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How omics technologies can enhance chemical safety regulation

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Environmental Toxicology & Chemistry

How OMICs Technologies Can Enhance Chemical Safety Regulation: Academia, Government and Industry Perspectives

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Abstract:	The current rapid pace of innovation of industry is delivering an ever-increasing number and diversity of chemicals within industrial and consumer products. Regulations require an evaluation of the environmental and human hazards of chemicals, ultimately characterizing the risks associated with their manufacture, use and disposal. In order for policy makers and regulators to carefully balance economic priorities with the need to protect vital ecosystems and public health, equally novel, revolutionary and coordinated scientific approaches to hazard and risk assessment are needed. Such approaches not only need to be robust and reliable they also need to be time and cost effective whilst avoiding, wherever possible, the use and reliance on animal testing. There are times in every field of science when technological advances set the stage for progress at a pace that was previously inconceivable. For regulatory science, the time is now. The goal of this Perspectives column is to discuss how stakeholders foresee the use of OMICs data to trigger a genuine and fundamental change to redress the escalating challenges faced by industries, governments and the public in the assessment of potential health and environmental hazards imposed by thousands of un(der)tested

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	<p>chemicals. We ask: How the different stakeholders see the current use of OMICs for chemical safety assessment and what are the critical advances required so it can deliver valuable solutions to improve our confidence in chemical safety assessment and ultimately be incorporated into global regulatory frameworks?</p>



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3 **How Omics Technologies Can Enhance Chemical Safety Regulation:**
4 **Academia, Government and Industry Perspectives**
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Acronyms

RA	Risk assessment
MoA	Mode of Action
WoE	Weight of Evidence
NC3Rs	National Centre for the Replacement Refinement & Reduction of Animals in Research
QSAR	Quantitative structure–activity relationship
IATA	Integrated Approach to Testing and Assessment
TK/TD	Toxicokinetic-Toxicodynamic
CBR	Critical Body Residue
PEC	Predicted Exposure Concentration
GC-MS	Gas chromatography–mass spectrometry. An analytical method that combines the features of gas-chromatography and mass spectrometry to identify different substances within a test sample.
RNA-seq	Ribonucleic acid sequencing. A transcriptome sequencing technique used to reveal the presence and quantity of RNA molecules in a biological sample at a given moment in time.

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Lead**Bruno Campos and John k. Colbourne**

The current rapid pace of innovation of industry is delivering an ever-increasing number and diversity of chemicals within industrial and consumer products. Regulations require an evaluation of the environmental and human hazards of chemicals, ultimately characterizing the risks associated with their manufacture, use and disposal. In order for policy makers and regulators to carefully balance economic priorities with the need to protect vital ecosystems and public health, equally novel, revolutionary and coordinated scientific approaches to hazard and risk assessment are needed. Such approaches not only need to be robust and reliable they also need to be time and cost effective whilst avoiding, wherever possible, the use and reliance on animal testing. There are times in every field of science when technological advances set the stage for progress at a pace that was previously inconceivable. For regulatory science, the time is now. The goal of this Perspectives column is to discuss how stakeholders foresee the use of OMICs data to trigger a genuine and fundamental change to redress the escalating challenges faced by industries, governments and the public in the assessment of potential health and environmental hazards imposed by thousands of un(der)tested chemicals.

We ask: How the different stakeholders see the current use of OMICs for chemical safety assessment and what are the critical advances required so it can deliver valuable solutions to improve our confidence in chemical safety assessment and ultimately be incorporated into global regulatory frameworks?

an Academia perspective**James B. Brown and Mark R. Viant***State of the science*

The current level of technological maturity attained by diverse platforms like Next Generation Sequencing as well recent advances in chromatography and mass spectrometry have enabled the high-content and high-quality molecular interrogation of biological systems. The process of comprehensively measuring, or, deeply sampling a molecular landscape is known as “Omics”. Dozens of “omes” are now open to study: the epigenome (heritable modifications to DNA or chromatin that do not alter the underlying genetic sequence); the transcriptome (RNA); the proteome (proteins); the metabolome (usually defined as all small molecules present in a system). It is now possible to globally survey gene products with single-cell resolution, to simultaneously measure hundreds of proteins and thousands of metabolites, and to use time series measurements to infer the relationships between these processes, as well as with tissue and organismal phenotypes [1, 2].

