EUROPEAN COMMISSION

Directorate-General for
Health & Consumers

## Scientific Committee on Emerging and Newly Identified Health Risks

## SCENIHR

# Scientific Basis for the Definition of the Term “nanomaterial”

Scientific Committees
on consumer safety
on emerging and newly identified health risks
on health and environmental risks

The SCENIHR approved this opinion by written procedure on 8 December 2010

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## SCENIHR

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## ABSTRACT

With the expected increase in the applications of nanotechnology, there is an urgent need to identify what can be considered as a nanomaterial by clear unequivocal descriptions. This need to identify a nanomaterial comes from the uncertainty regarding safety evaluation and the risk assessment of nanomaterials. As a result, the SCENIHR was invited to provide advice on the essential scientific elements of an overarching working definition for the term “nanomaterial” for regulatory purposes. The scientific opinion concluded that:

- Whereas physical and chemical properties of materials may change with size, there is no scientific justification for a single upper and lower size limit associated with these changes that can be applied to adequately define **all** nanomaterials.

- There is scientific evidence that no single methodology (or group of tests) can be applied to **all** nanomaterials.

- Size is universally applicable to define all nanomaterials and is the most suitable measurand. Moreover, an understanding of the size distribution of a nanomaterial is essential and the number size distribution is the most relevant consideration.

In order to define an enforceable definition of “nanomaterial” for regulatory use it is proposed to set an upper limit for nanomaterial size and to add to the proposed limit additional guidance (requirements) specific for the intended regulation. Crucial in the guidance that needs to be provided is the extended description of relevant criteria to characterise the nanoscale. Merely defining single upper and lower cut-off limits is not sufficient in view of the size distributions occurring in manufactured nanomaterials. Alternatively, a tiered approach may be required depending on the amount of information known for any specifically manufactured nanomaterial and its proposed use.

The scientific opinion recognises however that specific circumstances regarding risk assessment for regulatory purposes for certain areas and applications may require the adaptation of any overarching definition.

It should be stressed that 'nanomaterial' is a categorization of a material by the size of its constituent parts. It neither implies a specific risk, nor does it necessarily mean that this material actually has new hazard properties compared to its constituent parts or larger sized counterparts.

Keywords: nanomaterial, definition, scientific basis

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## TABLE OF CONTENTS

## EXECUTIVE SUMMARY

With the expected increase in the applications of nanotechnology, there is an urgent need to identify by clear unequivocal descriptions what can be considered as a nanomaterial and what should not be. This need to identify a nanomaterial comes from the uncertainty regarding the safety evaluation and risk assessment of nanomaterials.

The SCENIHR was invited to provide advice on the essential elements of a science-based overarching working definition of the term "nanomaterial". More specifically they were asked to identify:

- The types of physical and chemical properties particular to nanomaterials,
- The threshold(s) at which nanomaterial-specific properties could be expected to occur and
- Potential methodology for nanomaterial characterisation

The scientific opinion concluded that:

- Whereas physical and chemical properties of materials may change with size, there is no scientific justification for a single upper and lower size limit associated with these changes that can be applied to adequately define **all** nanomaterials.
- There is scientific evidence that no single methodology (or group of tests) can be applied to **all** nanomaterials.
- Size is universally applicable to define all nanomaterials and is the most suitable measurand. Moreover, an understanding of the size distribution of a nanomaterial is essential and the number size distribution is the most relevant consideration.

The scientific opinion recognised however that specific requirements regarding risk assessment for regulatory purposes for certain areas and applications may require modifications of any overarching definition.

The major question for both regulators and manufacturers is to identify when a material or product can be considered a nanomaterial. This opinion has tried to address the complexity and the uncertainties and provide advice on the essential scientific elements for a working definition of the term “nanomaterial” for regulatory purposes using specific examples that may be applicable to certain classes of nanomaterials.

It should be stressed that “nanomaterial” is a categorisation of a material by the size of its constituent parts. It neither implies a specific risk nor does it necessarily mean that this material actually has new hazard properties compared to its constituent parts. However, size influences bio-distribution (and distribution kinetics) in an organism or in an ecosystem which should be taken into consideration in the risk assessment of nanomaterials.

There is sufficient evidence to indicate that there can be a change in some properties of the material when it or its constituent parts are at the nanoscale which is, for instance, due to the increased surface-to-volume ratio. These nano-specific properties raise concerns over their potential to cause harm to humans and the environment. The reaction rate of nanoparticles often relates to the available surface area. Consequently, chemical reactivity per mass dose increases for smaller particles of the same type; this effect may or may not be associated with an increase in biological activity or toxicity. It is this possible change in reaction rate that warrants the careful evaluation of possible risks associated with nanotechnology products. However, it is not currently possible to identify a specific size at which a specific property would change or appear, or a specific property that is introduced or changed with size.

Several international and national organisations have proposed definitions for the nanoscale and for nanomaterials (summarised in Annex I). In most of the proposed definitions, the size refers to one or more external dimensions or an internal structure within a specified size range. An upper limit of 100 nm or approximately 100 nm is commonly used. However, there is no scientific evidence to qualify the appropriateness of this value or any other single upper limit. Some definitions also include a reference to specific properties or nano-specific properties.

It should be noted that, in the metric system, the "nanoscale" is the range below 1 micrometre (μm) and above 999 picometre (pm). Criteria relevant for the discrimination between nano and non-nano are discussed using a working definition for the nanoscale being approximately 1 to 100 nm. Any material with one or more internal or external dimensions in the nanoscale is then considered a nanomaterial. The feasibility of including specific properties as elements of a definition was assessed.

