NanoImpact 9 (2018) 1–13

Contents lists available at ScienceDirect

NanoImpact

journal homepage: www.elsevier.com/locate/nanoimpact

Review article

Risk assessment frameworks for nanomaterials: Scope, link to regulations, applicability, and outline for future directions in view of needed increase in efficiency

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ARTICLE INFO

Keywords:
Strategies
Tiered testing
Vitro-vivo comparison
Bioaccumulation
Delivered dose
Read-across
Decision-making

ABSTRACT

The increasing application of nanomaterials and the notion that their distinct features compared to larger sized counterparts should be considered in safety assessment, has led to the development of risk assessment frameworks that are specific to nanomaterials. These frameworks aim to prioritise, rank or assess the safety of a nanomaterial efficiently by targeting critical information in order to conserve resources. The present overview shows that each nanomaterial framework has its own scope, advantages and disadvantages and all except one lack details such as decision criteria to come to conclusions and enable actual application. Those frameworks directed towards gaining information and making decisions on regulatory submissions at national and EU level are principally of interest. Additionally, those aimed at informing decision-making in the innovation chain are important.

This manuscript also discusses issues relevant for exposure and hazard assessment of nanomaterials such as life cycle, bioaccumulation and delivered dose that should be considered in risk assessment frameworks. Elements for improving the feasibility to perform risk assessment in practice include standardised testing, knowledge on in vitro-in vivo comparison and functional assays. With this information and the need to increase the efficiency in risk assessment, future perspectives are outlined. Grouping and read-across approaches can bring some efficiency compared to a case-by-case approach. However, science is at present not advanced enough to fully substantiate decision criteria and specific protocols needed to considerably increase the efficiency. A possible way forward would be to pursue the development of a pragmatic and internationally accepted nanomaterial decision framework with decision criteria that can only be partially scientifically based. This would require the cooperation of policy makers, scientists and industry.

1. Introduction

Nanomaterials are increasingly used as their different features, compared to their larger sized counterparts, can be applied in innovative products and materials. Such changes in functionality can be made by modifying chemical make-up, size, shape, surface characteristics *et cetera*. The physicochemical properties that provide specific functionality, can also affect the behaviour of nanomaterials in the environment and humans, which may result in different exposures (including different sites in the environment or within the human body) and subsequent hazards. It is therefore relevant to consider the potential risks of nanomaterials. This should be done in such a manner that sufficient information becomes available to assess the risk of each nanomaterial and allows innovative nanotechnologies to be developed.

The basic components of risk assessment of chemicals are hazard and exposure assessments, dose-response estimation, risk characterisation, and accounting for uncertainty in the overall assessment. While this traditional risk assessment paradigm also holds for nanomaterials (SCENIHR, 2005, 2007, 2009; Sayre and Steinhäuser, 2016; OECD, 2012a), many of the tools, test protocols and guidelines for determination and assessment of physicochemical properties, fate, exposures, and effects used for conventional chemicals, need modifications when applied to (the regulatory) evaluation of nanomaterials (Sayre and Steinhäuser, 2016).

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http://dx.doi.org/10.1016/j.impact.2017.09.001
Received 17 July 2017; Received in revised form 12 September 2017; Accepted 13 September 2017
Available online 14 September 2017

In the context of this article, the term risk assessment "framework" is used in the same context that it is used by the National Academy of Sciences (NAS, 2009): it is intended to incorporate the traditional risk assessment paradigm applied to chemicals (NAS, 1983) in broader sense to allow for a flexible risk assessment approach for decision-making. This encompasses both human health and environmental endpoints, and incorporates concepts such as the following: default assumptions, read-across, overarching general risk assessment paradigms, and specific tiered-testing schemes. Recently, more risk assessment frameworks and assessment methodologies, sometimes also referred to as tiered-testing approaches or schemes, strategies or methodologies, have emerged that are specific to nanomaterials. These frameworks aim to prioritise, rank or assess the safety of a substance/nanomaterial efficiently by targeting critical information, *i.e.* aiming to obtain the necessary information for risk assessment, while conserving resources.

The aim of the present manuscript is to assess nanomaterial testing and assessment frameworks that are most useful in a regulatory context. Those frameworks, which are mainly directed towards gaining information and making decisions on regulatory submissions at national and EU-wide levels are principally of interest. Additionally, frameworks to inform decision-making in the innovation chain are important. Several nanospecific issues in risk assessment and elements for improving the feasibility to assess the risks of nanomaterials are addressed. The frameworks are discussed in relation to the need to increase the efficiency in information gathering for risk assessment of nanomaterials. Finally, recommendations, future perspectives and conclusions are provided and discussed. These include process-related considerations on how such perspectives can be achieved.

2. Methods and criteria to select and evaluating risk assessment frameworks

As noted in Sayre et al. (2017), experts in nine different disciplines (including those with expertise in regulatory assessments, physicochemical properties, fate, effects, modelling, and risk assessment) reviewed the relevant publications and reports of 23 research and regulatory bodies from the EU, the US, the OECD, and Germany, as well as references from open literature. In total, approximately 1000 references from both the peer-reviewed and grey literature were evaluated (Steinhäuser and Sayre, 2017). All experts commented on the utility of the risk assessment frameworks, and their components, that were contained in these publications.

The overarching criteria used to select and evaluate the risk assessment frameworks, covering human health and/or environment, are those applied by the OECD to judge the utility of any regulatory method, protocol, or data set: is the risk assessment framework both relevant (to predicting endpoints of interest for regulatory purposes) and reliable (OECD, 2005)? In addition, the risk assessment frameworks and testing schemes were evaluated relative to how responsive they were to a set of regulatory questions specific to nanomaterials, as generated by regulatory programs and experts who are involved in nanomaterial regulatory risk assessments (Sayre et al., 2017). These questions were developed to determine which risk assessment frameworks were most useful for use early in the innovation process, *versus* those which could be applied at an EU or national level for regulatory decisions. Of those that could become applicable in regulatory context, focus is put on the risk assessment frameworks that are more detailed and cover a broad range of nanomaterials and exposure routes. All risk assessment frameworks would benefit from being tested for reliability in case studies. These issues were considered in Table 1, addressing the aim, regulatory readiness, advantages, and disadvantages for the various risk assessment frameworks. The obtained insights, and how the use of the frameworks can facilitate the need for increased efficiency in information gathering for risk assessment of nanomaterials constitute a different evaluation process, relative to those done in the recent past (Grieger et al., 2012; Hristozov et al., 2016). In addition, the present manuscript includes recent developments relative to the assessment of the regulatory perspective on early frameworks in Hristozov et al. (2012).

3. Overview of risk assessment frameworks

The selected risk assessment frameworks that are specific for nanomaterials are listed in Table 1. Although the frameworks are based on the same risk assessment paradigm, consisting of hazard identification, exposure assessment and risk characterisation, the frameworks are diverse in their aim, applicability domain, basic assumptions and alignment to one or more regulations. Since each framework is specific to a purpose, it is not possible to take various components from them to construct an adequate risk assessment framework to suit all routes of exposure for mammalian and ecological receptors. Almost all the frameworks lack the specific decision points and associated methods needed for decision making that are required for actual application. For the one framework that is specific enough, the decision points and associated methods cannot be fully evaluated based on current scientific knowledge. For these reasons, it is not possible to clearly indicate the best or most useful framework(s).

3.1. Scope, advantages and disadvantages

All but one of the frameworks lack the specific decision points and associated methods needed for decision making that are required for actual application. The DF4nanoGrouping framework is the only fully elaborated risk assessment framework that transparently and in detail includes clear decision criteria, triggers/cut-off values and tools to assess inhalation risks (Arts et al., 2015, 2016). The framework also has specific associated case studies (Arts et al., 2016; Landsiedel et al., 2017). Just like other frameworks, however, an independent evaluation of these criteria, triggers and methods has not yet been conducted. The properties required by regulations such as REACH do not match with the intrinsic material and system dependent properties needed by the DF4nanoGrouping framework. Therefore, while the approach is developed, detailed and includes decision criteria, allowing it for to be evaluated, the regulatory acceptability of this framework remains unclear.

The more elaborated of the risk assessment frameworks without decision criteria, are the NANoREG nanospecific approach for risk assessment described by Dekkers et al. (2016), and the NanoRiskCat by Hansen et al. (2014). These frameworks are transparent and detailed, and underpin their choices using scientific information (as far as possible) and build upon existing approaches for 'conventional' substances (*i.e.* non-nanomaterials). These frameworks consider materials and products, respectively. For screening of inhalation exposure in an occupational setting the general risk banding framework for inhalation of low aspect ratio nanoparticles by Oosterwijk et al. (2016) can be useful, whereas for environmental risks the general test strategy for assessing the risks of nanomaterials in the environment by Hund-Rinke et al. (2015) is more advanced. Further details on the different frameworks can be found in Table 1. It should be noted that these frameworks remain qualitative.

The ECHA/JRC/RIVM approach on read-across between nanoforms (ECHA/JRC/RIVM, 2016) constitutes scientifically-founded guidance aimed at gathering information for a nanomaterial on one or more hazard endpoints by using information from other materials, if possible. The ECHA/JRC/RIVM read-across approach describes steps to consider if existing information can be used in such a manner that sufficient information is available to assess the risk/safety of an unassessed nanoform, and how a read-across hypothesis can be substantiated (with existing or additional information) (ECHA/JRC/RIVM, 2016). Read-across between structurally similar substances is a generally applicable approach in regulatory risk assessment of 'conventional' substances, as

Table 1
Overview of risk assessment frameworks for nanomaterials. A distinction is made between risk assessment strategies mainly directed towards gaining information for regulatory submission, and strategies mainly directed to inform decision making in the innovation chain.