‘OMICs’ have fundamentally transformed the approach used by basic scientists for investigating and characterising molecular processes in plants, animals and microbes [3]. Comprehensive information about biological systems has effectively industrialized the process of hypothesis generation: data mining procedures have leveraged advances in statistical machine learning to discover new molecular processes and pathways from the integrative analysis of multi-OMICs surveys [4]. High-profile data-driven discoveries based on the analysis of OMICs data include the role of epigenetics in transmitting the maternal “memory” of transcription to the egg and the molecular basis of over two thousand distinct genetic diseases. In principle, OMICs are therefore ideal tools for discovering key events (KEs), within Adverse Outcome Pathways (AOPs) [5]. If sufficiently reproducible, robust, and harmonized protocols can be developed for the characterization of chemical exposures in relevant and ethically credible biological and ecological systems, OMICs approaches are

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3 likely to emerge as the most plausible solution to the international chemical
4 safety crisis.
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9 Public demand for safer environments has been growing, and sophisticated
10 regulatory frameworks governing the manufacture and dispersal of chemicals
11 are now in place throughout the world. In Europe, the Registration, Evaluation,
12 Authorisation and Restriction of Chemicals (REACH, EC 1907/2006) requires
13 safety assessment and risk management for all chemicals used in quantities of
14 more than one tonne per year, and the Water Framework Directive (WFD,
15 2000/60/EC) requires assessment and management of ecological and chemical
16 water-quality for inland, transitional and coastal water bodies in Europe. In the
17 United States, The Frank R. Lautenberg Chemical Safety for the 21st Century
18 Act (H.R.2576, 114th Congress 2015-2016) has explicitly modernized toxic
19 substances testing requirements by opening the door to Integrated Approaches
20 to Testing and Assessment (IATA) in regulatory science. To support these
21 innovative legislations, novel and coordinated assessment procedures that are
22 rapid, cost effective, and consistent with ethical standards are urgently needed.
23 A proposed solution is the application of data-rich OMICs technologies in non-
24 mammalian organisms coupled with *in vitro* human-derived models to
25 systematically identify the MoA's of compounds. A collection of pioneering
26 studies have demonstrated the contribution of OMICs in a weight-of-evidence
27 approach helping the formulation of AOPs [6], and now these and related
28 studies need to be replicated to establish robust, transferable, and reliable
29 protocols for chemical safety assessment.
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43 44 45 46 *Proposed roles for OMICs technologies in chemical safety assessment* 47

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49 Accepting for a moment the premise that 'OMICs technologies can, in the near
50 term, contribute significantly to the discovery of the modes of action of
51 chemicals; we foresee three specific regulatory applications for this new
52 knowledge. **First**, KEs discovered from 'OMICs measurements and then
53 biologically validated in subsequent targeted studies could enable biologically
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3 based read-across – leading to more reliable and cost-effective prioritization of
4 chemicals for testing, as well as more accurate hazard prediction [7]. To
5 achieve this, chemicals would first be grouped according to the molecular KEs
6 that they activate, then the apical toxicity of some chemicals within a given
7 chemical group could be predicted from the known adverse outcomes of the
8 more extensively studied chemicals in that group. Read-across (of apical
9 toxicity) based upon the activation of a panel of molecular KEs exploits far
10 richer information than is presently used for quantitative structure-activity
11 relationship (QSAR) analysis. **Second**, molecularly defined AOPs could lead to
12 improved cross-species extrapolation. In this emerging field, we seek to apply
13 the principles of evolutionary biology to toxicology – to identify conserved (or
14 not) biochemical pathways that give rise to susceptibility or resiliency to
15 compounds. This is an essential step, because it could enable the translation of
16 tests conducted in tractable model organisms to the entire tree of life. **Third**,
17 and most simply, given the capacity of OMICs-enabled studies to identify KEs,
18 we expect rapid growth in the number of OECD-approved AOPs – which is
19 already apparent: at the time of writing, there were only six OECD endorsed
20 AOPs, and 113 under development. The stage is set for an exponential growth
21 in the collection of adverse outcome pathways with extensive molecular
22 characterisation in human and other metazoan systems.
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38 However, a key question remains: how predictive are the OMICs-based test
39 data of higher levels of biological organisation (the adverse outcome), including
40 growth and reproduction of organisms and population or ecosystem fitness? It is
41 increasingly clear that OMICs will radically enhance our capacity to discover
42 molecular predictors of adverse outcomes at the level of individuals [8], but
43 there is still little evidence to suggest that we can extrapolate to populations or
44 ecosystems. Transgenerational studies are beginning to show promise [9], but
45 ecological toxicology lacks the basic tool that serves as a foundation to virtually
46 all other biological science: the model system. We have model organisms,
47 model *in vitro* tissue cultures, but no model ecosystems. Mesocosms are a step
48 in the right direction, but are generally too expensive and slow to function on
49 time scales relevant to industry or government. Ecosystem models (in silico
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3 and/or in vivo) for toxicity testing as reproducible and rapid as the development
4 of the model organism *Drosophila melanogaster* or *Daphnia spp.* are needed to
5 enable this leap. Will these be freshwater microcosms? 3D-printed terrestrial
6 systems? A number of *consortia* are now racing to build such constructs
7 (<http://eco-fab.org/>) and we will be watching their progress with interest.
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11 12 13 14 *How to progress OMICs based safety assessments operationally*