When considering any definition for nanoscale and nanomaterial, size is the predominant feature. This requires that adequate validated methodologies are available for carrying out measurements at the nanoscale (i.e. below 1 μm). Any nanomaterial should be described by its size, and by its number size distribution, including the methodologies used for the measurement.

Solely referring to size as "one or more external dimensions" will NOT capture aggregates and agglomerates of primary particles or, importantly, more complex multi-component nanomaterials that are used in medical and cosmetic applications as their external dimension is likely to be larger than a specified upper size limit. The inclusion of a reference to "internal structure" with the same specified range as the external dimensions will include materials that consist of aggregates, agglomerates and multi-component assemblies within the scope of the definition. This would also include nanoporous and nanocomposite materials. If possible, also for the constituents of multi-component nanomaterials the size and number size distribution should be provided.

To distinguish a nanostructured material from a non-nanostructured material, the volume specific surface area (VSSA) can be a complementary criterion, based on its integral material surface area per unit volume. For powders and/or dry solid materials, the VSSA can be determined using a nitrogen absorption methodology called the BET method after Brunauer, Emmett and Teller (Brunauer et al. 1938). A limitation of the BET-method is that it is only applicable to powders and/or dry solid materials and not to nanomaterials embedded in solids and suspensions. Other methods exist that can be used for the analysis of both powders and particles dispersed in liquids but most still remain difficult to use on a routine basis. More developments are needed in the area of analysing powders and particles in liquids. Expressing the surface area related to the volume instead of mass allows for an additional criterion independent of the density and size or size distribution of the nanomaterial. A VSSA above $60 m^2/cm^3$ would indicate an average size below 100 nm, thus indicating a high nanomaterial or nanostructure content.

Data on the size distribution should be taken into account when describing a nanomaterial. When only a part of the material has a size within the size range of the definition or description, it should be clear whether and when such a material will be considered a nanomaterial. This may be by allowing a part (a certain percentage) of the number size distribution to be below a certain threshold or by using the information on the size distribution itself. Based on its geometric mean and geometric standard deviation, a material might be considered a nanomaterial when >0.15% of the material, as indicated by the number size distribution, has a size below the designated upper size limit.

As size is a key element in any definition of the term "nanomaterial", there seems to be a need for the development of validated standardised methods to determine size (including that of its constituent parts where relevant) and its corresponding distribution to ensure comparability of results.

There is a multitude of possibilities for the application of coatings and surface modifications to nanomaterials. Purposely applied and environmentally acquired coatings can have a major impact on nanomaterial interaction with biological systems. The coating and core together control the properties of a given nanomaterial. As a result, it is not useful to look at either the properties of the core or of the coating in isolation as they may not be representative of how the nanomaterial will behave in a given environment. The diversity in coatings of nanomaterials makes it impossible to include criteria based on surface properties within a definition as these properties may vary with coatings applied.

Several physico-chemical properties from the OECD Working Party on Manufactured Nanomaterials (WPMN) list of characteristics were evaluated as possible discriminators for the identification of a nanomaterial. They were crystalline phase, photocatalytic activity, zeta potential, redox potential, radical formation potential, water solubility and the octanol-water partition coefficient. It was concluded that while all of these properties are very useful for risk assessment, none of them appear to be universally applicable as a criterion within a definition for all nanomaterials.

Like any other material, nanomaterials can be degraded either mechanically, chemically or by dissolution; in fluids, they can form agglomerates or stable dispersions depending on solvent chemistry and surface coating. Features associated with solubility (and degradability) of nanomaterials are very important for risk assessment in view of the possibility for persistence and accumulation both in man and the environment. These features include size and shape, water solubility, surface charge and surface reactivity. However, these features cannot be translated into a definition as they are part of the characterisation of a nanomaterial and can change for each individual nanomaterial depending on chemical composition, surface modification and the immediate environment of the nanomaterial.

Certain nanomaterials and composite materials may contain internal or external structures at the nanoscale that were incorporated to confer nanospecific characteristics. As the external dimensions of nanocomposites would be typically larger than 100 nm, a definition based solely on external size would not consider most nanocomposites to be nanomaterials. The internal structure with a size at the nanoscale would be an element to include in a definition, as then nanocomposites will be included in the definition of a nanomaterial. There are also nanocomposites where one phase is the solid bulk. Exclusion criteria would have to be developed to avoid considering macroscopic composite objects as nanomaterials

In order to specifically designate purposely made nanomaterials within regulations, the term “engineered” or “manufactured” may be used. When considering purposely made nanomaterials, the meaning of “engineered” or “manufactured” needs to include the processing (e.g. grinding or milling resulting in size reduction, or chemical processing) of materials to obtain materials at the nanoscale.

In conclusion, size (and its distribution) is universally applicable to all nanomaterials and is the most suitable measurand. A defined size range would facilitate a uniform interpretation. For regulatory purposes the number size distribution should also be considered using both the geometric mean size and its geometric standard deviation for further refinement of the definition. Alternatively a specific fraction of the number size distribution might be allowed to be within the specified size ranges of the definition. For dry powders, the volume specific surface area (VSSA) may be added to the size as a complementary discriminator to identify nanomaterials. In addition, the definition should include both external and internal nanostructures.