Framework; author	Type of framework and aim	Link to current regulation	Advantages and key details	Disadvantages
Risk assessment strategies mainly directed towards regulatory submission				
NanoRiskCat; Hansen et al. (2014)	Screening level assessment framework. Aim: A systematic tool that can support companies and regulators in their first tier assessment and communication on what they know about the hazard and exposure potential of consumer products containing nanomaterials. Input is given on how the potential for exposure, human health hazard and environmental hazard can be assessed.	The tool can be used for a first risk assessment of existing consumer products containing nanomaterials, *i.e.* not as such applicable to substance-based legal frameworks like REACH. Also useful for prioritisation of existing products. It gives a qualitative exposure potential for professional end-users, consumers and the environment, as well as information on the hazard potential for humans and the environment.	Descriptors for the potentials have been listed, and where possible cut-off values are proposed (mostly in analogy to non-nanomaterials). The human hazard potential is based on HARN-information (*i.e.* whether it is a nanomaterial with a high-aspect ratio), Classification & Labelling information of the bulk, and information on genotoxicity/mutagenicity, respiratory toxicity, cardiovascular toxicity, neurotoxicity, reproductive toxicity, carcinogenicity and organ accumulation. For the environment indicators are: Is the nanomaterial reported to be hazardous to environmental species? Is the nanomaterial persistent or bioaccumulative? Could use of the nanomaterial lead to potentially irreversible harm? Is the nanomaterial readily dispersed? Is the nanomaterial novel?	Qualitative output on exposure and hazard. No integration of exposure and hazard information.
DF4nanoGrouping framework; Arts et al. (2015, 2016)	Specific framework focussed on human health hazards, *via* inhalation. Aim: Efficient strategy to sort out nanomaterials that could undergo hazard assessment without further testing.	No direct link to current regulations. Proposal by a group of industries organized within the European Centre for Ecotoxicology and Toxicology Of Chemicals (ECETOC).	Clear framework with description of tools, decision criteria and specific nanomaterial cases.	Regulatory acceptability unclear. Independent evaluation of triggers and protocols used has not been conducted. Focus on local inhalation toxicity (not all endpoints).
MARINA Risk Assessment Strategy; Bos et al. (2015)	General strategy. Aim: To develop a flexible and efficient approach for data collection and risk assessment. The generated information should be sufficient to assess the risks of nanomaterials.	Generally applicable on high level. Information on physicochemical properties, exposure, toxicokinetics/fate and hazard are to be integrated. No direct link to regulations.	Generally applicable. Different potential applications of grouping and read-across within the MARINA Risk Assessment Strategy have been discussed in detail (Oomen et al., 2015).	General strategy, it provides a blueprint of a risk assessment strategy. It is not specific and not very elaborated with regard to requirements on basic information set, decision criteria, *etc.*
Nanomaterial categorization for assessing risk potential; Godwin et al. (2015)	Screening level assessment framework. Aim: Prioritise nanomaterials for further testing. To avoid 90-day inhalation studies when possible.	Prioritisation methodology to target materials of high concern that need additional scrutiny, while material categories that pose the least risk can receive expedited review.	Nanomaterials for which no exposure or potential hazard is expected require no further testing. Subsequently, hazard is investigated with alternative testing strategy assays, and the adverse outcome pathway is investigated (tier 1). If further testing is required, a short-term bolus administration *in vivo* study is performed (tier 2). If needed in tier 3 an aerosol inhalation (90 d) study is performed. Presents a case that focuses on CNTs and health risks under the Toxic Substances Control Act of USA. Provides general framework or evaluation.	General strategy without clear information on how to proceed from one tier to another. Focus on inhalation. Chance of false negatives (no further testing required whereas it in reality poses a health risk) cannot be assessed. The suitability of alternative testing strategies and of the *in vivo* short-term bolus approaches to inform on chronic toxicity is not clearly addressed.
Test strategy for assessing the risks of nanomaterials in the environment; Hund-Rinke et al. (2015)	Towards a specific framework for nanomaterials in the environment. Aim: To develop a test and risk assessment strategy for nanomaterials which specifically addresses environmental fate and effects.	Aligned to the risk quotient (PEC/PNEC) as applied in environmental regulations. The strategy is not yet sufficiently developed to fulfil the information requirements of specific legislation (*e.g.* plant protection act, biocide regulation, REACH).	Generally applicable for environmental risk assessment of nanomaterials. Bioaccumulation is taken into consideration as an alternative endpoint delivering additional information on ecotoxicity. Different stages along the life cycle of the nanomaterial(s) are considered by assessing whether there is a potential for the nanomaterial to be released into the environment. Test systems and strategies of data collection, and evaluation are provided. A screening on durability (*i.e.* the extent to which nanomaterials remain intact) in the initial compartment needs to be performed as a	Still a conceptual framework (although more concrete). Screening tests are required to identify a potentially significant effect, but no suitable screening tests have been identified. Trigger values have not yet been set.

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Table 1 (*continued*)

Framework; author	Type of framework and aim	Link to current regulation	Advantages and key details	Disadvantages
			first step. In case of medium or high durability, in tier 1 the risk quotient for environment is assessed in an initial compartment for the pristine material. In tier 2, pristine and aged nanomaterials are considered and secondary compartments are included.	
A strategy towards grouping and read-across; Sellers et al. (2015)	General strategy. Aim: To develop testing strategies for nanomaterials in order to characterise the potential risks to human health and the environment.	Comprises testing strategies for nanomaterials that are in compliance with REACH.	Generally applicable. Two hypothetical case studies.	General strategy. Life cycle changes are not considered.
Risk assessment and grouping strategy based on clouds of predefined test strategies; Walser and Studer (2015)	General strategy. Aim: To describe the need and outline of a risk assessment framework for nanomaterial identification and grouping, using 'clouds'.	Sketch of issues on nanomaterials in current regulations and general outline of potential ways to deal with those issues.	The clouds mainly relate to modes of action (MOA) and adverse outcome pathways (AOP). The need for pragmatic solutions is indicated.	Details on how clouds of predefined test strategies can be formed are lacking. Not clear how and when read-across between members of the same 'cloud' can be substantiated and performed. Limited considerations on fate and toxicokinetics.
NANoREG nanospecific approach for risk assessment; Dekkers et al. (2016)	Screening level assessment framework. Aim: To prioritise those nanomaterial applications that may lead to high risks for human health. To identify those aspects of exposure, kinetics and hazard assessment that are most likely to be influenced by the nanospecific properties of the material under assessment.	The proposed approach provides alternative ways to address the risk assessment of nanomaterials, by prioritising those applications with the highest potential health risk. Approaches for (Q)SARs, grouping and read-across are integrated.	A generally applicable approach to prioritise nanomaterials for potential risk. Provides a more concrete framework by listing and discussing which aspects of exposure, kinetics and hazard of nanomaterials are most relevant and how they can be taken into consideration using several flow charts. Provides a sorting of nanomaterials into 3 classes for further human health assessments: soluble, high aspect ratio nanomaterials, and all others for which case-by-case analysis criteria are generally described.	Can currently only be used for prioritisation and directions on type of data needed for scientific justification to perform risk assessment across different nanoforms (*e.g.* using (Q)SARs, grouping or read-across). Illustration by case studies not yet performed. Cut-off values still need to be defined.
Risk banding framework; Oosterwijk et al. (2016)	Screening level assessment framework for inhalation route. Aim: Development of a scientifically based risk banding tool by combining information on deposition of particles in the respiratory tract, lung burden and clearance, diffusion through lung mucus layer, translocation and cellular uptake and local and systemic toxicity.	Output is qualitative risk banding, which can be used to advice on risk management measures in occupational settings.	Integrating scientific knowledge to give an estimate on risk. Transparent description on the assumptions behind the framework. Available information on relationships between physicochemical properties and the processes mentioned above is used.	Qualitative output. Only for inhalation route.
NANoREG framework for the safety assessment of nanomaterials (Gottardo et al., 2017)	Review of current framework in view of nanomaterials, and outlook to screening level assessment framework. Aim: To analyse the applicability of the current EU regulatory framework to nanomaterials and to giving concrete, practical direction to industry and regulatory authorities on how to address nanomaterials in a legislative context, with focus on REACH.	To analyse the applicability of the current EU regulatory framework for nanomaterials, with focus on REACH.	Comprehensive overview of safety assessment of nanomaterials under REACH, including nanospecific considerations. Outlook scenarios are addressed, which comprise 1) the NANoREG nanospecific approach for risk assessment, as also published by Dekkers et al. (2016), and 2) safe by design, as also described in other NANoREG deliverables (*i.e.* D6.04, 2016).	No clear recommendations for nanospecific adaptations of the current regulations are provided.
Sustainable Nanotechnology Decision Support System (SUNDS); Subramanian et al. (2016)	Conceptual decision framework in which various tools and models have been integrated.	Tools (*e.g.* PROAST on dose-response) and exposure models (such as Stoffenmanager Nano and ConsExpo) are part of the overarching framework. Links to REACH have been established.	It considers links to risk governance and risk management approaches. It provides the user a broader overview.	The present assessment is on main features as the final SUNDS framework has not yet been published.
Risk assessment strategies mainly directed towards the innovation chain				
LICARA nanoSCAN; Van Harmelen et al. (2016)	Generally applicable risk comparison framework. Aim: A screening tool for SMEs that provides a	The tool is preferably to be used at an early stage in the innovation chain with the aim to	Risk and benefits are addressed in a transparent manner.	Not providing information that can be used directly in regulatory frameworks.

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Table 1 (*continued*)

Framework; author	Type of framework and aim	Link to current regulation	Advantages and key details	Disadvantages
	qualitative evaluation of the potential benefits and risks of a new or existing nano-enabled product. A comparison against a reference product or "doing nothing" is made.	facilitate the development of sustainable and competitive nano-enabled products.	Easy to use. Relevant tool to support safe innovation.	
Alternatives assessments for nanomaterials; Hjorth et al. (2016)	Alternative assessment framework. Aim: To assess the overall applicability of alternatives assessment methods for nanomaterials and to outline recommendations. Alternatives assessment for nanomaterials is complicated by the sheer number of nanomaterials possible.	Alternatives assessment is not obligatory. The decision context might allow more use of high-throughput data (comparison *vs.* assessment). It is concluded that although science may not (yet) be in the position to predict or explain nanotoxicology, science may be (more) ready for making better and safe(r) choices.	Principles for the design of safer nanotechnology (adapted from Morose, 2010) have been discussed. Examples of alternatives assessments have been discussed (mitigating oxidative stress, encapsulation, doping approaches, surface properties).	Link to innovation chain not clearly made. How the actual comparison with and decision making on alternatives is performed remains unclear.
NANoREG D6.04 (2016)	Generally applicable risk screening framework. Aim: Strategy to efficiently screen for indicators of potential risks during early stages of an innovation process. Based on six key risk potentials: solubility/dissolution rate, stability of coating, accumulation, genotoxicity, inflammation and ecotoxicity.	If the indicator for a specific 'risk potential' suggests low risk, risk assessment can be performed in the conventional, *i.e.* non-nanospecific, way. If the potential is high, nanospecific data/testing is required. Strategy is relevant for all actors along the innovation chain but mainly for academics and industry working on an innovation.	Generally applicable for nanomaterials and products containing nanomaterials at early stages of innovation. Detailed in the sense that the parameters relevant for the key risk potentials are described, and methods to investigate these parameters are listed. Parameters relevant for the key risk potentials are listed and discussed. Analytical methods suitable to investigate these parameters are also listed and referenced.	Some information on the relationship between the parameters and the risk potential is provided, but clear direction on how to assess and integrate the experimental information of the parameters is lacking.

in REACH (Regulation EC/1907/2006). The ECHA/JRC/RIVM approach on read-across between nanoforms (of the same substance) provides direction on how this concept can be applied to nanomaterials. The stepwise approach, in which a hypothesis should be built to show similarity (or be conservative) between the source and target material, is the backbone of the read-across approach. The hypothesis can be based on a kinetic argument in combination with a hazard argument. The kinetic argument shows that a similar (or smaller) amount of a target material reaches the (toxicological) target site in comparison to the source material, while the hazard argument shows that the target material is hazardous to a lesser or similar extent in comparison with the source material.