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16 While many academics remain convinced of the power of ‘OMICs technologies
17 in regulatory toxicology, several challenges do remain. For example, while
18 ‘OMICs have a proven record of discovering novel molecular mechanisms of
19 disease, aging, and toxicity, a standardized design of the OMICS study
20 (particularly the design of the method performance criteria) and the
21 statistical/computational strategies for discovering molecular KEs have not yet
22 been established. The distillation of OMICs approaches to precisely defined and
23 exquisitely reproducible protocols has yet to happen, and while some cross
24 platform validation has already taken place [10], much more is needed.
25 Bioinformatics and statistical tools also need to be standardized, and entire
26 pipelines, from the lab bench to quantitative conclusions, need to be packaged
27 and made distributable. But this is no easy task – credible use of these
28 technologies will require massive efforts towards validation and standardization.
29 In the genomics community, the Encyclopaedia of DNA Elements Consortium
30 (ENCODE), a multinational, NIH-funded endeavour that has cost over a quarter
31 of a billion dollars, operated since 2003, and has been a major contributor to the
32 standardization of analytical pipelines in the genomics community
33 (<https://www.encodeproject.org/>). We need to scale up our commonly artisanal
34 projects to validate findings and procedures in “ring tests” ensuring and
35 quantifying reproducibility. At a minimum, a collection of case studies need to
36 be conducted at four or more laboratories with high biological replication, from
37 data generation to knowledge extraction. We posit that such case studies, along
38 with effective cross-disciplinary and cross-sector communication and education,
39 are the necessary foundation of future regulation and policy. More broadly, the
40 community needs to think collaboratively – and grandly – to imagine and
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3 establish ENCODE-scale Consortia suited to tackling the enormous challenges
4 presented by the science of molecular toxicology.
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9 Although substantial challenges remain, we suggest that a clear goal has come
10 into view for the future of OMICs in regulatory science: the generation of a
11 publicly owned Knowledge Base organizing and distributing data and
12 conclusions derived from molecular and OMICs studies. Here we don't refer to
13 (existing) repositories of experimental OMICS data, such as GEO or
14 MetaboLights, rather a database of molecular measurements derived from
15 standardized assays of highly informative KEs organized in a semantic way
16 enabling chemical producers to query molecular responses in model systems
17 for new compounds against a library of existing test data, and make projections
18 for the activity of the new compound across the entire tree of life – a tantalizing
19 prospect. Such a resource stands to provide enormous dividends for both public
20 and private sectors.
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29 Given the immediate legislative needs for setting regulatory priorities and the
30 economic benefits of manufacturing safer products by design, now is the time to
31 apply high-throughput multi-OMICs technologies for 3R-compliant toxicity
32 assays. The SETAC community can and should be key part of this effort, and
33 should further coordinate global efforts to achieve the objective of migrating
34 towards 21st century solutions to (eco)toxicology.
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A Government Perspective