A size of 1 nm is proposed as the lower limit for the definition of nanomaterials. However, around 1 nm the distinction between molecules, nanoclusters and nanoparticles is unclear. In general molecules should be excluded. However, exceptions can be made to allow for inclusion of certain specific entities.

At the moment, no scientific data are available to indicate that a specific size associated with special properties due to the nanoscale can be identified for nanomaterials in general. There is no scientific evidence in favour of a single upper limit. Although an upper limit of 100 nm is commonly used, there is no scientific evidence to qualify the appropriateness of this value. Notably, the use of a single upper threshold value might be too limiting for the classification of nanomaterials and a differentiated approach might be more appropriate. This approach could be based on a relatively high upper threshold for which it is assumed that the size distribution at the lower end will most likely be above the lower, more critical threshold. The lower threshold would be the critical one for which extensive nano-specific information would be needed in order to perform case-by-case risk assessment.

For specific areas and applications, modifications of any overarching definition may be needed due to specific requirements regarding risk assessment for regulatory purposes.

## 1. BACKGROUND

The services of the European Commission urgently need to elaborate a working definition for the term "nanomaterial" to ensure the consistency of forthcoming regulatory developments to guide, as appropriate, the effective implementation of existing regulation, and to contribute to international work and dialogue on nanotechnology definitions.

The SCENIHR adopted a scientific opinion on "The scientific aspects of the existing and proposed definitions relating to products of nanoscience and nanotechnologies" at the 21st plenary meeting on 29 November 2007.[1] Moreover, both SCENIHR[2] and the predecessor to the Scientific Committee on Consumer Safety (SCCS)[3] have provided further advice on the definitions of the term nanomaterial and other related terms in their opinions. Moreover, the European Food Safety Authority used the terms and definitions suggested by the SCENIHR in the opinion on "The Potential Risks Arising from Nanoscience and Nanotechnologies on Food and Feed Safety" on 10 February 2009.[4]

In order to prepare a science-based definition of nanomaterials, the services of the European Commission need clarification on the size ranges and other relevant characteristics and corresponding metrics reported in the scientific literature, the types of physical and chemical properties particular to nanomaterials, the relevant thresholds, as well as the most appropriate metrics to express such thresholds.

The development of the policy and regulatory activities on nanotechnologies requires the establishment of a working definition of nanomaterials as a matter of urgency. Therefore, SCENIHR is requested to provide a scientific opinion on the issues mentioned below in accordance with the accelerated procedure referred to in Article 9.13 of the Rules of Procedure, in co-operation with other Scientific Committees of the European Community and, as appropriate, with external experts.

## 2. TERMS OF REFERENCE

Advice on the essential elements of a science-based working definition: Based on current knowledge, the Committee was invited to provide advice on the essential elements of a science-based working definition of "nanomaterials" and, specifically, to identify the most appropriate metrics to define materials at the nanoscale, taking into account:

(i) Reported size ranges and other relevant characteristics and corresponding metrics: The size ranges and other relevant characteristics (e.g., specific surface area, shape, density, spatial arrangements, aggregation, agglomeration, etc.) and corresponding metrics of materials reported as "nanomaterials" in the scientific literature;

(ii) Characteristics: A first indication of possible characteristics and associated mechanisms that alone or in various combinations may lead to different properties;

(iii) Physico-chemical properties: The physical and chemical properties that materials may show as a result of being at the nanoscale or having a nanoscale structure;

(iv) Threshold(s): The threshold(s) at which properties identified in (iii) above may be expected to occur (the threshold(s) may be "below" or "above" depending on the relevant characteristic(s) and associated metric(s)).

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[1] http://ec.europa.eu/health/ph_risk/committees/04_scenihr/docs/scenihr_o_012.pdf
[2] http://ec.europa.eu/health/ph_risk/committees/04_scenihr/docs/scenihr_o_023.pdf
[3] http://ec.europa.eu/health/ph_risk/committees/04_sccp/docs/sccp_o_099.pdf
[4] http://www.efsa.europa.eu/en/scdocs/scdoc/958.htm

# 3. SCIENTIFIC RATIONALE

## 3.1. Introduction

The rapid development, increased production and use of nanomaterials have raised concerns that such materials may introduce new hazards during occupational exposure, consumer exposure and/or on environmental exposure. Nanomaterials are being engineered for their specific physico-chemical and biological characteristics thus providing novel materials with promising technological advances.

Reduction in size can result in materials with specific physico-chemical properties that distinguish them from the bulk[5] (larger sized) of the same material (Auffan et al. 2009, Gleiter 2000, Jiang et al. 2008, SCENIHR 2006). The properties of a material generally depend on its chemical composition and on the environment at the interface (such as the surrounding medium (air, liquid, solid), temperature, and pressure). With decreasing particle or structure size there is an increase in surface area in relation to the volume. This results in an increase in the number of molecules/atoms at the surface which can potentially change the surface reactivity.

With the progress in nanoscience, nanomaterials are typically engineered to have specific properties. Bottom up methods such as chemical synthesis and self-assembly yield nanomaterials that are often not directly comparable to any “bulk” counterpart. Bottom-up methods typically yield nanomaterials that are composed of multiple components. Many nanomaterials are engineered and manufactured for their specific properties, often with well known chemical composition. Although the toxicological profile of the chemical components of a given nanomaterial may be well known, there may be cases where its specific properties raise concerns about their specific potential to harm humans and the environment. This raises the question as to whether the current risk assessment methodology as used for "classic" substances (chemicals) in the EU can be used for nanomaterials or whether there is a need to change the risk assessment methodology (Kreyling et al. 2006, Oberdörster et al. 2007, Oberdörster 2010).