Depending on the available information of the source material and the substantiation of a read-across hypothesis, for each specific combination of endpoint and nanoform, a data gap may be filled by read-across. In that manner, the burden of testing that would otherwise be required from a regulatory point of view to provide the necessary risk assessment information for many nanoforms can be reduced. The ECHA/JRC/RIVM approach on read-across between nanoforms of the same substance outlines overarching principles on read-across to assess if it is feasible to use information from one or more (source) nanoforms, and how to substantiate such a read-across hypothesis (ECHA/JRC/RIVM, 2016). This scientific reference paper has been used by ECHA to develop guidance on grouping and read-across between nanoforms (ECHA, 2017b). Hence, read-across provides a regulatory accepted scientifically-based approach to fill an information gap on hazard, if possible, and as such is an overarching principle that can be incorporated in other risk assessment frameworks.

As indicated, the risk assessment frameworks presented in Table 1 are very heterogeneous in their aim, scope, input and output information. Hristozov et al. (2016) observed that the nanospecific frameworks tend to become more quantitative and containing higher-tier models in time. Some frameworks are directed towards risk assessment required for regulatory approval before market admittance, such as the DF4nanoGrouping framework (Arts et al., 2015, 2016) and the testing strategy for nanomaterials in the environment by Hund-Rinke et al. (2015). The LICARA nanoSCAN (Van Harmelen et al., 2016) that provides a qualitative assessment of potential benefits and risks of a new 'nanoproduct', and the strategy described in NANoREG D6.04 (NANoREG, 2016) are mainly intended to be used at an early stage in the innovation chain so as to facilitate the development of sustainable safe(r) nanoproducts. Such premarket frameworks aiming at safe(r) innovations can therefore provide a win-win situation for policy makers and industry. The flow chart by Dekkers et al. (2016) is applicable to nanomaterials that are already on the market, and facilitates prioritisation of those nanomaterials that may lead to high exposure or high toxic potential. Those elements related to exposure, toxicokinetics and hazard that are expected to be mostly influenced by nanospecific properties can also be used in safe innovation processes (*i.e.* early in the innovation chain), as well as in grouping and read-across approaches. Although there is no evidence of a hazard that is only caused by nanomaterials and not by other substances, nanomaterials can have different effects and/or potencies than the same material in a non-nanoform. Also the NanoRiskCat tool by Hansen et al. (2014) can be used for prioritising nanomaterials, though the framework is more directed towards products containing those nanomaterials. To assess the human hazard potential they consider information on the aspect ratio, Classification & Labelling information of the bulk, and existing information on genotoxicity/mutagenicity, respiratory toxicity, cardiovascular toxicity, neurotoxicity, reproductive toxicity, carcinogenicity and organ accumulation most relevant. These aspects are in part similar to the aspects considered most relevant for nanospecific human hazard by Dekkers et al. (2016) and NANoREG D6.04 (2016): solubility/dissolution rate, stability of coating/nanomaterial transformation, accumulation, genotoxicity and immunotoxicity/inflammation. The SUN Decision Support system (Subramanian et al., 2016) proposes the use of risk governance

approaches to manage nanotechnology risks and sustainability, and considers the links between these concepts.

The anticipated users of many frameworks are mostly manufacturers and developers, *i.e.* to give – in an economically efficient manner – direction to the development of safe and sustainable products or materials, and to gather the information needed for regulatory approval. The frameworks are also useful for risk assessors at governmental bodies considering the need for additional information or risk reduction measures, and to prioritise those applications and situations that need to be considered first or most. All of the frameworks, except the DF4nanoGrouping (Arts et al., 2015, 2016), are qualitative: they do not provide clear triggers, protocols, and cut-offs for hazard and risk decisions and thus require further development. Policy makers are working on implementing or improving nanospecific adaptations to current legislation or in some cases developing new pieces of legislation, for which the present frameworks can provide valuable input. For mainstream application of risk assessment frameworks by industry, regulatory acceptance is needed for their use and output.

Most frameworks focus on the hazard assessment of the pristine nanomaterial. Some frameworks such as the DF4nanoGrouping (Arts et al., 2015, 2016), the MARINA Risk Assessment framework (Bos et al., 2015), the testing strategy for nanomaterials in the environment (Hund-Rinke et al., 2015) and the ECHA/JRC/RIVM read-across approach (ECHA/JRC/RIVM, 2016) explicitly consider changes in physicochemical properties of the nanomaterials during their life cycle, but further detailed results will be needed to apply this in practice. The idea of the DF4nanoGrouping framework is to mark out the entire life cycle by determination of 'hotspots' with likely occupational or consumer exposure. The ECHA/JRC/RIVM report envisions a read-across type of hypothesis between the pristine nanomaterial and the nanomaterial as present in different life cycle stages. To address the issue of life cycle, Potthoff et al. (2015) developed a series of decision trees and flow charts to support the testing of nanomaterials, for example differentiating between relevant/probable conditions and worst-case conditions. In the case of the latter, stable dispersions of the pristine material are used. In case of relevant/probable conditions, this should be reflected in the type of test material and the potential for agglomeration. Nowack et al. (2016) developed an approach to obtain sufficient quantities of released materials to study these materials.

Some frameworks build on previously-established frameworks. This is for example the case for the DF4nanoGrouping framework, which consists of a series of publications by Arts et al. (2014, 2015, 2016) and Landsiedel et al. (2017), and builds upon previous work by Gebel et al. (2014). The ECHA/JRC/RIVM report on read-across between nanoforms (2016) relies on previous work, *e.g.* by Sellers et al. (2015) and references above, as well as on considerations on grouping and read-across approaches for risk assessment of nanomaterials as described by Oomen et al. (2015). The flow chart by Dekkers et al. (2016) and the screening strategy described in NANoREG D6.04 (2016) are both products that were coordinated by RIVM as part of the NANoREG project, and in both documents the same risk potentials or aspects of exposure, kinetics and hazard assessment that are considered most likely to be influenced by the nanospecific properties have been put forward.

3.2. Physicochemical properties used in risk assessment frameworks

Identification and characterisation of nanomaterials is considered a prerequisite first step in most risk assessment strategies. This information allows identification of existing information of those particular materials, and to target the information gathering process needed for risk assessment. Differences between frameworks can be observed with regard to the physicochemical properties that are considered. Physicochemical properties used for the most elaborated risk assessment frameworks as discussed in Section 3.1 are listed in Table 2 below. Analytical methods for physicochemical characterisation are discussed by Lowry et al., in this issue.

Some frameworks such as DF4nanoGrouping (Arts et al., 2015, 2016), Dekkers et al. (2016) and NANoREG D6.04 (2016) mention, list and/or make reference to test methods that can be used to measure the indicated key physicochemical properties or aspects considered key for risk assessment of nanomaterials. However, further guidance on how to interpret the data of such testing is lacking, except for the DF4nanoGrouping framework.

As can be seen in Table 2, although differences in information needs in the different risk assessment frameworks are visible, the chemical composition, presence of impurities, shape, particle size and surface properties are typically mentioned. Information on solubility/dissolution is also considered important.

4. Specific considerations in risk assessment

As already indicated, the current principle of risk assessment that combines information on exposure and hazard endpoints applies to nanomaterials. However, nanomaterials show particle-specific behaviour and, as a consequence, require some specific considerations. These nanospecific issues should be considered in risk assessment frameworks. Several important considerations are addressed below in Sections 4.1 and 4.2.

4.1. Nanoforms

It has been shown that different nanoforms can display different behaviour both in their fate/toxicokinetics and hazard, and thus in risk. It is therefore logical to consider this in those dossiers for regulatory acceptance that comprise more than one nanoform. A start is being made by guidance in development by ECHA on the term 'nanoform' (ECHA, 2017a). As long as different nanoforms are not addressed, this can be considered a regulatory gap which requires further research and policy considerations.

4.2. Life cycle and exposure

It is known that physicochemical properties of nanomaterials relevant for their potential risk may change during their life cycle (OECD, 2012a; Gottardo et al., 2017), whereas normally the pristine material is used for toxicity testing. Therefore, the issue of including life cycle considerations in the risk assessment of nanomaterials has been raised by many scientists and considered in general terms in risk assessment frameworks (see Section 3.1) (Bos et al., 2015; Oomen et al., 2014; Sellers et al., 2015; Arts et al., 2015; Hendren et al., 2015; Nowack et al., 2016; Erdely et al., 2016; Dekkers et al., 2016). To enable toxicity testing of released nanomaterials, standardised procedures to obtain fragmented products from nanomaterials incorporated in products are being developed (Kuhlbusch et al., in this issue). Hazard studies with well-dispersed pristine materials would probably result in most cases in an overestimation of the risk, as these well dispersed pristine materials are expected to represent a worst-case situation (Potthoff et al., 2015). In specific conditions, however, more hazardous nanomaterials may be formed during the life cycle, for example due to disintegration of a coating or photo-activation. Further investigation when these life cycle changes may lead to more hazardous nanomaterials than their pristine counterpart may be relevant. On the other hand, hazard testing of the material as representative for the realistic situation rather than the well-dispersed pristine material would result in more realistic risk assessment. This would require additional work, *i.e.* to obtain the relevant material for the (potentially many) relevant situation(s) for human health and the environment, though from a scientific point of view this would result in the scientifically most correct risk characterisation.

In order to consider changes of the nanomaterial during the life cycle in the risk assessment, it must be possible to measure the nanomaterials and their physicochemical properties in complex matrices. Quantitative measurement of concentrations of nanomaterials at

Table 2
Overview of physicochemical properties that are considered key in several risk assessment frameworks for nanomaterials.

