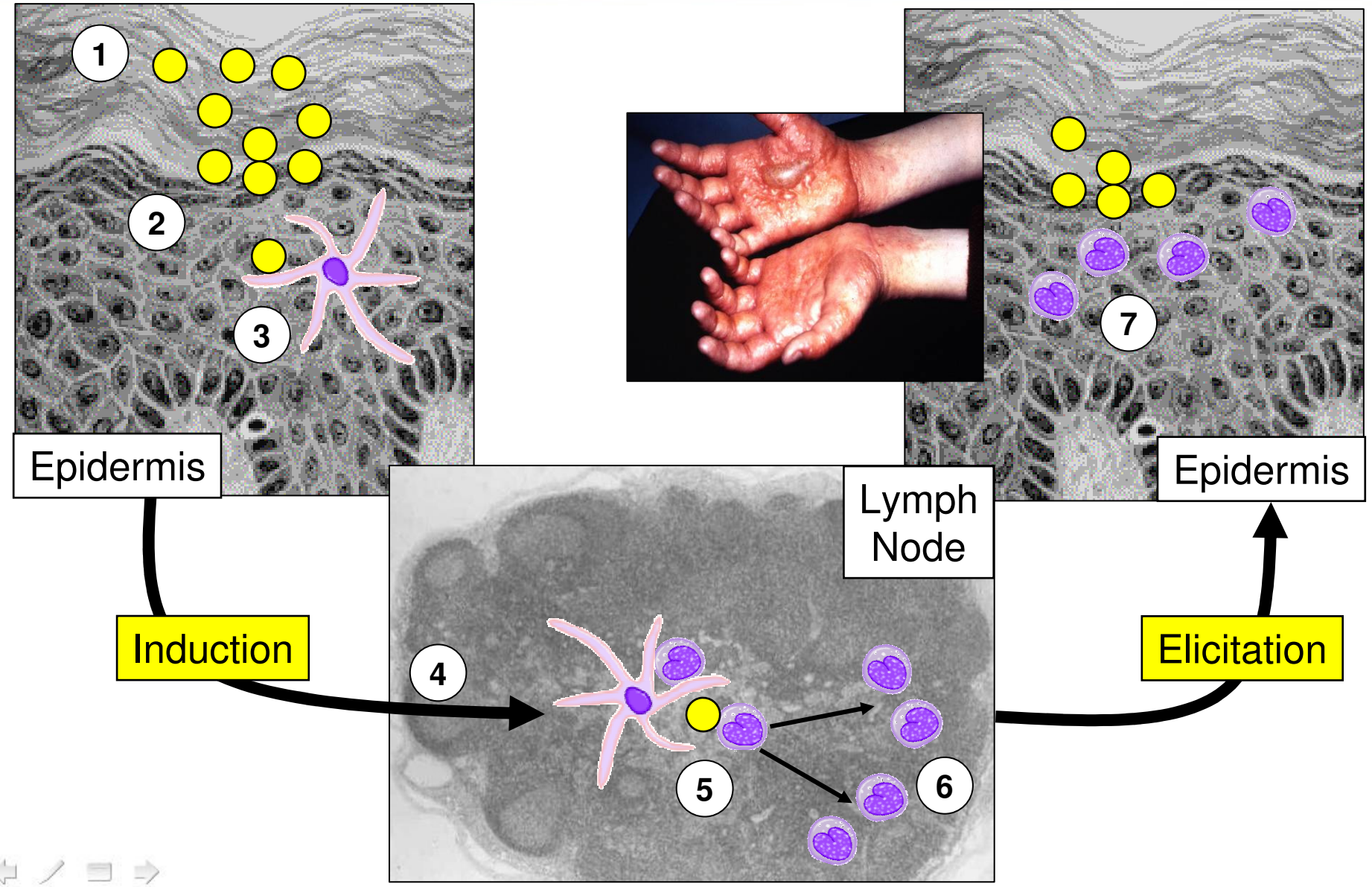
New Risk Assessment Approach for Skin Sensitisation

Key questions moving forward:

- What information do we require to inform our risk assessment approach for skin sensitisation (without animal testing)?
- How might our new risk assessment approach need to evolve to accommodate these new types of information?