Adam D. Biales, Kathryn Gallagher, Tala R. Henry, Keith G. Sappington

State of the science

Despite decades of toxicological research and advances in the chemical regulatory landscape throughout the world, the need for improving the efficiency and accuracy of our chemical risk assessment process has never been greater. Characterizing chemical risk is accomplished through the linkage of measured or modeled exposure and toxicity values. The current paradigm for risk estimation has limitations based on the types of tools available, as well as the availability of data to expand the existing tools and/or create new, improved tools. Traditional methods are ill equipped to robustly assess risks associated with the 140,000+ chemicals in commerce worldwide in an efficient and timely manner, let alone other perennial issues such as mixtures. It has been suggested that the various OMICs modalities, such as transcriptomics, proteomics, metabolomics and epigenomics, may be used to augment traditional methods, or be combined with other next generation technologies to address limitations and provide a better characterization of chemical risk [1, 2]. Alterations on the molecular, biochemical, and/or cellular levels of organization, typically measured using OMICs technologies often represent the initial responses of organisms to a chemical exposure and are thought to be initiating events in the pathway to adverse changes on higher biological levels. Many of the technological platforms available (e.g., microarrays, GC-MS, RNA-seq) are able to measure changes across large proportions of the total OMICs response of an organism, tissue, or cell (i.e., 100s to 1000s of genes, proteins, or metabolites). The sheer number of simultaneously measured endpoints can be applied to chemical risk assessment in any number of ways (discussed below). Lastly, OMICs-based approaches are amenable to high and medium throughput experimental formats, suggesting their utility for the rapid screening and characterization of untested chemicals.

Potential of OMICs technologies in environmental regulation

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3 The high dimensionality of OMICs data sets suggests the possibility of
4 developing OMICs-based fingerprints for a chemical or activated biological
5 pathway. These fingerprints have the potential to be applied to both exposure
6 and hazard assessment in an unsupervised or non-targeted manner to
7 simultaneously screen all activated biological pathways within in vivo or in vitro
8 systems, requiring no a priori information regarding mode of action (MOA) [3, 4].
9 Though there are few, if any examples, of these tools being used in a strict
10 regulatory framework, there are several examples demonstrating their potential
11 within the human clinical world where they have been shown to outperform gold
12 standard tests and have reached the lofty bar of FDA approval [5, 6]. The
13 ability to develop MOA-specific fingerprints makes this concept amenable to
14 discovery and characterization of multiple cellular pathways simultaneously in a
15 single experiment, as opposed to screening against a panel of focused
16 bioassays, providing needed time and resource efficiency. Throughput,
17 however, is highly dependent on the development of automated pipelines for
18 data analyses and interpretation. The ability to screen for all activated MOA
19 simultaneously has particular benefits in characterizing real-world exposures.
20 Environmental samples are often highly complex mixtures, with chemical
21 constituents that have the potential to interact, altering both toxicity and
22 exposure parameters. Mixture constituent interactions are not reflected in
23 analytical chemistry measures; however, the OMICs endpoints are effects-
24 based measures, thus they should effectively integrate potentially confounding
25 factors. Fingerprints can also be used for hazard assessment, where they can
26 be used to identify MOA of untested or uncharacterized chemicals by
27 establishing relationships among chemicals based on similarities of their OMICs
28 responses [7]. OMICs-based fingerprints can also be related to the
29 manifestation of disease states, suggesting the potential for predictive
30 measures of apical response. An extension of this is the ability to leverage
31 publicly available datasets, reducing the need for additional toxicity testing. The
32 utility of these existing datasets may be further increased by the possibility to
33 extrapolate across experimental platforms, cell lines and species, suggesting
34 the potential for rapidly increasing the coverage of the total chemical space
35 without the need for further testing [8].
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Regulatory challenges potentially addressed using OMICs technologies

The relatively high costs and lengthy timeframes associated with producing measured toxicity data puts practical limits on the number of conditions that can be reasonably tested. This subsequently introduces uncertainties when extrapolating beyond the relatively simplistic laboratory conditions. Measured toxicity data exists for only a limited number of chemicals relative to the total number of chemicals in commerce, and generally focus on the discrete parent compounds or commercial mixtures as manufactured rather than their metabolites or environmental degradation products and employ a limited set of exposure conditions. Because OMICs experimental platforms are amenable to high throughput formats (think TOXCAST®, NGS, among others), it is possible to rapidly and efficiently expand testing to cover a greater range of experimental conditions, chemical formulations or mixtures, and metabolites which should lead to reduced uncertainty in assessing chemical hazard and risk. Additionally, OMICs endpoints may be most informative when taken relatively soon after the exposure is initiated, as these responses are more readily associated with the particular chemical exposure rather than somewhat generic pathways leading to apical outcome(s). Comparatively rapid OMIC-based assays further contribute to the potential for a highly efficient and relatively inexpensive platform that would allow a greater coverage of the total chemical space and allow testing to expand beyond the parent compound and include an increased set of exposure conditions. OMICS measures taken in exposed cells, organs or organisms should capture all biological pathways that are altered by a chemical or environmental sample. Their ability to integrate across MOA suggests that they may have increased utility relative to traditional bioassays targeting single events, such as receptor binding, single gene or enzymatic activation. Many currently used bioassays focus on initial single events, such as direct binding of a ligand to its cognate receptor. As a result, they may not be able to identify indirect effects or activation of a particular MOA at a point downstream of receptor binding. Finally, as many components of biological pathways are well characterized, changes in expression of many components of a biological