DF4nanoGrouping, (Arts et al., 2014, 2015, 2016)[a,b]	ECHA/JRC/RIVM Read-across (2016) and MARINA RA Strategy (Oomen et al., 2015)[c]	NANoREG D 6.04 (2016)[d] Strategy to efficiently screen for potentials during early stages of an innovation process	NanoRiskCat (Hansen et al., 2014)	Risk banding framework for inhaled low aspect ratio nanoparticles, Oosterwijk et al. (2016)[e]	Test strategy for assessing the risks of nanomaterials in the environment Hund-Rinke et al. (2015)
Chemical composition Crystallinity	Chemical composition (including crystal structure and crystalline phase) Impurities	Composition (including crystal structure and crystalline phase) Impurities	It should be assessed if there is a bulk form of the material		Physicochemical properties not specified, noted that physicochemical information on the pristine material is needed
Shape and aspect ratio (including rigidity)	Shape	Shape	It should be determined if the material is a HARN (high-aspect ratio nanomaterial)		
	Particle size	Particle size (distribution) Agglomeration, aggregation	It should be determined if the material is a nanomaterial	Size and size distribution(s)	
	Surface area (including porosity)	Surface area			
Surface reactivity Surface composition	Surface characteristics (including coating chemistry, functionalisation (e.g. capping agents), surface charge (e.g. zeta potential)).	Surface charge, surface coating, hydrophobicity, lipophilicity Degree of coating		Zeta potential (or net charge at a specific pH) Surface hydrophobicity or hydrophilicity	Information on modification of the coating during testing
Water solubility, dissolution in relevant media, dispersibility		Solubility/dissolution, acid dissociation		Solubility at a specific pH	Dissolution rate to assess 'durability'
		Reactivity, photoreactivity		Conduction band energy (for metals, metal oxides, quantum dots, etc.)	

[a] Also information from functional assays is considered key.
[b] Information from all tiers (that may not be applicable in all cases).
[c] The listed physicochemical properties are also considered essential for nanomaterial identification for read-across between nanoforms (ECHA/JRC/RIVM., 2016), and for risk assessment, read-across and grouping (Oomen et al., 2015). Also information on properties related to 'where they go' and 'what they do' (such as reactivity) is needed for read-across substantiation. Some of these properties can again be linked to physicochemical properties as listed by NANoREG D 6.04 (2016).
[d] The table shows the key physicochemical properties related to the risk potentials for safety screening of nanomaterials. In addition to the physicochemical properties also coating stability, hydrolytic stability, exposure route, dose, protein binding, clearance, cell uptake, cytotoxicity, generation of reactive oxygen species (ROS), cytokine induction, test medium are mentioned.
[e] The risk banding framework by Oosterwijk et al. (2016) is only applicable for inhaled low aspect ratio nanoparticles.

realistic conditions is very complicated, expensive and time consuming. This is especially the case if also physicochemical properties, such as degree of agglomeration, size distribution, *etc.*, are to be measured under realistic conditions in complex matrices. Development of computational models to estimate such exposures and changes in physicochemical properties during the life cycle is therefore highly recommended.

4.3. Delivered dose

Attention should be paid to the tendency of nanomaterials to agglomerate, as that can have serious impact on factors like dilution, internalised dose, and application of assessment factors (Lützhøft et al., 2016; van Kesteren et al., 2014; Holden et al., 2016). High dose studies can give results not representative for lower concentrations due to agglomeration and subsequent lower biological uptake, biological barrier penetration, or sedimentation affecting the internal exposure (van Kesteren et al., 2014; Holden et al., 2016), or in cases where effect mechanisms do not scale with concentrations (Holden et al., 2016). As a consequence, the degree of agglomeration at the concentration that is identified as a No Observed Effect Concentration (NOEC) may be higher than the safe concentration that is derived from the NOEC after application of assessment factors (Lützhøft et al., 2016). Also flotation on the (water) surface may affect the degree of uptake and thus the internalised dose (Hund-Rinke et al., 2015). Hence, the internalised and nominal dose may thus not be related (Holden et al., 2016; Lützhøft et al., 2016; OECD, 2012a). In other words, if the results of a high dose study are used to set a (no) effect concentration, which is subsequently used to estimate a safe concentration by applying assessment factors, the risk may be "underpredicted" (van Kesteren et al., 2014; Lützhøft et al., 2016), *i.e.* such an estimated safe concentration may not be safe. It is therefore recommended to determine the form (physicochemical properties) and concentrations of nanomaterials in the exposure medium during and after testing (Holden et al., 2016), and to determine representative internal concentrations (such as in liver and spleen or environmental test organisms) in animal toxicity studies to obtain insight into the amount of nanomaterial actually taken up in time (and thus in the internalised dose at the target site). Examples of risk assessment based on internal concentration are provided in van Kesteren et al. (2014) and Heringa et al. (2016).

Information on internalised dose is thus considered highly relevant, though still technically challenging. For the time being, making such information mandatory may thus be difficult, but standard assessment in research projects (including EU projects) can be considered. For *in vitro*, a recent paper by DeLoid et al. (2017) proposes a model that can estimate the nanomaterial dose that is delivered to cells over the course of an *in vitro* exposure study based on limited input data specific to the *in vitro* test situation. It should be considered that determination of the internalised dose in *in vivo* studies would in many cases require additional animals. Nevertheless, such information is also useful to correlate *in vitro* and *in vivo* data in order to find ways forward for alternative testing (Oberdörster et al., in this issue, and Rothen-Rutishauser and Drasler, in this issue), which may save animals in the long run.

4.4. Bioaccumulation

It is considered that bioaccumulation is a process delivering additional information for risk assessment of nanomaterials on ecotoxicity (Hund-Rinke et al., 2015; Bos et al., 2015) and human health (Bos et al., 2015; Dekkers et al., 2016; NANoREG D6.04, 2016; Hansen et al., 2014; Arts et al., 2015, 2016) that is particularly important for nanomaterials as their elimination from tissues can be very slow (*i.e.* years). It should be noted that bioaccumulation is also relevant for hydrophobic non-nanomaterials, where the octanol-water partition coefficient (K_{ow}) provides in a suitable functional assay. However, the tissues where accumulation of such hydrophobic non-nanosubstances occur and basic mechanisms of uptake and elimination differ from those of nanomaterials. In fact, the behaviour of nanomaterials cannot be described by chemical equilibrium. As the octanol-water partitioning coefficient assumes an equilibrium between the dissolved concentrations in the octanol and water phases, it is not a predictor for bioaccumulation or environmental fate of nanomaterials (see Baun and Rose, in this issue, and Nowack et al., in this issue).

For environmental risk assessment of nanomaterials, a pragmatic screening procedure may be to determine the nanomaterial concentration in suitable test organisms (Hund-Rinke et al., 2015). For human health risk assessment of nanomaterials, information on dissolution rate in physiologically relevant media such as lysosomal fluid may give insight in the potential for accumulation. A more comprehensive view can be obtained by measuring internal tissue concentrations (*i.e.* internalised dose) in toxicity studies at a few key time points, and/or including measuring internal tissue concentrations before and after a clearance period. Such data can be used to better address the uptake (see delivered dose (Section 4.3)) and elimination and thus the potential for accumulation. Linking toxicokinetic data, including information on accumulation and elimination, to physicochemical properties of the nanomaterial and to their dissolution rate in physiologically relevant media may make this issue easier to handle in the future.

4.5. Assessment factors

The use of assessment factors for *e.g.* interspecies and intraspecies differences is common practice in risk assessment of chemicals, although also for chemicals a solid scientific rationale for the magnitude of the factors is mostly lacking. It remains unclear if these standard assessment factors for chemicals are also applicable for nanomaterials. The OECD (2012b, 2015) indicates that there are insufficient data to verify if the present standard assessment factors (AFs) of 10 for both interspecies and intraspecies differences are applicable to nanomaterials. Indeed, insufficient studies are available that can be directly compared to each other to allow assessment of the variation between studies. Toxicity information for non-nanosubstances has accumulated over decades and has a history-of-use, whereas for nanomaterials, acquisition of data, and especially of good-quality data, is increasingly emerging in the past few years. Furthermore, the variation in physicochemical properties of a nanomaterial of the same elemental composition hampers direct comparison between studies: is a different response the result of the intra- or interspecies variability, or due to differences in the nanomaterial itself? To the best of our knowledge, there are at present no datasets available that allow an adequate assessment. Hence, for the time being it cannot be assessed whether nanomaterial-specific assessment factors are needed, and if so, what factor that would be.

However, for the application of AFs for nanomaterials, pragmatic choices could be made that are based on existing data, inherent uncertainties of this data and extent of extrapolation, and consequent incorporation of sufficient margins of safety (Dourson and Stara, 1983). As there is no clear evidence that the current AFs for non-nanomaterials are inappropriate, it could be an option to use the present standard AFs until science allows for a better assessment. Alternatively, a more conservative approach could be followed, in which an extra AF for nanomaterials is introduced, in which case a discussion and decision making on limited scientific evidence is needed on what specific factor is appropriate.

4.6. Route-to-route extrapolation

In human health risk assessment, route-to-route extrapolation of information occurs on a regular basis. However, there are few data to support route-to-route extrapolation for nanomaterials (Gottardo et al., 2017). The media for exposure *via* lung (air), gastrointestinal tract (gastrointestinal fluids), skin, and intravenous application typically

differ greatly. These media as well as the conditions at the site of contact may affect the properties of a nanomaterial considerably, and thus their behaviour and risk. Further insight into the possibilities for extrapolation of hazard information that is gained from studies with different exposure routes is therefore recommended.