What are we trying to prevent?
- Allergic Contact Dermatitis

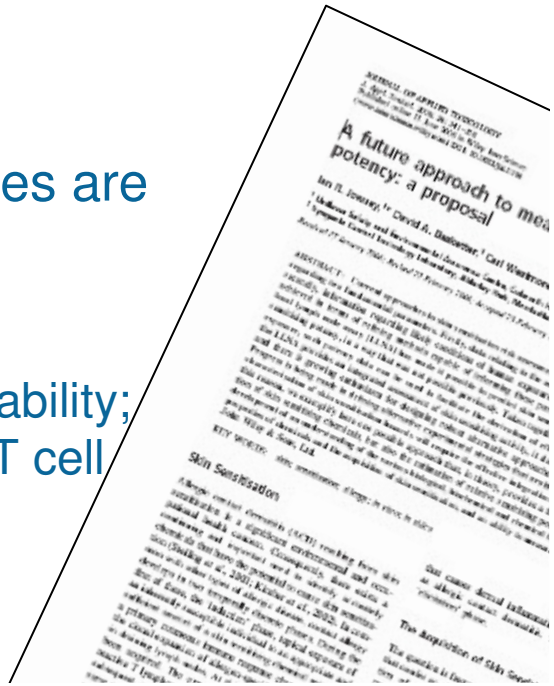


Non-animal test methods for Hazard Characterisation

- Current consensus: several non-animal hazard characterisation test methods will be required to predict skin sensitiser potency & dose response information

- Several major programmes of research and method development underway (e.g. COLIPA Skin Tolerance TF and Sens-it-*iv* EU Framework VI project)

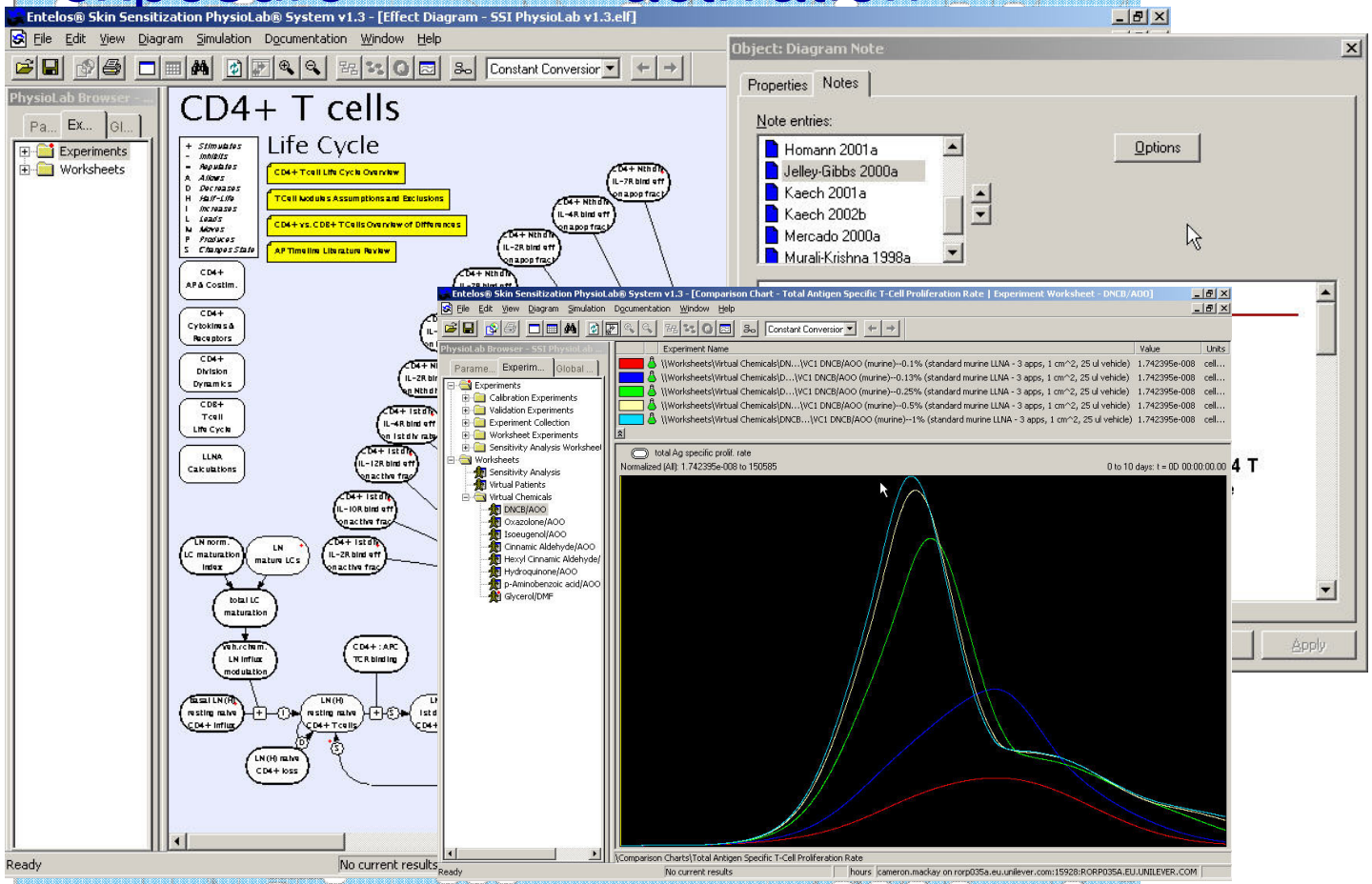
- A variety of *in silico, in chemico* and *in vitro* approaches are being developed to encompass key events in Skin Sensitisation induction:
  - Chemical / Peptide reactivity; Skin disposition / bioavailability; Skin inflammation; Dendritic cell activation/maturation; T cell proliferation