Part of the concern can be attributed to the fact that it is not yet known whether the assays currently used for hazard identification and risk assessment of substances (chemicals) can also be applied to risk assessment of nanomaterials or whether they need to be modified. Obviously, a sufficiently precise assessment of the chemical ingredients of a nanomaterial is a pre-requisite for such an analysis. Currently, the OECD is running a sponsorship programme aimed at investigating whether the assays described in the various OECD guidelines for the testing of chemicals (OECD 2009a) can be applied to nanomaterials. These guidelines comprise five sections: 1-Physical chemical properties; 2-Effects on biotic systems; 3-Degradation and accumulation; 4-Health effects; and 5-Other test guidelines. An “OECD series on principles of good laboratory practice and compliance monitoring” and an “OECD series on testing and assessment” complement these guidelines. A major issue is that so far the testing guidelines were developed for chemicals with only limited attention given to the testing of particles (powders) for risk assessment.

Since 1990, a growing number of nano-sized products have been approved for routine human use as nanopharmaceuticals and nano-sized imaging agents in the pharmaceutical area. In this context, the methodology used to assess preclinical safety of both specific nanomaterials and first generation nanomedicine products is already documented (Gaspar and Duncan 2009). Moreover, for such products there has been considerable post-market patient surveillance documenting both safety and efficacy.

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[5] In particle toxicology, the term “bulk” is often used to distinguish nanoparticles to larger particles of the same chemical substance. Equally relevant is the comparison of the nanoparticulate form of a chemical with the free (atomic, ionic, molecular) gaseous or dissolved species. All possible species (gaseous/dissolved, nanoform, aggregates/agglomerates and conglomerates with other materials) may play a role in the way nanomaterials affect organisms. In this text, the term “bulk” is used to refer to all non-nano species of a nanomaterial.

Previously, the use of a case-by-case approach for safety evaluation and risk assessment of nanomaterials has been recommended (EFSA 2009, FDA 2007, SCENIHR 2009) as extrapolation from one nanomaterial to another is not considered feasible even when the basic chemical composition is the same. For example, a nanomaterial with a particle size of 20 nm must be considered different from a nanomaterial of 80 nm. There is growing pressure to set a definition that will allow identification of those "nanomaterials" for which a separate or alternative safety evaluation and/or risk assessment is needed rather than the standard methodology applied to "classic" materials/chemicals.

It should be stressed that the term "nanomaterial" is a categorisation of a material by the size of its constituent parts. It neither implies a specific risk nor does it necessarily mean that this material actually has new hazard properties compared to any smaller constituent parts of the nanomaterial or the bulk (or larger sized powder form) if such exists. Engineering of "nano" size can, but not necessarily does, result in a change of physico-chemical properties. However, a decreasing size will always result in a corresponding change in biodistribution (and distribution kinetics) in an organism or in an ecosystem. There is an analogy with the toxicological assessment of chemical compounds (with some new issues however), in that regardless of whether a compound is synthetic, synthesised to be nature identical or extracted from natural substances, this does not provide any clue about its toxicity profile. Moreover, it is well known that due to differences in distribution and/or metabolism even different isomeric forms of the same compound can have differing toxicity and efficacy.

In general the current risk assessment methodology is also applicable for nanomaterials as the methodology allows for sufficient flexibility. Although most of the existing toxicological and ecotoxicological methods for hazard identification are likely to be appropriate, they may not be sufficient to address all the hazards of nanomaterials. Therefore not all currently applied methodologies for hazard identification or exposure assessment of chemicals may be applicable to nanomaterials. The assays may need to be supplemented by additional tests, or replaced by modified tests, as it cannot be assumed that current scientific knowledge has elucidated all the potential adverse effects of nanoparticles.

Standard chemistry and physics define atoms, molecules, polymers, supra-molecular aggregates, colloids, particles and (nano)clusters, and how they are discriminated or characterised. In the context of material properties, it is important to note that the size of large individual molecules reaches well into the nanometre range, and that, like in any other case, the property of such molecules can be designed by chemical synthesis. Similarly, the size of nanoparticles and nanoclusters may reach into the sub-nanometre range.

The key question to be answered is which elements or characteristics should be included in a definition of "nanomaterial" for regulatory purposes. However, one should not confuse the definition of a subject with the necessary characterisation of that subject. It is very useful to know some parameters and they may also be part of the characterisation/description of the nanomaterial. It is also important to note that a parameter or characteristic would have to be applicable to all nanomaterials if it were to be included in a universal definition for "nanomaterial". In this opinion, several scientific and practical considerations are discussed that may affect the description of a nanomaterial.

## 3.2. Existing and proposed definitions

Several definitions for nanomaterials have been formulated by various national and international bodies including BSI, FDA, Health Canada, ISO 2008, OECD 2008, SCENIHR 2007 (see Annex I) and considerations regarding a definition for regulatory purposes have recently been provided by the Joint Research Centre of the European Commission (Lövestam et al. 2010). A common feature of many definitions has been to specify an upper size limit for nanomaterials of 100 nm for one or more external dimensions. Others have argued that it is difficult to ascribe the specific upper threshold to this value. An upper limit of 1,000 nm has been proposed for pharmaceuticals (Brouwer et al. 2010, FDA 2010). It is argued that a single upper threshold of 100 nm (or a 1 nm–100 nm range for nanomaterial size) cannot be considered to properly distinguish nanomaterials from their bulk counterparts as it does not take into account issues like size range, size distribution and specific properties (electrical, mechanical, optical) at the nanoscale. Thus a more elaborate description of a nanomaterial may be needed that can be used as a working definition for regulatory purposes. Equally, it must be noted that other elements included in a definition must be applicable to all possible nanomaterials.