4.7. Functional assays in risk assessment frameworks

Because the understanding of the relationship between physicochemical properties of nanomaterials and their toxicokinetic/fate behaviour and hazard potential still is limited, several functional assays have been suggested in literature as additional tests and triggers for use in nanomaterial risk assessment. Such functional assays give a read-out depending on the combination of a nanomaterial and specific – external – conditions. Functional assays properties include the *surface affinity* (also referred to as 'stickiness') as suggested by Hendren et al. (2015) and the *dissolution rate* under relevant environmental or physiological conditions for characterising nanomaterial behaviour in a variety of important systems. Hendren et al. (2015) proposes functional assays to measure nanomaterial behaviour in environmentally relevant systems, as current approaches to predict risk directly from intrinsic nanomaterial properties are problematic. The outcome of such a functional assay could be relevant for assessing the most relevant environmental fate routes and highlight needs for associated risk assessment endpoints and could thus be integrated to improve the efficiency of risk assessment frameworks for nanomaterials. Surface affinity (α) describes the probability of particle attachment when particles collide with another particle or a stationary "collector" surface. It determines the mobility of nanomaterials (and other small particles) in environmental matrices such as porous media, and relates to their propensity to (hetero)aggregate and, in some cases, the reactivity of nanoparticle aggregates. Surface affinity measures of nanomaterials may therefore have implications for both the fate of a nanomaterial in the environment, and its potential toxicity/ecotoxicity. It is suggested that functional assays can support near-term regulatory guidance and sustainable product development. Yet, as for other tools and methods that aim to become regulatory accepted, this would require consensus on the application of the functional assay, including how to standardise the assay, and how to interpret the assay results in a regulatory context. This would imply a major change in the current practice of regulatory frameworks and scientific and regulatory efforts to come to decisions on the use and application of such functional assays. If clear relationships between output of a functional assay and a risk related parameter can be made, functional assays such as for surface affinity can indeed provide added value in risk assessment. For example, in regulatory environmental risk assessment of non-nanosubstances, the octanol-water partitioning coefficient is a convenient functional assay for likelihood of soil adsorption and bioaccumulation of that substance.

Other functional assays that are considered promising for nanomaterials are *dissolution rate* in relevant media, and surface reactivity (Lowry et al., in this issue). Dissolution rate describes the speed at which the nanoparticles form a solution in a given solvent, and may be critical for predicting both the persistence, toxicokinetics/fate of and hazard posed by nanomaterials. Dissolution rate in physiologically relevant conditions is also proposed and applied in the DF4nanoGrouping risk framework and NANoREG nanospecific approach for risk assessment by Dekkers et al. (2016). Fast dissolution in physiologically or environmentally relevant media in view of physiologically and environmentally relevant time frames is considered to provide options for read-across to the solute at an early stage of the risk assessment process as proposed by Oomen et al. (2015). When the nanomaterial dissolves only partially within an environmentally or physiologically relevant time-frame, things become more complicated with behaviours similar to mixture toxicity. In a review of available data and knowledge gaps for nano-silver by Wijnhoven et al. (2009), it was suggested that the toxic effects of nano-silver can be due to a combination of the specific properties of silver nanoparticles and the generation of ions from them. Determining whether future research would need to focus on nano-silver particles only, on silver ions only, or both, would be key. Additional efforts would be needed for a thorough risk assessment, especially in the case that both the ion and the particulate form need to be considered, or similarly, in case a coating is degraded and both the core nanomaterial and the nanomaterial with a coating need to be considered. Dissolution and agglomeration are in the OECD validation process as TGs, containing initial considerations on the interpretation of the assay results for regulation. Yet, before application in regulatory frameworks can be considered, currently available functional assays appear to require better understanding and validation.

Dekkers et al. (2016) and NANoREG D6.04 (2016) consider dissolution as one of the six aspects that are most relevant for nanomaterial exposure, kinetics and/or hazard. Hund-Rinke et al. (2015) suggests a screening on the *durability* of a nanomaterial, referring to the extent that nanomaterials remain intact. Durability by Hund-Rinke et al. (2016) is exemplified by rapid dissolution, indicating that dissolution and durability are related. Taken together, dissolution rate under relevant conditions can provide unifying information that is relevant for regulation. However, similar to surface affinity, further insights are needed on the relevant media and conditions, as well as information on the reproducibility and reliability, and the interpretation of the outcome.

Surface reactivity is considered a potential unifying factor for a mode-of-action of nanomaterials (Delaval et al., 2017; Gandon et al., 2017). Surface reactivity can *e.g.* induce oxidative stress, which may result in inflammation related effects. Surface reactivity is considered a key descriptor for hazard in grouping and read-across approaches (ECHA/JRC/RIVM, 2016; Oomen et al., 2015 – see Lowry et al., in this issue and Oberdörster et al., in this issue on generation of reaction oxygen species ROS). Similar to other functional assays, assays on surface reactivity would require better understanding and validation before application in regulatory frameworks can be considered.

The relevance and need for developing internationally accepted guidance and test guidelines especially for surface affinity, dissolution in relevant media, and surface reactivity is clear. These assays should be suitably placed in risk assessment frameworks to make use of their potential to address nanospecific behaviour and hazard. It is also essential to develop guidance on the interpretation of the outcome of such tests.

4.8. Case studies

Some publications are available on the risk assessment of specific nanomaterials incorporated in products (Voelker et al., 2015; Dekkers et al., 2013; van Kesteren et al., 2014; Heringa et al., 2016; Wohlleben et al., 2011). This case-by-case approach allows for detailed adjustments relevant to the specific case, and provides food-for-thought by pointing towards important elements to be considered in the risk assessment of nanomaterials.

5. Elements for improving the feasibility to assess the risk of nanomaterials

The present section addresses issues to increase the feasibility to perform risk assessment of nanomaterials in practice.

5.1. Standardised testing

As can be observed in the other manuscripts of this Special Issue, the knowledge to correctly apply and/or modify tools, tests and methods to obtain information relevant for risk assessment of nanomaterials is increasing. Interferences, artefacts and unstable or unknown exposure conditions are better understood and dealt with, or alternatives are (to be) sought. To bring this knowledge into practice, protocols, guidance

and suitable controls and reference materials are being developed, and standardisation activities are ongoing or can be launched. Internationally acknowledged guidance documents to standardise testing of nanomaterials are therefore considered highly necessary for risk assessment frameworks. For example, the OECD Guidance on Sample Preparation and Dosimetry (OECD, 2012b) is helpful to consider when conducting both health and ecotoxicity testing. The OECD Guidance document on Aquatic and Sediment Toxicological Testing of Nanomaterials (OECD, 2017), the OECD Fate Decision Trees (OECD, 2014) and the OECD Guidance document on Fish Dietary Accumulation Studies for Engineered Nanomaterials (OECD, 2015) – some still to be finalised – are useful for prioritising testing needs. Worldwide harmonisation of such testing approaches *via* the OECD is considered valuable in general due to their consensus-driven review and acceptance by regulatory authorities from many countries.

Standardisation of test protocols and assurance of high quality data (including sufficient quality controls) is of the utmost relevance for acquiring reliable and reproducible data that can be used in risk assessment of nanomaterials in general.

5.2. Development of in vitro-in vivo comparison

Scientific understanding to enable linking *in vitro* and *in vivo* tests is required to rely more on *in vitro* results in risk assessment. The risk assessment frameworks reviewed here do not suggest any unifying dose metric for linking *in vitro* and *in vivo* tests. Information on the dose reaching the target site (the internalised dose) both in *in vitro* and *in vivo* situations would probably help when comparing the results, as the fraction of nanomaterial reaching the target site is highly influenced by the experimental setting, both in *in vitro* and *in vivo* testing. Based on such comparisons, further considerations may then help to link *in vitro* and *in vivo* tests and to assess the applicability of *in vitro* data for risk assessment purposes of nanomaterials.

Also, information on the internalised dose, *i.e.* the amount of nanomaterials reaching the target site (*i.e.* environmental organism, cells in *in vitro* test systems, organs in test animals, *etc.*) is highly relevant for correct interpretation of the experimental results.

In vitro studies can be very useful for hazard and biokinetic assessment, though direct correlation of the *in vitro* studies for environmental or human health risk is still not yet possible. As a start, Landsiedel et al. (2014) compared data form *in vitro* assays to *in vivo* instillation and inhalation data. In the short term, *in vitro* studies can be expected to become useful for mechanistic information, *i.e.* to assess which mode of action (MOA) is expected to be most relevant for a specific nanomaterial. In addition, ranking hazard potency and biokinetic behaviour (*i.e.* cellular uptake, fate, and translocation across barriers) by *in vitro* studies is likely to provide relevant information for regulatory risk assessment. This is especially the case when such *in vitro* information can be compared to information from reference nanomaterials. Finally, test batteries which comprise different organ systems such as described by Farcal et al. (2015) may be used as a general screening tool for nanomaterials, though further comparison of the predictability of such *in vitro* test batteries to the *in vivo* situations would be needed. Further discussion on *in vitro* and *in vivo* tools and approaches for nanomaterials are provided in Oberdörster et al., in this issue, and Rothen-Rutishauser and Drasler, in this issue. Continuation of these comparison and standardisation activities is thus highly recommended.

5.3. Benchmark materials

Nanomaterials with good-quality physicochemical characterisation and exposure and/or (chronic) hazard studies are needed as standard and to assess the reliability and reproducibility of other tests.

5.4. In silico approaches

In silico approaches may help to target testing of nanomaterials. The present *in silico* approaches mainly relate to straightforward *in vitro* data, as only on such endpoints sufficient data are available, and have therefore limited accuracy for *in vivo* situations. The limited amount of available data also limits the applicability domain of available *in silico* approaches. Therefore, the current added value of *in silico* approaches is limited, although it is expected to increase in the future. Further assessment on *in silico* approaches is provided in Burello et al., in this issue.

6. Efficiency and uncertainty in risk assessment frameworks

A range of different risk assessment frameworks aiming to deal with the complexity of risk assessment of nanomaterials have been described in the literature (see Table 1). Most of the risk assessment frameworks lack details regarding decision criteria and tools to allow actual application. All frameworks struggle with how to deal efficiently with the multitude of potentially different nanomaterials due to (slightly) varying physicochemical properties. When potential consequences for human health and for the environment due to this multitude of varying properties are more thoroughly addressed (*e.g.* by gathering/generating more data), the efficiency of the framework decreases. It is anticipated that a major hurdle in constructing and implementing efficient risk assessment frameworks in product and substance/material regulations, will be the choices/decisions to be made that result in uncertainty in the number of false negatives. In the case of a false negative, a nanomaterial is allowed on the market where it poses an unacceptable risk. For example, if a cut-off value for dissolution rate is proposed, this value is either highly conservative (only the immediately dissolving materials can be waived to their solutes, meaning that many nanomaterials still remain to be assessed), or uncertainty arises from the consequences of a more pragmatic choice. Science is at present not sufficiently advanced to give a quantitative estimation of the chance of false negatives. This uncertainty is biggest for potential effects after chronic exposure. As this kind of information (mostly from long-term animal studies) is ethically debatable and expensive, knowledge can be expected to progress slowly here (Gottardo et al., 2017). Yet, it has been shown that nanoparticles can accumulate in tissues/organisms over time, indicating that with increasing tissue/organism loading in time also the likelihood of adverse effects increases. This means that it is important to investigate whether effects occur after long-term exposure. Also potential immunotoxicity and neurotoxicity have been linked to nanomaterials, though are difficult to measure and data still mainly comes from long-term *in vivo* tests. Taken together, these considerations show a major challenge for risk assessment of nanomaterials: the urgent need for efficient risk assessment with a focus on potential chronic effects, limited availability of existing, good-quality information, and high cost and time efforts needed to increase this information. From a policy point of view, the efficiency of risk assessment of large numbers of nanomaterials can be increased with practical, only partially scientifically underpinned choices. Alternatively, read-across, prioritisation and comparative techniques can bring some efficiency, but would clearly not solve the entire issue, especially not for the near future, considering the required substantiation of grouping and read-across and the limited available information. These options are further developed under future perspectives in this chapter.