A future approach to mea
potency: a proposal
Skin Sensitisation

Defining and quantifying the relative value of the key parameters of Skin Sensitisation

Chemical exposure

Epidermal cell activation



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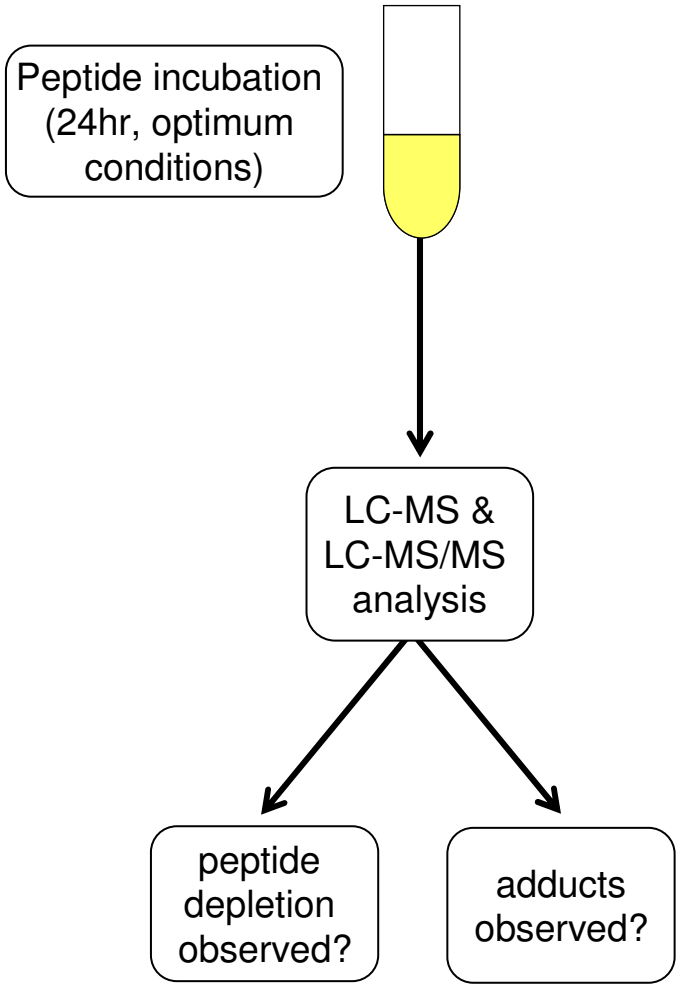
Maxwell G. & MacKay C. 2008. *ATLA*. **36**. 521-556

Testing Strategies for Skin Sensitisation

- Chemical-specific data generated across different *in vitro* approaches using well-characterised chemicals to provide 'lines of evidence':
  - Dermal Kinetics
  - Peptide Reactivity
  - Epidermal Inflammation
  - Dendritic cell activation
- Data analysed using statistical models (PCA, PLS, Linear regression, Clustering, Decision trees) to establish predictive capacity of measurements
- Probabilistic (Bayesian) approach is being developed to explore strategies for integration of different data types to predict sensitiser potency & dose response information