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3 pathway may provide an increased weight of evidence suggesting a particular
4 MOA.
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7 The same practical realities that limit our ability to explore more varied exposure
8 scenarios, also limits the number and variety of species that we can test.
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10 Regulatory programs generally intend to be protective of the majority of species
11 likely to be exposed. Given the huge diversity of species and the fact that
12 toxicity data are lacking for most species, it can be difficult to quantify the
13 degree of uncertainty underlying the assumption. Similarly, toxicity tests
14 conducted in one life stage may not capture vulnerabilities of other life stages,
15 as exposure routes and sensitivities may differ [9, 10]. OMICs may provide a
16 means to extrapolate from model species to less characterized species.
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18 Though chemical-induced OMICs profiles may be related to MOA, they may
19 also provide insights into compensatory mechanisms (clearance, metabolism,
20 etc). Susceptibility, at least in part, may therefore be “coded for” in the
21 sequences of genes related to xenobiotic metabolism and clearance or directly
22 to the MOA [11-13]. The cost of sequencing genomes continues to decrease,
23 which is resulting in a huge increase in the number of non-model species with
24 sequence level information [14]. It may be possible to leverage sequence level
25 data and epigenetic modifications in combination with life history information
26 and phylogenetic relationships to predict sensitivities of untested organisms.
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28 Sequence data has been used to accurately predict sensitivities across species
29 [15] as has phylogeny [16].
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41 *Challenges and limitations of OMICs technologies in environmental regulation*

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43 Though OMICs show great promise in a myriad of applications related to
44 chemical risk, there are limitations that must be addressed before their use in
45 regulatory settings. The majority of OMICs-based studies are aimed at
46 discovery rather than at the development of tools useable within regulatory
47 contexts. The performance of OMICs-based tools must be characterized and
48 their applications standardized under the conditions that they will likely be
49 employed. For the application of OMICs data and OMICs-based tools to
50 emerge/evolve, it is critical for these data/tools to be put into practical and “fit-
51 for-purpose” assessment framework(s) and that they demonstrate an added
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3 benefit to the currently used measurement/assessment endpoints. Another
4 potential limitation of OMICs-based tools is the difficulty in interpreting OMICs
5 change relative to risk, as they are often reversible and are early events in the
6 pathways leading to higher order adverse outcomes. For OMICs-based
7 data/tools to be effectively used to predict adverse outcomes, it will be critical to
8 establish quantitative relationships among the events along the adverse
9 outcome pathway and with the apical endpoint of regulatory concern. These
10 tools must be honed to their intended regulatory application; however, it must
11 be recognized that OMICs-based tools may not easily lend themselves to
12 traditional means of estimating chemical risk and for them to be maximally
13 useful we must consider adapting practices to accommodate them. For
14 example, many regulatory actions rely upon quantitative concentrations as a
15 trigger for action or for protective values, yet it is not clear if OMICs endpoints
16 are amenable to a single value. Rather they may provide more semi-
17 quantitative data (high, medium or low) or even qualitative data, which, in
18 combination with other toxicity measures may be result in a greater weight of
19 evidence supporting the likelihood of an adverse outcome. How these can be
20 incorporated into environmental regulation will require close communication
21 between the regulatory community and those developing the OMICs-based
22 tools and is somewhat constrained by the statutes that the regulatory
23 community is tasked with implementing.
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Disclaimer: The views expressed in this article are those of the author(s) and do not necessarily represent the views or policies of the U.S. Environmental Protection Agency.