A way to shift the issue of uncertainty on false negatives in risk assessment frameworks is prioritising those nanomaterial-applications with the highest potential risk, as proposed by Dekkers et al. (2016). Also frameworks that aim at considerations on human health or environmental risk early on in the innovation chain can be based on comparisons or risk indications rather than quantitative risk assessment. Although highly useful also in a regulatory sense, this does not allow for efficiently fulfilling the regulatory requirements for

legislation, *e.g.* REACH. To enable decisions to be made about efficient risk assessment frameworks in the near future, or application of (parts of) risk assessment frameworks in existing regulations, international agreement is required on practical cut-off and trigger values, while realising that these can only be scientifically founded to a limited extent. This would require dialogue between stakeholders, in which the uncertainty associated with the state-of-the-art science is addressed, made transparent and dealt with. In order to make such a dialogue a success, all stakeholders should be constructive and address and move beyond underlying issues such as the lack of information on nanomaterials in the current regulatory frameworks and the fear of extensive additional testing. Communication to the public at large that "zero risk" does not exist is essential to provide fair expectations on risk assessment.

If no action is taken, either innovation can be substantially hampered due to the need for a huge number of case-by-case risk assessments, or large numbers of nanomaterials reach the market without or with only limited knowledge on their potential risk. Hence, international agreement to come to efficient risk assessment of nanomaterials is recommended. This should take science into account as far as possible, while acknowledging that scientific insights are still premature for complete substantiation. Other risk assessment approaches that focus on prioritisation, substitution and on safety considerations during the innovation chain are relevant, and it is recommended that efforts in these fields continue. Further future perspectives are provided below.

It should be noted that also false positives are unwanted. False positives in the present setting are nanomaterials/products that are not allowed to the market because of assumed unacceptable risk, where in fact the risk is acceptable. This would hamper innovation.

7. Future perspectives to increase efficiency in risk assessment of nanomaterials

A key issue in acceptance and application of risk assessment strategies in a regulatory setting is that adequate information for safety assessment of nanomaterials is obtained.

Outside the regulatory legal framework arena, addressing human or environmental risk early on in the innovation chain is relevant to reduce the number of nanomaterials with a safety issue and avoid investment in further product development which will eventually not yield a marketable product due to high risk for humans and/or the environment. This can be a win-win situation for innovators and regulators, as both benefit from reduced uncertainty on risk.

With regard to nanomaterials that reach regulatory evaluation, two broad options can be distinguished to increasing the efficiency in gathering information for risk assessment.

7.1. Option 1

Continue in line with current regulation (*e.g.* REACH). For nanomaterials some increase in efficiency is possible by grouping and read-across approaches according to the outline described in the scientific reference paper by ECHA/JRC/RIVM (2016). This document is currently the most aligned with REACH. Based on this document further guidance has been developed (ECHA, 2017b).

Parallel to this development, a practical approach would be possible for existing nanomaterials that are already on the market. A multi-dimensional map of all forms within one substance differentiating in key physicochemical properties can be made. The nanomaterials that represent the most extreme key physicochemical properties, *i.e.* the lowest and highest value of a range of *e.g.* size, or the worst case forms if identifiable with reliable scientific substantiation, can be used to obtain hazard information that may be used for a group of nanoforms.

7.2. Option 2

Stratify the information needs according to the anticipated potential for hazard/risk in order to focus more on nanomaterials with the greatest potential for hazard/risk. To further increase the efficiency in addressing nanomaterials in risk assessment, pragmatic, partially scientifically underpinned choices for decision criteria (cut-off values, trigger values, *in vitro* tests, functional assays and other tools like high throughput systems (HTS) and - in future - *in silico* approaches, *etc.*) would be applied to risk assessments of nanomaterials. Such choices can only be partially scientifically based, as science is not sufficiently advanced and is not expected to provide well-founded science-based mechanisms to efficiently deal with risks in the near future, especially where chronic exposure is relevant. Such an approach would require cooperation of policy makers, scientists and industry and agreement on an international level. Attention should be paid to the operational process in such cooperation, for example by starting with a dialogue. If such an approach would be pursued, aspects most likely to be influenced by nanospecific properties can be used as a starting point. Several studies indicate that exposure, solubility/dissolution rate, stability of coating, accumulation, genotoxicity, inflammation, ecotoxicity and environmental fate are important nanospecific properties (Dekkers et al., 2016; NANoREG D6.04, 2016, and to some extent also Hansen et al., 2014; Arts et al., 2015, 2016; Hund-Rinke et al., 2015, and Oosterwijk et al., 2016).

Also, the decision criteria and tools as proposed by the DF4Nano Framework (Arts et al., 2015, 2016) can be independently investigated on their methodology and the likelihood of giving false negative outcomes. The outcome of the case studies as performed by the authors of the DF4NanoFramework should be included in this process (Arts et al., 2016).

Another tool to address uncertainties in such analyses is to apply additional risk management controls such as workplace engineering controls, protective equipment and restricted environmental releases until additional data are provided.

For nanomaterials already on the market, aspects of option 2 can be applied as a start. The thus obtained insights can be used in the first place to regulate the most hazardous nanoforms according to the current understanding. Subsequently, a more generic understanding on the behaviour of nanomaterials can grow continuously. This will add to a knowledge base to allow for the improved applicability of grouping and read-across of nanomaterials in regulatory frameworks, and to the feasibility of pragmatic, partially scientifically underpinned choices for decision criteria, tests and tools.

It should be noted that the present assessment is based on scientific knowledge that is gained from 'the first generation' of nanomaterials, which comprise relatively simple inorganic and carbon-based nanomaterials. It is recommended to actively monitor scientific developments of future innovations and assess potential consequences for human and environmental risk, and determine whether the legal frameworks are sufficiently equipped to deal with these innovations.

8. Conclusions

In conclusion, ongoing efforts in improving the current risk assessment frameworks to include nanomaterials need to be continued. With nanomaterials, some issues such as exposure (stable dispersions, relevance of high dose studies, assessment of internalised dose), bioaccumulation, *in vitro-in vivo* comparison and long-term effects deserve special attention. Furthermore, activities to standardise and develop protocols and guidance documents, including for several functional assays, as well as suitable controls and reference materials should continue.

Risk assessment frameworks that aim to prioritise or substitute nanomaterials of highest concern, as well as frameworks that aim to introduce safety considerations into the innovation chain, are gradually

becoming more concrete. Further evaluation of such frameworks *via* case studies and success stories are needed to improve these frameworks and pave the way for mainstream application, also for regulatory needs. To include nanomaterials (or nanoforms) in risk assessment regulations in an efficient manner, *i.e.* beyond the case-by-case approach, consensus is needed with regard to *e.g.* cut-off values and benchmark materials as well as suitability of simple tools and tests for which the present knowledge base is insufficient. Grouping and read-across approaches can bring some efficiency to the case-by-case assessment. Considering the required scientific substantiation of grouping and read-across and current limitations in available data and knowledge on relationships between physicochemical properties and nanomaterial behaviour, however, these approaches are also not expected to bring the required efficiency in risk assessment of nanomaterials for the near future. Nevertheless, developments in grouping and read-across may accelerate the assessment of existing relevant information and gaining of new information. In addition to pragmatic decisions to deal with nanomaterials in the current situation, further research is needed to increase the scientific knowledge on the risks of nanomaterials. The focus should be on systematic studies that facilitate the understanding of the behaviour of nanomaterials, especially related to long-term effects. In such studies information on internal concentrations (also referred to as internalised dose) would have added value. Such information would be useful to substantiate or adapt cut-off values and other decisions in risk assessment frameworks, and facilitate in grouping and read-across approaches.

Finally, attention should be paid to organising the operational process to get to efficient risk assessment approaches for nanomaterials, including cooperation between parties to come to harmonised approaches and investing in curation processes to acquire a reliable set of data on nanomaterials.

Conflict of interest

The authors declare that they have no competing interests.

Acknowledgement

This research was funded by the EU H2020 project ProSafe (promoting the implementation of Safe by Design), grant agreement 646325. The other members of the ProSafe Task Force are acknowledged for in-depth and fruitful discussions resulting in better integration of different disciplines.

References

Arts, J.H., Hadi, M., Keene, A.M., Kreiling, R., Lyon, D., Maier, M., Michel, K., Petry, T., Sauer, U.G., Warheit, D., Wiench, K., Landsiedel, R., 2014. A critical appraisal of existing concepts for the grouping of nanomaterials. Regul. Toxicol. Pharmacol. 70, 492–506.

Arts, J.H., Hadi, M., Irfan, M.A., Keene, A.M., Kreiling, R., Lyon, D., Maier, M., Michel, K., Petry, T., Sauer, U.G., Warheit, D., Wiench, K., Wohlleben, W., Landsiedel, R., 2015. A decision-making framework for the grouping and testing of nanomaterials (DF4nanoGrouping). Regul. Toxicol. Pharmacol. 71, S1–27.

Arts, J.H., Irfan, M.A., Keene, A.M., Kreiling, R., Lyon, D., Maier, M., Michel, K., Neubauer, N., Petry, T., Sauer, U.G., Warheit, D., Wiench, K., Wohlleben, W., Landsiedel, R., 2016. Case studies putting the decision-making framework for the grouping and testing of nanomaterials (DF4nanoGrouping) into practice. Regul. Toxicol. Pharmacol. 76, 234–261.