In the definitions proposed so far, the lower size varies from approximately 1 nm, 0.2 nm, and 0.1 nm to no lower limit for one or more external dimensions. It is necessary to include a lower limit in a definition for nanomaterials because around 1 nm the distinction between molecules, atoms and nanoparticles is unclear. Moreover, molecules and atoms should not be included in a definition of a nanomaterial. The size of some individual large molecules may reach well into the nanometre range. While some are usually considered as a nanomaterial (e.g. fullerenes), others may just be large molecules without any indication of them being a nanomaterial.

Most definitions also refer to the internal structure with the same specified size range. The latter includes aggregates and agglomerates of particles within the scope of the definition. Aggregates and agglomerates would not be captured by a definition solely based on external dimensions. This internal structure seems to be a logical criterion that could be included. However, including it would also include nanoporous materials such as membranes within the definition and this may seem counter-intuitive to those who are more familiar with the concept of nanomaterials as nanoparticles. In addition, the change in physicochemical characteristics is included in some definitions/descriptions. In some definitions, this criterion is in addition to the size specification while in others it supersedes the size requirement. The latter would allow materials that are larger than 100 nm that show specific properties to be included in the definition/description of a nanomaterial. This criterion implicitly assumes that there is a corresponding bulk counterpart for the nanomaterial to compare properties with. If there is no bulk counterpart, this criterion regarding changes in physical-chemical properties cannot be used as the frame of reference is missing.

In addition, some regulatory definitions are already present in the legislation (e.g. EU Cosmetics Directive 76/768/EC) or presented by governmental institutions as a basis for general discussion (e.g. Health Canada, 2010).

A hierarchical approach to the terminology within nanotechnology and nanosciences has been described by ISO where a nanomaterial can be either a nano-object (one or more external dimensions in the nanoscale) or a nanostructured material (internal structure or surface structure in the nanoscale) (ISO/TS 27687:2008, ISO/TS 80004-1:2010). In this context, a nanomaterial is solely defined by its size which can refer to either internal or external dimensions. The SCENIHR 2007 opinion describes a framework that follows mainly existing terminology with additions only for specific keywords within nanotechnology and nanosciences (SCENIHR 2007).

In conclusion, the definitions/descriptions of nanomaterials formulated so far have i) given a general size frame for nanomaterials in both external and internal dimensions and ii) referred in some to the unique physico-chemical characteristics of the specific material under discussion. Whilst such broad definitions can be scientifically justified, they are not easy to apply within the context of a regulatory framework.

## 3.3. Parameters to be considered

A "nanomaterial" may consist of a single element or numerous different elements. The term may apply to both simple and complex inorganic and organic substances as well as derivatives thereof and mixtures, including nano-composite materials. All nanomaterials may be chemically and/or physically modified. They may be coated (covalently or non-covalently) or otherwise functionalised to obtain their specific, technologically required performance.

Typically "nanomaterials" are manufactured using "top-down" (e.g. milling) or "bottom-up" methodology (e.g. chemical synthesis, supramolecular assembly, covalent conjugation involving multiple components, colloid chemistry). Complex multi-component nano-sized materials have been engineered using a combination of both of these routes, and indeed many emerging nanomaterials are prepared by the sequential application of several different manufacturing steps.

The nature of the manufacturing process, and any integral purification steps, will ultimately govern the purity of the final product, particle size distribution and heterogeneity of other parameters. In addition to the specific physico-chemical properties of each material these factors, together with storage conditions and degree of aggregation, play a major role in determining ultimate biological behaviour.

This scientific opinion discusses key criteria that should to be considered when using a definition for "nanoscale" and/or "nanomaterials" in a regulatory context. These criteria are a non-exhaustive list but are considered the most important ones for which guidance might be needed when applying any regulatory definition for nanotechnology.

### 3.3.1. Size

Size is the universal element that is included in all proposed definitions thus far. The use of size is indicated by the prefix "nano-", which specifically means a measure of $10^{-9}$ units, the nature of this unit being determined by the word that follows. For the metric scale, this means 1 to 999 nm (the size below 1 μm).

Upper Size Limit

As mentioned in section 3.2, most previous reports specify an upper limit for a nanomaterial of approximately 100 nm (and a lower limit of approximately 1 nm for one or more external dimensions).

The upper physical size-range for nanomaterials may also be considered depending on the intended application of the cut-off (i.e. material properties or risk). While the upper size limit of 100 nm would catch the most obvious nanomaterials, such as primary metal based nanoparticles of various shapes, hierarchical assemblies of primary particles such as agglomerates and/or aggregates often have external dimensions greater than this. Aggregation and agglomeration can occur due to a number of deliberate and accidental mechanisms (see for example the review by Schneider and Jensen (2009)). When hierarchical assemblies, aggregates and agglomerates are included in the determination of the size, their presence induces a shift to larger sizes.

Particulate nanomaterials may also be reported with larger sizes due to coatings or functionalisation by long-chained organic compounds. Hence, for risk assessment purposes, it may be necessary to define a threshold size based on the size of the core of the individual particulate nanomaterial and not on the complete functionalised particle.