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An Industry Perspective

Stuart Marshall and Graham Whale

State of the science

Industry has responsibility to assure safe production, handling, use and disposal of chemicals. It does this through assessing the associated hazards and risks and by implementing relevant risk reduction measures. Companies have an ethical responsibility for this irrespective of regulatory requirements. Therefore, industry is keen to see development of science-based assessment of chemicals for both internal safety assessments and in regulatory frameworks. Hazard assessment in current regulatory frameworks was founded on testing representative or surrogate organisms in vivo. A key incentive for the development of sub-individual level assessment methods such as OMICs is the drive towards animal alternative methods, i.e. assessing hazardous properties without using protected stages of vertebrates. Depending on the level of testing and refinement needed, in vivo testing can be cost and time intensive. Alternative methods are desirable but it is important that there is confidence and reassurance that assessments are fit for purpose and that approaches can be easily communicated to relevant stakeholders.

The question of using OMICs in Risk Assessment (RA) is not new [1, 2] but developments over recent years have accelerated progress, especially in the capability for using gene expression data that cover a large proportion of cellular components to identify potential biomarkers and in advanced bioinformatics and statistical methods to discriminate control versus treated molecular responses. The availability of a limited number of fully sequenced organisms (including fish, daphnia, nematodes, slime moulds and algae), the high throughput nature of the technology and data processing capacity now bring opportunities for the rapid assessment of chemicals using several types of OMICs assays. Guidance on data collection and processing is also developing to improve the quality and robustness of OMICs screening tests (MERIT (MEtabolomics standaRds Initiative in Toxicology) www.ecetoc.org). Commercialisation of the technology is helping to reduce variability. However,

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3 even with these advances, many of the approaches are still being used as
4 research rather than RA tools. This is due to a range of factors such as lack of
5 standardisation and complexities in interpreting data in terms of representing
6 apical endpoints. Nevertheless, there is a need for lower cost, more rapid, less
7 animal intensive studies to help screen potential new products in order to
8 identify those which raise concerns and may require additional assessment.
9 Such approaches are also required to help underpin chemical grouping or
10 category approaches for UVCB substances. The Cat-App project, initiated and
11 funded by CONCAWE, is investigating practical strategies for grouping and
12 read across approaches by developing a framework based on chemical-
13 biological read-across [3]. The approach is to integrate innovations in (i) in vitro
14 testing, (ii) high-throughput genomics and (iii) integrative data analyses and
15 visualisation into a transparent workflow for read-across assessment of UVCBs
16 in regulatory programmes (<https://www.concawe.eu/cat-app/>).
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28 *Opportunities for OMICS technologies in chemical hazard and risk assessment*