Bos, P.M., Gottardo, S., Scott-Fordsmand, J.J., van Tongeren, M., Semenzin, E., Fernandes, T.F., Hristozov, D., Hund-Rinke, K., Hunt, N., Irfan, M.A., Landsiedel, R., Peijnenburg, W.J., Sanchez Jimenez, A., van Kesteren, P.C., Oomen, A.G., 2015. The MARINA risk assessment strategy: a flexible strategy for efficient information collection and risk assessment of nanomaterials. Int. J. Environ. Res. Public Health 12, 15007–15021.

Dekkers, S., Bouwmeester, H., Bos, P.M., Peters, R.J., Rietveld, A.G., Oomen, A.G., 2013. Knowledge gaps in risk assessment of nanosilica in food: evaluation of the dissolution and toxicity of different forms of silica. Nanotoxicology 7 (4), 367–377. http://dx.doi.org/10.3109/17435390.2012.662250.

Dekkers, S., Oomen, A.G., Bleeker, E.A., Vandebriel, R.J., Micheletti, C., Cabellos, J., Janer, G., Fuentes, N., Vázquez-Campos, S., Borges, T., João Silva, M., Prina-Mello, A., Movia, D., Nesslany, F., Ribeiro, A.R., Emílio Leite, P., Groenewold, M., Casse, F.R., Sips, A.J.A.M., Dijkzeul, A., van Teunenbroek, T., Wijnhoven, S.W., 2016. Towards a nanospecific approach for risk assessment. Regul. Toxicol. Pharmacol. 80, 46–59. http://dx.doi.org/10.1016/j.yrtph.2016.05.037.

Delaval, M., Wohlleben, W., Landsiedel, R., Baeza-Squiban, A., Boland, S., 2017. Assessment of the oxidative potential of nanoparticles by the cytochrome *c* assay: assay improvement and development of a high-throughput method to predict the toxicity of nanoparticles. Arch. Toxicol. 91, 163–177.

DeLoid, G.M., Cohen, J.M., Pyrgiotakis, G., Demokritou, P., 2017. Preparation, characterization, and in vitro dosimetry of dispersed, engineered nanomaterials. Nat. Protoc. 12, 355–371.

Dourson, M.L., Stara, J.F., 1983. Regulatory history and experimental support of uncertainty (safety) factors. Regul. Toxicol. Pharmacol. 3, 224–238. http://www.tera.org/Publications/UFHIST83.pdf.

ECHA, 2017a. Appendix 4: Recommendations for Nanomaterials Applicable to the Guidance on Registration. Draft (Public) Version 1.0 January 2017. https://echa.europa.eu/documents/10162/23047722/appendix_4_nano_registration_committees_en.pdf/1abb12d1-88a2-b386-0907-c67d05105378.

ECHA, 2017b. Appendix R.6-1: Recommendations for Nanomaterials Applicable to the Guidance on QSARs and Grouping of Chemicals. Draft (Public) Version 1.0 May 2017. https://echa.europa.eu/documents/10162/23036412/appendix_r6_nanomaterials_en.pdf.

ECHA/JRC/RIVM, 2016. Usage of (Eco)Toxicological Data for Bridging Data Gaps between and Grouping of Nanoforms of the Same Substance. Elements to Consider. Available via. https://echa.europa.eu/documents/10162/13630/eco_toxicological_for_bridging_grouping_nanoforms_en.pdf.

Erdely, A., Dahm, M.M., Schubauer-Berigan, M.K., Chen, B.T., Antonini, J.M., Hoover, M.D., 2016. Bridging the gap between exposure assessment and inhalation toxicology: some insights from the carbon nanotube experience. J. Aerosol Sci. 99, 157–162.

Farcal, L., Torres Andon, F., Di Cristo, L., Rotoli, B.M., Bussolati, O., Bergamaschi, E., Mech, A., Hartmann, N.B., Rasmussen, K., Riego-Sintes, J., Ponti, J., Kinsner-Ovaskainen, A., Rossi, F., Oomen, A., Bos, P., Chen, R., Bai, R., Chen, C., Rocks, L., Fulton, N., Ross, B., Hutchison, G., Tran, L., Mues, S., Ossig, R., Schnekenburger, J., Campagnolo, L., Vecchione, L., Pietroiusti, A., Fadeel, B., 2015. Comprehensive in vitro toxicity testing of a panel of representative oxide nanomaterials: first steps towards an intelligent testing strategy. PLoS One 10, e0127174.

Gandon, A., Werle, K., Neubauer, N., Wohlleben, W., 2017. Surface reactivity measurements as required for grouping and read-across: an advanced FRAS protocol. J. Phys. Conf. Ser. 838 conference 1. http://dx.doi.org/10.1088/1742-6596/838/1/012033.

Gebel, T., Foth, H., Damm, G., Freyberger, A., Kramer, P.J., Lilienblum, W., Rohl, C., Schupp, T., Weiss, C., Wollin, K.M., Hengstler, J.G., 2014. Manufactured nanomaterials: categorization and approaches to hazard assessment. Arch. Toxicol. 88, 2191–2211.

Godwin, H., Nameth, C., Avery, D., Bergeson, L.L., Bernard, D., Beryt, E., Boyes, W., Brown, S., Clippinger, A.J., Cohen, Y., Doa, M., Hendren, C.O., Holden, P., Houck, K., Kane, A.B., Klaessig, F., Kodas, T., Landsiedel, R., Lynch, I., Malloy, T., Miller, M.B., Muller, J., Oberdorster, G., Petersen, E.J., Pleus, R.C., Sayre, P., Stone, V., Sullivan, K.M., Tentschert, J., Wallis, P., Nel, A.E., 2015. Nanomaterial categorization for assessing risk potential to facilitate regulatory decision-making. ACS Nano 9 (4), 3409–3417.

Gottardo, S., Alessandrelli, M., Amenta, V., Atluri, R., Barberio, G., Bekker, C., Bergonzo, P., Bleeker, E., Booth, A.M., Borges, T., Buttol, P., Carlander, D., Castelli, S., Chevillard, S., Clavaguera, S., Dekkers, S., Delpivo, C., Di Prospero Fanghella, P., Dusinska, M., Einola, J., Ekokoski, E., Fito, C., Gouveia, H., Grall, R., Hoehener, K., Jantunen, P., Johanson, G., Laux, P., Lehmann, H.C., Leinonen, R., Mech, A., Micheletti, C., Noorlander, C., Olof-Mattsson, M., Oomen, A., Quiros Pesudo, L., Letizia Polci, M., Prina-Mello, A., Rasmussen, K., Rauscher, H., Sanchez Jimenez, A., Riego Sintes, J., Scalbi, S., Sergent, J.-A., Stockmann-Juvala, H., Simko, M., Sips, A., Suarez, B., Sumrein, A., van Tongeren, M., Vázquez-Campos, S., Vital, N., Walser, T., Wijnhoven, S., Crutzen, H., 2017. NANoREG Framework for the Safety Assessment of Nanomaterials, EUR 28550 EN. http://dx.doi.org/10.2760/245972.

Grieger, K.D., Linkov, I., Hansen, S.F., Baun, A., 2012. Environmental risk analysis for nanomaterials: review and evaluation of frameworks. Nanotoxicology 6, 196–212.

Hansen, F.S., Alstrup Jensen, K., Baun, A., 2014. NanoRiskCat: a conceptual tool for categorization and communication of exposure potentials and hazards of nanomaterials in consumer products. J. Nanopart. Res. 16, 2195. http://dx.doi.org/10.1007/s11051-013-2195-z.

Hendren, C.O., Lowry, G.V., Unrine, J.M., Wiesner, M.R., 2015. A functional assay-based strategy for nanomaterial risk forecasting. Sci. Total Environ. 536, 1029–1037. http://dx.doi.org/10.1016/j.scitotenv.2015.06.100.

Heringa, M.B., Geraets, L., van Eijkeren, J.C.H., Vandebriel, R.J., de Jong, W.H., Oomen, A.G., 2016. Risk assessment of titanium dioxide nanoparticles via oral exposure, including toxicokinetic considerations. Nanotoxicology 10 (10), 1515–1525.

Hjorth, R., Hansen, S.F., Jacobs, M., Tickner, J., Ellenbecker, M., Baun, A., 2016. The applicability of chemical alternatives assessment for engineered nanomaterials. Integr. Environ. Assess. Manag. http://dx.doi.org/10.1002/ieam.1762.

Holden, P.A., Gardea-Torresdey, J.L., Klaessig, F., Turco, R.F., Mortimer, M., Hund-Rinke, K., Cohen Hubal, E.A., Avery, D., Barceló, D., Behra, R., Cohen, Y., Deydier-Stephan, L., Lee Ferguson, P., Fernandes, T.F., Herr Harthorn, B., Matthew Henderson, W., Hoke, R.A., Hristozov, D., Johnston, J.M., Kane, A.B., Kapustka, L., Keller, A.A., Lenihan, H.S., Lovell, W., Murphy, C.J., Nisbet, R.M., Petersen, E.J., Salinas, E.R., Scheringer, M., Sharma, M., Speed, D.E., Sultan, Y., Westenhoff, P., White, J.C., Wiesner, M.R., Wong, E.M., Baoshan, X., Steele Horan, M., Godwin, H.A., Nel, A.E., 2016. Considerations of environmentally relevant test conditions for improved evaluation of ecological hazards of engineered nanomaterials. Environ. Sci. Technol. 50