In particular, more complex nanomaterials such as liposomes that can be loaded with drug particles or metal particles and that are used in medicinal and cosmetic applications would typically have external dimensions greater than 100 nm for one or more external dimensions. The inclusion of "internal structure" with the same specified upper limit would however define such structures as nanomaterials provided the components are within the specified upper limit. The next-generation hybrid nanomaterials are already being developed and are typically based on the concept of a hierarchical assembly of many components (e.g. quantum dot superlattices, dendrimers and polymers) (Love et al. 2005, Daniel and Astruc 2004, Schmidt and Bodmeier 1999). Therefore, a reference to the internal structure should be included within the definition as this would then also capture complex assemblies provided the internal structure was within the specified range. Any definition should aim to be over-arching to also include next-generation nanomaterials in order to avoid quickly becoming obsolete (Roco 2004).

### Lower size limit

Focusing on the external dimensions, the lower cut-off size for nanomaterials presents another challenge as 1 nm or less also includes the size of atoms, molecules and clusters. Depending on the specific compounds, there will be a transition from atoms to nanoclusters or molecules to particles at different sizes which makes the definition of a specific lower size limit challenging. A lower limit is needed, but will not definitively avoid all the problems stated, as molecules are defined by the covalent links between atoms and range from sub-nanometre dimensions (CO, $O_2$ etc.) up to the size of some globular proteins (~5 nm) to macroscopic dimensions in the case of interlinked structural proteins (e.g. collagen) and many technical polymers. It should be noted that at sizes around 1 nm there is an ambivalence between molecules, and certain specific entities such as fullerenes, dendrimers, graphene, nano-clusters and complex hybrid molecular structures (Abad et al. 2007). In addition, there are tubes and fibres which may have a diameter below 1 nm and a length above 100 nm. (Zhou et al. 2002)

### Methods for characterisation of size

The specific method(s) used to determine nanomaterial size is very important. There are currently very few standard methods for determining size. There is also an apparent lack of awareness among the producers that not all techniques measure the same thing. For example, Transmission Electron Microscopy (TEM) and Atomic Force Microscopy (AFM) measure the size without any organic coating whereas the size determined by Dynamic Light Scattering (DLS) includes the organic coating as well as the ionic double layer in the measurement. Various authors (Park and Grassian 2010, Domingos et al. 2009, Frank et al. 2008) reported that size could deviate considerably from that indicated by the manufacturer depending on the methodology used for characterisation. For example, differences of two to three orders of magnitude were reported, with the methods in closest agreement to the manufacturer's information being TEM and AFM. Sizing a particulate material needs to be done using different techniques depending on whether the nanoparticles occur as a powder, are dispersed in a liquid, are coated or are embedded a solid material.

In conclusion, it should be realised that, at the lower nanoscale, there are molecules and clusters with sizes well above the proposed 1 nm lower limit that should be taken into consideration when designating a material as nanomaterial based on the lower limit of size. In general, molecules and clusters should not be included in a definition of a nanomaterial but certain specific entities may need to be included (e.g fullerenes, dendrimers, and complex hybrid molecular structures). Conversely, some molecules with a size lower than 1 nm should be taken into consideration (e.g. certain nanotubes). Some (nano)tubes and (nano)fibres may have a diameter below 1 nm and a length above 100 nm. For the upper limit, the presence of complex structures, agglomerates and/or aggregates should be taken into consideration. To overcome problems with a definition associated with complex structures a reference to the size determination of possible internal structures might be included. In addition, the methodology with which the size was determined should be indicated as this affects the outcome of the measurement. More reliable information is obtained when the determined size is reported using a few different complementary standardised methods. As size is a key element in any definition, it can be concluded that there is a need for the development of validated standardised methods to determine size.

### 3.3.2. Size distribution

One cannot consider size without addressing the issue of size distribution. Although it has been demonstrated that it is possible to produce monodisperse nanoparticles, it is important to note that most nanomaterials produced will have a size distribution (Park et al., 2005). Thus, for every threshold proposed, there remains the question of whether a product should be defined as a nanomaterial (or not) when a fraction of the size distribution falls below and/or above the chosen threshold(s). The following text on size distribution refers to the distribution in the size of one or more external dimensions, specifically primary particle size. The external size of aggregates/agglomerates of primary particles is not considered as these may not be at the nanoscale although the same considerations would apply to the constituent internal primary particles.

Clearly, it is important that any figure given for the size of a particular product (i.e. nanomaterial formulation or production) must be qualified with a specific descriptor of the size distribution. If the size distribution follows a normal distribution the size can be described by its mean and the distribution by ± the standard deviation (SD). However, most nanomaterials do not show a normal distribution, and many can be described by a log-normal distribution. This can be described by the geometric mean diameter and the geometric standard deviation. For all nanomaterials it is essential that the appropriate statistical measure of both average size and size distribution is presented.

It should be stressed that data on size and number size distribution should be complemented by information describing the number concentration of the particles as well as the mass concentration. Clearly, a low mass concentration of nanoparticles in a product may still represent a high number of particles and a mass based distribution can be skewed by the presence of relatively few large and thus heavy particles. A specification would be required that indicates whether the size distribution is provided for the mass, volume concentration, or number concentration.