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30 OMICs technology is unlikely to address the challenges of increasing the
31 environmental relevance of RA, i.e. assessing protection goals set at
32 population, community and ecosystem levels [4]. Indeed, even at the individual
33 level, more knowledge of systems biology is required to link molecular
34 responses to physiological processes at increasing levels of complexity,
35 covering all life stages. This includes the knowledge needed to interpret OMICs
36 data in terms of adaptive or adverse effects e.g. quantitatively linking up/down
37 regulation to apical endpoints via plausible biological pathways and accounting
38 for temporal changes in signals. Other influences on OMICs responses such as
39 environmental factors (water quality, temperature, food) and the genetic
40 variability of a species - both inter-individual variation in expression and
41 differences between lab and field populations, also require consideration
42 although it should be noted that this is not made in most conventional testing
43 guidelines. These confounding influences mean that OMICs assays must be
44 well standardised and validated in terms of replicability, reproducibility and
45 repeatability. Consequently, in the near to mid-term, expectations for applying
46 OMICs in RA may be tending away from the prediction of apical endpoints
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3 towards their use in identifying and understanding KE as part of an integrated
4 approach to testing and assessment (IATA).
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7 Overcoming the considerable challenges in applying OMICs to RA will only
8 occur if there is confidence in the techniques and clarity on the applicability
9 domains. Increasing confidence in RA may be achieved through a WoE
10 approach to hazard identification and characterisation e.g. using existing
11 ecotoxicity data, QSARs, chemical grouping (by structure and bioassay
12 responses), TK/TD modelling and generating new in vitro or in vivo ecotoxicity
13 data. OMICs data are starting to play a useful role in by providing supporting
14 information to help explain differences in species sensitivity, for use in
15 qualitative grouping/read-across and in the determination of MoA.
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19 Determination of MoA is an important and useful input to a RA strategy. It would
20 be beneficial to develop a library of MoA fingerprints for a range of species to
21 compare with OMICs response data to test chemical exposure. These
22 fingerprints would need to be resolved and described at a suitable level, e.g. for
23 a range of tissues, sexes, life stages and account for changes in response over
24 time. If response patterns are sufficiently similar, then it may be possible to infer
25 the same MoA. For example, if a chemical could be categorised as a baseline
26 toxicant then knowledge of the internal critical body residue (CBR), for this
27 generic MoA could be used to simplify the RA by comparing the CBR with
28 external exposure (PEC) using conservative TK models or chemical activity [5].
29 However, OMICs may be more effective in assessing chemicals with a specific
30 MoA/receptor where a characteristic OMICs response can be identified. This
31 information could add to conventional structural approaches, e.g. to assessing
32 chemicals with specific Moa's [6]. As previously discussed, these data could
33 also be used to categorise chemicals; the value of OMICs for this purpose is
34 currently being investigated by CONCAWE to assess and confirm petroleum
35 product categories under the EU REACH regulations.
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51 *Using OMICs to reduce animal use in health and environmental risk*
52 *assessments*
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3 OMICs data are also showing promise in understanding species differences in
4 sensitivity to chemicals with specific Moa's, e.g. by searching for target genes
5 across species (gene homology) [7]. Identifying conserved key genetic
6 pathways has promise in enabling non-vertebrates such as nematodes to be
7 used to provide hazard screening data for fish and mammals.
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11 In terms of models, fish are vertebrates and therefore share a high degree of
12 sequence and functional homology with mammals, including humans. Due to
13 the conservation of cell biological and developmental processes across all
14 vertebrates, studies in fish can give great insight into potential effects on higher
15 vertebrates and even human disease processes. For example, studies with
16 proteins have shown these have a similar function in fish and mammals [8]. In
17 addition to fish other alternative developmental biology model systems are
18 currently being explored, ranging from whole organism systems with nematodes
19 and slime moulds, to cell based systems using mouse or human embryonic
20 stem cells. Groups working with these systems are embracing and exploiting
21 recent developments in molecular biology and genetics to better understand the
22 underlying pathways driving normal embryogenesis and the effects of
23 perturbations to these pathways. For example, the potential of some of these
24 test systems has recently been investigated as part of an NC3Rs challenge
25 (www.crackit.org.uk/challenge-10-predart). Using research from this challenge
26 from nematode (*Caenorhabditis elegans*) and zebrafish models have been
27 shown to have potential to screen for mammalian relevant pre-DART
28 (development and reproductive toxicity) phenotypic endpoints [9]. The pre-
29 DART challenge omics results also indicated DART specific responses on RNA
30 and protein level and that 4- RNAi in *C. elegans* indicated critical genes for
31 toxicological response although these results are yet to be published. However,
32 building on aspects of this research the UK NC3Rs launched a new DART
33 paths challenge which aims to develop a cross-organism mapping strategy and
34 framework (<https://crackit.org.uk/challenge-26-dartpaths>). Industrial companies
35 with policies in place to reduce animal use are supporting these initiatives to
36 help unlock the potential of these alternative test systems. It is envisaged that
37 this will be achieved by improving the understanding of conservation of key
38 pathways across different species and identifying and linking genes with the
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3 same function and role in driving DART between organisms. Although the
4 current focus is on health endpoints these test systems have relevance for
5 ecological endpoints as they can inform on potential concerns related to longer
6 term effects of chemicals in fish as well as potential concerns related to
7 mammals. In the future, such test systems could potentially provide more
8 holistic health and environmental assessments of environmental samples (e.g.
9 in contaminated site, surface water quality and plant protection product field
10 investigations).

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18 In summary, there are a number of exciting opportunities for the application of
19 OMICs technologies within health and environmental risk assessments. They
20 offer potential to provide high throughput more holistic screening assessments
21 with reduced reliance on traditional in-vivo (i.e. using 'protected stages)
22 vertebrate models. However, the challenges to application of OMICs in such
23 assessments are broad and numerous. Overcoming these challenges will
24 require a co-ordinated effort across academic, industry and regulatory sectors
25 with the specific aim of applying/implementing the technology in chemical safety
26 and environmental risk assessments.
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