(12), 6124–6145. http://dx.doi.org/10.1021/acs.est.6b00608.
Hristozov, D., Gottardo, S., Critto, A., Marcomini, A., 2012. Risk assessment of engineered nanomaterials: a review of available data and approaches from a regulatory perspective. Nanotoxicology 6, 880–898.
Hristozov, D., Gottardo, S., Semenzin, E., Oomen, A., Bos, P., Peijnenburg, W., van Tongeren, M., Nowack, B., Hunt, N., Brunelli, A., Scott-Fordsmand, J.J., Tran, L., Marcomini, A., 2016. Frameworks and tools for risk assessment of manufactured nanomaterials. Environ. Int. 95, 36–53. http://dx.doi.org/10.1016/j.envint.2016.07.016.
Hund-Rinke, K., Herrchen, M., Schlich, K., Schwirn, K., Völker, D., 2015. Test strategy for assessing the risks of nanomaterials in the environment considering general regulatory procedures. Environ. Sci. Eur. 27, 24. 2015. http://dx.doi.org/10.1186/s12302-015-0053-6.
Hund-Rinke, K., Baun, A., Cupi, D., Fernandes, T.F., Handy, R., Kinross, J.H., Navas, J.M., Peijnenburg, W.J., Schlich, K., Shaw, B.J., Scott-Fordsmand, J., 2016. Regulatory ecotoxicity testing of nanomaterials - proposed modifications of OECD test guidelines based on laboratory experience with silver and titanium dioxide nanoparticles. Nanotoxicology 1–16. http://dx.doi.org/10.1080/17435390.2016.1229517.
van Kesteren, P.C., Cubadda, F., Bouwmeester, H., van Eijkeren, J.C., Dekkers, S., de Jong, W.H., Oomen, A.G., 2014. Novel insights into the risk assessment of the nanomaterial synthetic amorphous silica, additive E551, in food. Nanotoxicology 1–10. http://dx.doi.org/10.3109/17435390.2014.940408.
Landsiedel, R., Sauer, U.G., Ma-Hock, L., Schnekenburger, J., Wiemann, M., 2014. Pulmonary toxicity of nanomaterials: a critical comparison of published in vitro assays and in vivo inhalation or instillatiogn studies. Nanomedicine 9 (16), 2557–2585.
Landsiedel, R., Ma-Hock, L., Wiench, K., Wohlleben, W., Sauer, U.G., 2017. Safety assessment of nanomaterials using an advanced decision-making framework, the DF4nanoGrouping. J. Nanopart. Res. 19 (5), 171.
Lützhøft, et al., 2016. Environmental effects of engineered nanomaterials. Estimation of predicted no-effect concentrations (PNECs). In: Report Technical University of Denmark. Danish EPA.
Morose, G., 2010. The 5 principles of “Design for Safer Nanotechnology". J. Clean. Prod. 18, 285–289.
NANoREG 6.04, 2016. Inventory of Existing Regulatory Accepted Toxicity Tests Applicable for Safety Screening of MNMs. http://rivm.nl/en/About_RIVM/International/International_Projects/Completed/NANoREG/deliverables/NANoREG_D6_04_DR_Inventory_of_existing_regulatory_accepted_toxicity_tests.org.
NAS, 1983. Risk Assessment in the Federal Government: Managing the Process. National Academy of Sciences, Washington, D.C. https://www.nap.edu/download/366 (193 pages).
NAS, 2009. Science and Decisions: Advancing Risk Assessment. National Academy of Sciences, Washington, D.C. https://www.nap.edu/download/12209 (423 pages).
Nowack, B., Boldrin, A., Caballero, A., Hansen, S.F., Gottschalk, F., Heggelund, L., Hennig, M., Mackevica, A., Maes, H., Navratilova, J., Neubauer, N., Peters, R., Rose, J., Schäffer, A., Scifo, L., van Leeuwen, S., von der Kammer, F., Wohlleben, W., Wyrwoll, A., Hristozov, D., 2016. Meeting the needs for released nanomaterials required for further testing-the SUN approach. Environ. Sci. Technol. 50 (6), 2747–2753. http://dx.doi.org/10.1021/acs.est.5b04472.
OECD, 2005. Guidance Document on the Validation and International Acceptance of New or Updated Test Methods for Hazard Assessment [ENV/JM/MONO(2005)14]. http://www.oecd.org/officialdocuments/publicdisplaydocumentpdf/?doclanguage=en&cote=env/jm/mono(2005)14.
OECD, 2012a. Important Issues on Risk Assessment of Manufactured Nanomaterials, Series on the Safety of Manufactured Nanomaterials No. 33. http://www.oecd.org/officialdocuments/publicdisplaydocumentpdf/?cote=env/jm/mono(2012)8&doclanguage=en.
OECD, 2012b. Guidance on Sample Preparation and Dosimetry: Series on the Safety of Manufactured Nanomaterials No. 36. http://www.oecd.org/officialdocuments/publicdisplaydocumentpdf/?cote=ENV/JM/MONO(2012)40&docLanguage=En.
OECD, 2014. Ecotoxicology and Environmental Fate of Manufactured Nanomaterials: Test Guidelines, Expert Meeting Report, Series on the Safety of Manufactured Nanomaterials No. 40. http://www.oecd.org/officialdocuments/publicdisplaydocumentpdf/?cote=ENV/JM/MONO(2014)1&doclanguage=en.
OECD, 2015. Preliminary Guidance Notes on Nanomaterials: Interspecies Variability Factors in Human Health Risk Assessment, Series on the Safety of Manufactured Nanomaterials No. 58. http://www.oecd.org/officialdocuments/publicdisplaydocumentpdf/?cote=env/jm/mono(2015)31&doclanguage=en.
OECD 2017. Guidance Document on Aquatic and Sediment Toxicological Testing of Nanomaterials (Draft).
Oomen, A.G., Bos, P.M., Fernandes, T.F., Hund-Rinke, K., Boraschi, D., Byrne, H.J., Aschberger, K., Gottardo, S., von der Kammer, F., Kuhnel, D., Hristozov, D., Marcomini, A., Migliore, L., Scott-Fordsmand, J., Wick, P., Landsiedel, R., 2014. Concern-driven integrated approaches to nanomaterial testing and assessment—report of the NanoSafety Cluster Working Group 10. Nanotoxicology 8, 334–348.
Oomen, A.G., Bleeker, E.A., Bos, P.M., van Broekhuizen, F., Gottardo, S., Groenewold, M., Hristozov, D., Hund-Rinke, K., Irfan, M.A., Marcomini, A., Peijnenburg, W.J., Rasmussen, K., Jimenez, A.S., Scott-Fordsmand, J.J., van Tongeren, M., Wiench, K., Wohlleben, W., Landsiedel, R., 2015. Grouping and read-across approaches for risk assessment of nanomaterials. Int. J. Environ. Res. Public Health 12, 13415–13434.
Oosterwijk, M.T., Feber, M.L., Burello, E., 2016. Proposal for a risk banding framework for inhaled low aspect ratio nanoparticles based on physicochemical properties. Nanotoxicology 10 (6), 780–793. http://dx.doi.org/10.3109/17435390.2015.1132344.
Potthoff, A., Weil, M., Meißner, T., Kühnel, D., 2015. Towards sensible toxicity testing for nanomaterials: proposal for the specification of test design. Sci. Technol. Adv. Mater. 16, 065006. http://dx.doi.org/10.1088/1468-6996/16/6/065006.
Sayre P, Steinhäuser K, ProSafe – Roadmap for Members of Task Force When Reviewing Data, Protocols, Reports and Guidance Notes for Regulatory Relevance. Final version, 2016.
Sayre, P., Steinhäuser, K., van Teunenbroek, T., 2017. Methods and data for regulatory risk assessment of nanomaterials: questions for an expert consultation. NanoImpact (this issue).
SCENIHR, 2005. Opinion on the Appropriateness of Existing Methodologies to Assess the Potential Risks Associated With Engineered and Adventitious Products of Nanotechnologies. European Commission, Brussels Available: http://ec.europa.eu/health/ph_risk/committees/04_scenihr/docs/scenihr_o_003b.pdf, Accessed date: 11 September 2016.
SCENIHR, 2007. The Appropriateness of the Risk Assessment Methodology in Accordance with the Technical Guidance Documents for New and Existing Substances for Assessing the Risks of Nanomaterials. European Commission Health & Consumer Protection Directorate-General, Brussels Available: http://ec.europa.eu/health/ph_risk/committees/04_scenihr/docs/scenihr_o_010.pdf, Accessed date: 11 September 2016.
SCENIHR, 2009. Opinion on: Risk Assessment of Products of Nanotechnologies. European Commission, Brussels Available: http://ec.europa.eu/health/archive/ph_risk/committees/04_scenihr/docs/scenihr_o_023.pdf.
Sellers, K., Deleebeeck, N.M.E., Messiaen, M., Jackson, M., Bleeker, E.A.J., Sijm, D.T.H.M., van Broekhuizen, F.A., 2015. Grouping Nanomaterials - A strategy towards grouping and read-across. In: RIVM Report 2015-0061. National Institute for Public Health and the Environment (RIVM) & Arcadis, Bilthoven, The Netherlands Available from: http://www.rivm.nl/bibliotheek/rapporten/2015-0061.html.
Steinhäuser, K., Sayre, P., 2017. Reliability of methods and data for regulatory assessment of nanomaterial risks. NanoImpact (this issue).
Subramanian, V., Semenzin, E., Hristozov, D., Zabeo, A., Malsch, I., McAlea, E., Murphy, F., Mullins, M., van Harmelen, T., Ligthart, T., Linkov, I., Marcomini, A., 2016. Sustainable nanotechnology decision support system: bridging risk management, sustainable innovation and risk governance. J. Nanopart. Res. 18, 89. http://dx.doi.org/10.1007/s11051-016-3375-4.
Van Harmelen, T., Zondervan-van den Beuken, E.K., Brouwer, D.H., Kuijpers, E., Fransman, W., Buist, H.B., Ligthart, T.N., Hincapié, I., Hischier, R., Linkov, I., Nowack, B., Studer, J., Hilty, L., Som, C., 2016. LICARA nanoSCAN - a tool for the self-assessment of benefits and risks of nanoproducts. Environ. Int. 91, 150–160. http://dx.doi.org/10.1016/j.envint.2016.02.021.
Voelker, D., Schlich, K., Hohndorf, L., Koch, W., Kuehnen, U., Polleichtner, C., Kussatz, C., Hund-Rinke, K., 2015. Approach on environmental risk assessment of nanosilver released from textiles. Environ. Res. 140, 661–672. http://dx.doi.org/10.1016/j.envres.2015.05.011.
Walser, T., Studer, C., 2015. Sameness: the regulatory crux with nanomaterial identity and grouping schemes for hazard assessment. Regul. Toxicol. Pharmacol. 72 (3), 569–571. http://dx.doi.org/10.1016/j.yrtph.2015.05.031.
Wijnhoven, S.W.P., Peijnenburg, W.J.G.M., Herberts, C.A., Hagens, W.I., Oomen, A.G., Heugens, E.H.W., Roszek, B., Bisschops, J., Gosens, I., van de Meent, D., Dekkers, S., de Jong, W.H., van Zijverden, M., Sips, A.J.A.M., Geertsma, R.E., 2009. Nano-silver - a review of available data and knowledge gaps in human and environmental risk assessment. Nanotoxicology 3, 109–138.
Wohlleben, W., Brill, S., Meier, M.W., Mertler, M., Cox, G., Hirth, S., von Vacano, B., Strauss, V., Treumann, S., Wiench, K., Ma-Hock, L., Landsiedel, R., 2011. On the lifecycle of nanocomposites: comparing released fragments and their in-vivo hazards from three release mechanisms and four nanocomposites. Small 7, 2384–2395.