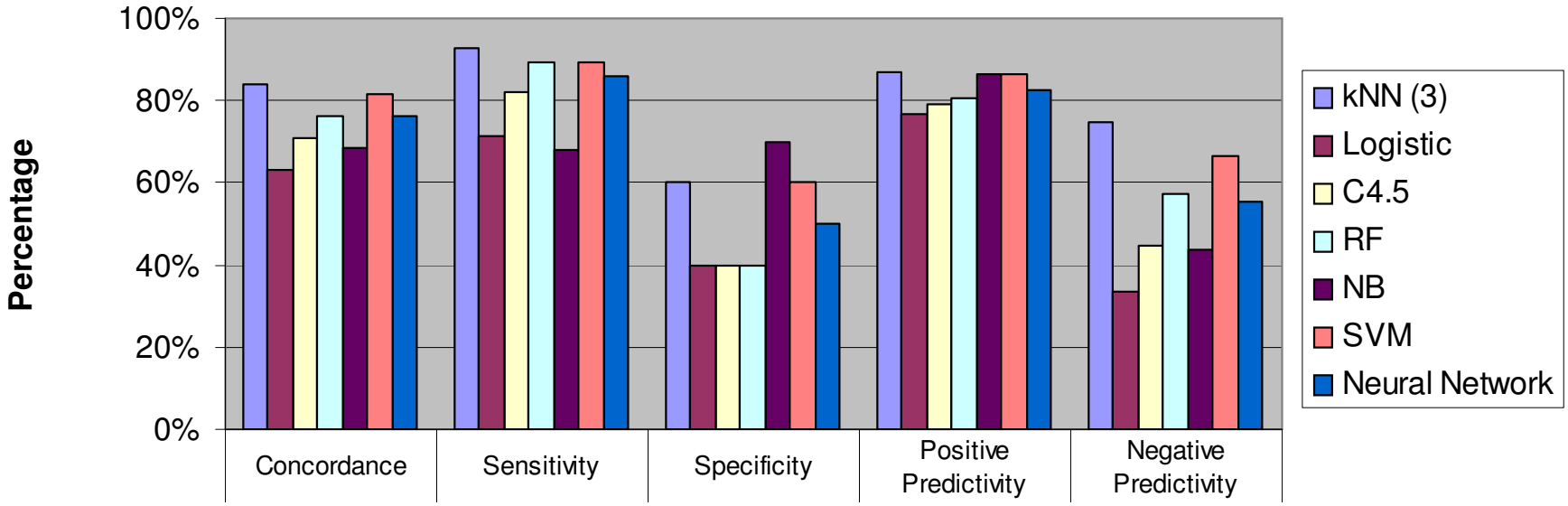
Example: Peptide Reactivity



- **Hypothesis**: covalent modification of protein must occur for a chemical to be a sensitiser
- Peptide depletion and adduct formation measured by LC/MS/MS
- Six different target amino acids each within a different model peptide
- If no adducts are observed chemical is assumed to be non-reactive and therefore non-sensitising (without transformation)
- Hierarchical clustering indicates that high depleting chemicals are more likely to be potent sensitisers