The standard deviation of the size distribution may be used for fine tuning the definition of a nanomaterial. For example, a material consisting of spherical particles might be defined as being a nanomaterial when more than 0.15% has a diameter below 100 nm. Assuming that the distribution is log-normal, this criterion is fulfilled if the mean of the $\log_{10}$-transformed sizes minus three times the Standard Deviation (of the $\log_{10}$-transformed sizes) is smaller than two (which is the $\log_{10}$ of 100 nm). It should be realised that nanomaterials may not show a log-normal size distribution (e.g. they might have a bi-modal size distribution). For determining the fraction below 100 nm in bi-modal distributions specific statistical software would be needed.

Reporting the summary statistics of a size distribution should be done in one of the following ways:

- Mean and SD of the log-transformed sizes; and

- Back-transformed values of the mean and SD, i.e. the geometric mean and geometric SD, respectively.

Reporting the arithmetic means and SDs of the sizes themselves is not considered appropriate.

In conclusion, the size distribution cannot be ignored and should be taken into account when defining/describing a nanomaterial. When only a part of the material has a size within the size range of the definition/description it should be clearly described whether and when such a material will be considered a nanomaterial or not. This may be by allowing a part (certain %) of the size distribution to be below a certain threshold or by using the information on the size distribution itself. The size distribution of a material should be presented as size distribution based on the number concentration and not on the mass concentration of a nanomaterial product as a small mass concentration may contain the largest number fraction.

### 3.3.3. Specific surface area

Since some nanoparticulate materials may have a broad size distribution it is difficult to draft a suitable definition based on a single parameter such as the median or mean diameter of a distribution. Indeed this definition may be extended to provide additional parameter(s) such as the standard deviation of a normal distribution or the geometric standard deviation of a log-normal distribution.

Even with the addition of a second size related parameter, this size-based definition falls short in the case of highly agglomerated and/or aggregated particles. For this reason, most definitions include a reference to the internal structure which would capture such structures, provided the primary particle size of the aggregate is within the specified size range. However, there are difficulties associated with the measurement of primary particle size in aggregates that should not be overlooked. For agglomerates, aggregates, and porous materials, measurement of the surface area may be more meaningful. Such materials may exhibit a large surface area even when a sizable fraction of the external size distribution (e.g. the aggregates and agglomerates) is much larger than 100 nm.

To overcome this problem the specific surface area of a particulate powder material is an important complementary parameter. Kreyling et al. (2010) suggested using the volume specific surface area (VSSA) as an additional parameter. The VSSA is an integral parameter determined from the entire particulate powder material including the whole size range distribution, with all external and/or internal surfaces. It characterises the entire particulate surface area per volume of a solid and/or powder material.

A threshold for the distinction of nano- versus micro-sized material can be derived based on a proposed threshold of the minimal surface area of the particulate material. For example based on the upper size limit of 100 nm spherical particles of unit-density for a nanomaterial, such a threshold of the VSSA would be set at:

$$\mathrm{VSSA} \geq 60\ [\mathrm{m^2/cm^3}]$$

This means that any material with a VSSA $\geq$ 60 $\mathrm{m^2/cm^3}$ will fall in the category of nanostructured material, even though it may consist of a broad size distribution and be aggregated and/or agglomerated.

This VSSA threshold limit can be generalised to other values of VSSA when deriving from other diameters D:

$$\mathrm{VSSA}(D) \geq 6000/D\ [\mathrm{m^2/cm^3}] \qquad \text{(D given in units of nanometre)}$$

A practical advantage of the VSSA parameter is its simple calculation from two parameters usually available for each commercial nano- or micro-structured powder material (e.g. in CAS specifications); the two parameters are the density ($\rho$) given in $\mathrm{g/cm^3}$ and the mass specific surface area (SSA) given in $\mathrm{m^2/g}$ of nanostructured material; the latter is usually determined by nitrogen absorption methodology called BET method after Brunauer, Emmett and Teller (Brunauer et al. 1938).

$$\mathrm{VSSA} = \mathrm{SSA} * \rho$$

To be able to determine the VSSA, the density of the material has to be known. In most cases, the density of the bulk material can be used. However, when there is no bulk equivalent of the nanomaterial, the density of the nanomaterial itself should be determined. The BET-method allows surface area or porosity measurements within pores or other nanostructures as small as about 1 nm. Hence, the density of only the material without the empty spaces in between is required. Thus, using the bulk density ($\rho$) provides a reasonable integrated first estimate of the atomic or molecular structural alignments in the smallest material units. A limitation of the BET-method is that it is only applicable to powders and/or dry solid materials and not to nanomaterials embedded in solids and suspensions. Other methods exist that can be used for the analysis of both powders and particles dispersed in liquids but most still remain difficult to use on a routine basis. (Dekany and Turi 1997, Szekeres et al. 2002, Fairen-Jimenez et al. 2006, Mascotto et al. 2009). More developments are needed in this domain. It is worth noting that the estimated VSSA of a material is valid for the entire material as analysed. If a fraction or subset of the material (e.g. fractionated by size) is analysed, it will have a different $VSSA_{fr}$ which may be higher or lower than the VSSA of the initial entire material.

In conclusion, the VSSA is a complementary qualifier (criterion) to distinguish dry solid nanostructured material from non-nanostructured material based on its integral material surface area per material volume. The proposed VSSA threshold limit is 60 $m^2/cm^3$ above which the material is considered to be a nanomaterial.