Aleksic et al (2009) Toxicol Sci, **108,** 401-11

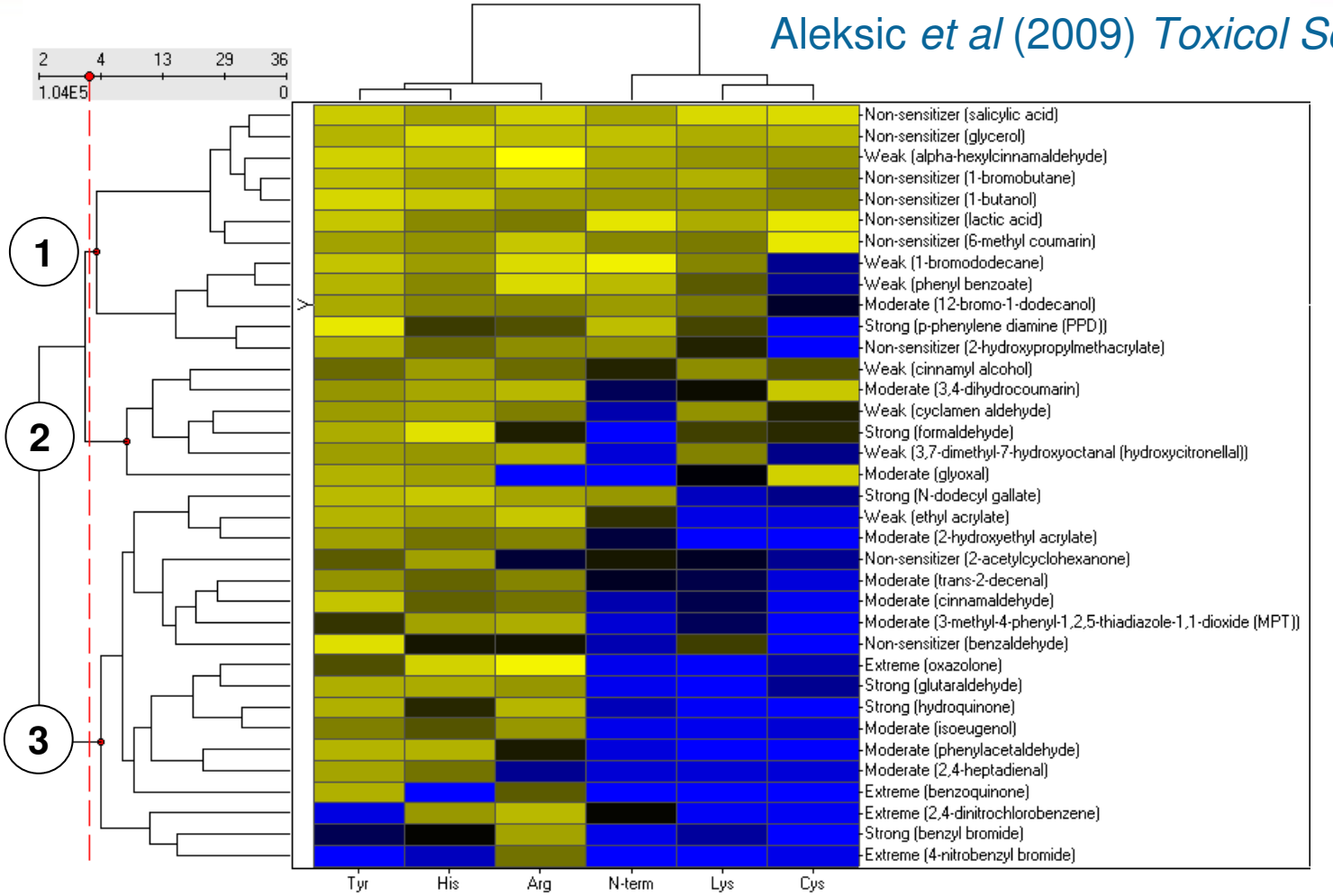
Peptide reactivity depletion data provides valuable information on sensitisation potential



- Peptide reactivity data from 28 sensitisers & 10 non-sensitisers analysed for sensitiser potential predictions using 7 different forms of statistical analysis
- Reactivity data found to be more accurate when identifying sensitisers than when identifying non-sensitisers
  - Identification of non-sensitisers is challenging due to the existence of reactive non-sensitisers
- Integration of data from other ‘lines of evidence’ is required to improve the overall prediction of sensitisation potential

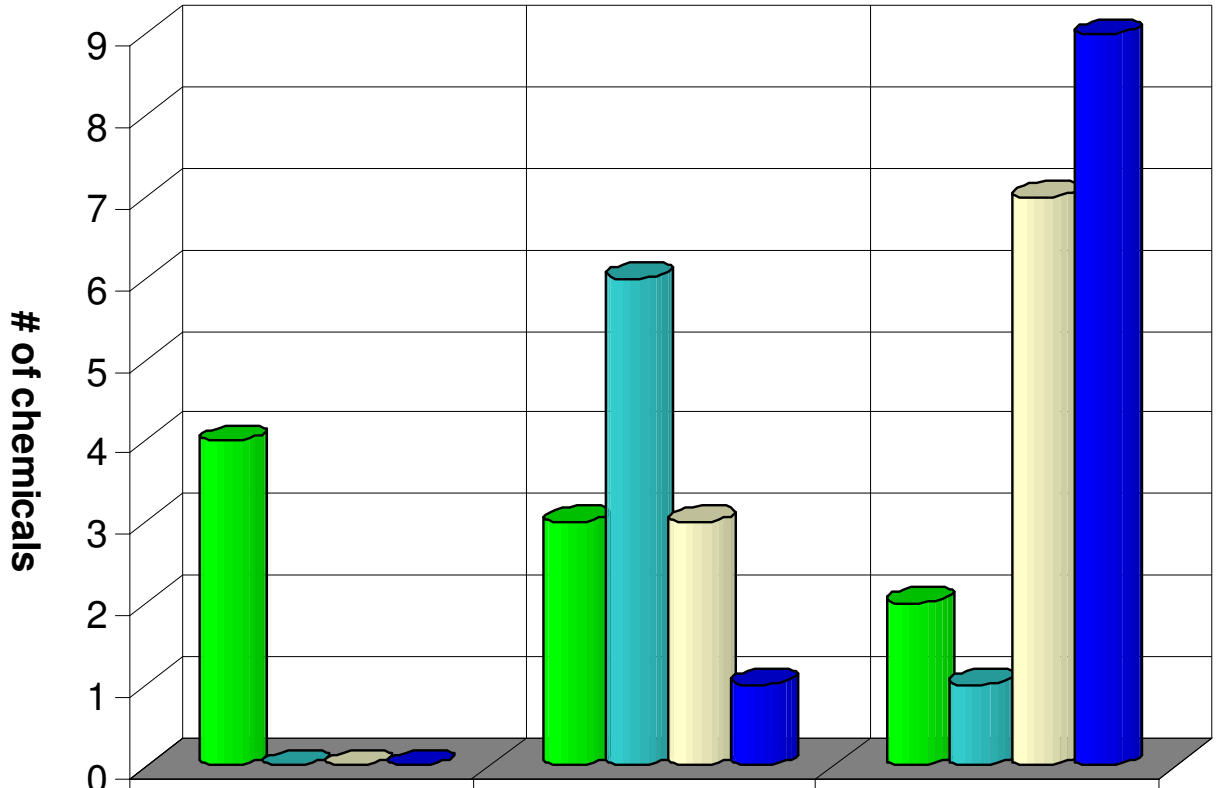
Sensitiser potency and model peptide depletion

Aleksic *et al* (2009) *Toxicol Sci,* **108,** 401-11



- Peptide reactivity data analysed using hierarchical clustering method
- Level of Depletion - Blue = high; Black = medium; Yellow = low

Promiscuity of chemical binding vs. Sensitiser potency (36 chemicals)



	0 Peptides	1-3 Peptides	4-6 Peptides
Non-sensitizer	4	3	2
Weak	0	6	1
Moderate	0	3	7
Strong/Extreme	0	1	9

Non-sensitizer
Weak
Moderate
Strong/Extreme

Number of peptides in which one or more adducts are formed

Summary

- How can we assess consumer safety without new animal test data?

- Skin allergy case study [Maxwell G. *et al*. 2008. *ATLA*. **36**. 557-568]
  - Understanding the key parameters driving human disease induction
  - Develop & evaluate non-animal models to predict these parameters
  - Interpret and integrate information from these models
  - Develop exposure-driven risk assessment approaches to accommodate these new types of hazard information

- Cancer & General Toxicity
  - Even more challenging - mode/mechanism of action is not well-defined for individual chemicals and can vary significantly between chemicals

Scientific Partnerships

Unilever, SEAC

Yeyejide Adeleye, Maja Aleksic, Nora Aptula, Simon Aveston, Christine Boucheron, Clare Bourner, Phil Briggs, Paul Carmichael, Phil Carthew, Kristina Castle, Peter Chapman, Catherine Clapp, Brett Cochrane, Stella Cochrane, René Crevel, Michael Davies, Matthew Dent, Sue Edwards, Julia Fentem, Sam Fletcher, Annette Furniss, Andrew Garrow, Nicky Gilmour, Steve Gutsell, Michael Hughes, Sandrine Jaquoilleot, Donna Jefferies, Penny Jones, Ian Jowsey, Denis Kan-King-Yu, Anja Lalljie, Sue Larbey, Jin Li, Martin Macfarlane, Cameron MacKay, Sophie Malcomber, Gavin Maxwell, Leona Merolla, Helen Minter, Sandeep Modi, Claire Moore, Tom Morrow, Denise Nason, Deborah Parkin, Camilla Pease, Ruth Pendlington, Ed Price, Trannum Rahim, Fiona Reynolds, Bob Safford, David Sanders, Andrew Scott, Sharon Scott, Mary Spurgeon, Vicki Summerfield, Emma Thain, Andrew White, Sam Windebank, Carl Westmoreland and Raniero Zazzeroni

"Part of Unilever's ongoing effort to develop novel ways of delivering consumer safety"

Thank You – Any questions?

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