The VSSA could be considered as an additional criterion that could be used to identify whether a material is a nanomaterial. However, for materials with a VSSA below 60 $m^2/cm^3$ there is still the possibility that a fraction may have a VSSA above 60 $m^2/cm^3$ in view of the size distribution. In addition, it should be noted that not all nanomaterials are easily amenable to VSSA determination by the BET-method (e.g. dispersions, nanocarriers etc.). A limitation of the BET-method is that it is only applicable to powders and/or dry solid materials and not to nanomaterials embedded in solids and suspensions. More developments are needed in the area of analysing powders and particles in liquids. Also for complex assemblies that have an external size greater than 100 nm, e.g. nanocarriers in drug delivery, the specific surface area of the internal components is intrinsically not measurable.

### 3.3.4. Surface modification

Surface modification of a nanomaterial can either be done by coating, functionalisation or other means, which may be chemical (organic, inorganic or both) or physical (e.g. irradiation, surface attrition) (Burda et al. 2005, Daniel and Astruc 2004, Love et al. 2005). Nanoparticles can be composed of a single external interface or more sophisticated multilayered shells. For example, a $TiO_2$ nanoparticle can be coated with a thin layer of $SiO_2$ (inorganic surface treatment) and the $SiO_2$ layer can be further treated with an alkylsilane (organic surface treatment). These form so-called "core-shell" type nanomaterials where the coating is the shell and a given core can have multiple shells (Chen et al. 2007, Selvan et al. 2007, Yu et al. 2005). The term "coating" may also refer to the charged groups on the surface of a nanomaterial, e.g. the hydroxyl groups on the surface of a silica particle. The type of coating on the outer surface of a given nanomaterial determines its stability to degradation or aggregation in a given medium. The choice of coating is usually application driven and has a direct influence on the binding of the nanomaterial with biomolecules, lipids proteins etc. and thus can affect the interaction of the nanomaterial with biological systems (Chen et al. 2007, Selvan et al. 2007).

It is important to realise the enormous variety of surface modifications that are possible. Nanomaterials can have identical surface coatings but completely different cores or vice versa. There can be multiple coatings on a given surface (Chen et al., 2007). Moreover, a nanomaterial may acquire coating materials (proteins, lipids etc.) from the environment by natural processes. The stability of the coating (it may degrade or dissociate with time or it may leave remnants of covalent anchoring chemistry attached to the nanomaterial; etc.) is also a key factor. The properties of a given nanomaterial are determined by both the core and coating, i.e. the coating is an intrinsic part of the nanomaterial and cannot be considered separately.

In conclusion, there is a multitude of possibilities for the application of coatings on nanomaterials. Purposely applied and environmentally acquired coatings can have a major impact on nanomaterial interaction with biological systems. The coating and core together control the properties of a given nanomaterial and it is not useful to look at either the properties of the core or the coating in isolation as they may not be representative of how the nanomaterial will behave in a given environment. Thus, each combination of a nanomaterial and a coating has to be considered as an individual case when safety evaluation of a specific nanomaterial is considered.

The variability in coatings on nanomaterials challenges the feasibility of including elements based on properties within a definition as these properties may vary with coatings as highlighted above.

### 3.3.5. Other physical-chemical characteristics

There are other physical-chemical characteristics that may be of relevance when considering which elements should be included within a definition

Nanoparticles and nanostructured materials may be characterised by a number of physico-chemical characteristics. The OECD WPMN has specified parameters for describing the primary nanomaterial of which several are related to the material properties and not to the physical appearance and state of the material (OECD 2008b).

Some of these parameters and their relevance are discussed as potential criteria to be included in a definition.

<u>Crystalline phase, crystallinity and small structures</u>

Specific inorganic compounds show changes in crystalline structure and morphology as function of particle size/specific surface area (e.g. Navrotsky et al. 2008). For example, $TiO_2$ occurs in at least four different natural polymorphs (amorphous, anatase, brookite and rutile) changing with decreasing specific surface area (e.g. Ranade et al. 2002, Zhang and Banfield 1998). Based on determined crystallite sizes, Ranade et al. (2002) showed that all transitions occurred within the 1 to 100 nm range where the stability field between the X-ray amorphous state and crystalline anatase was observed between 150 and 433 $m^3$/g.

Auffan et al. looked into the definition for nanomaterials, reviewed the effects of size on physico-chemical properties of several semi-conducting nanomaterials and suggested that the evidence for novel size-dependent properties alone, rather than particle size, should be the primary criterion in any definition (Auffan et al., 2009). For the evaluated nanoparticles, unique properties were generally identified as occurring when the diametre of nanoparticles was below 30 nm due to changes in crystalline structure or surface-to-volume ratio that enhanced their interfacial reactivity. However, the analysis appears not to be based on defined requirements for percent change. Another older review including other compounds indicates that changes in conduction bands and redox activity could occur at larger particle sizes (Gilbert and Banfield 2005). However, whether these properties are associated with a change in toxicological risk is still unknown.

When solid compounds are very small, complete translational symmetry disappears. Such atomic structures that can no longer be adequately described in the same terms as the bulk material are called “clusters” or “nanoclusters”. Typically, nanoclusters consist of only a few to a few thousand atoms, so their size may range from far below 1 nm to approximately 10 nm (Aiken and Finke 1999). The smallest full-shell metal particle clusters contain 13 metal atoms and the number increases by $10*n^2+2$, where n is the number of filled atom shells in the icosahedral symmetry.

However, it is clear that not all nanomaterials are crystalline or consist of clusters. Therefore, while crystalline phase and structures are very important properties for specific nanomaterials, they are not applicable to all. Thus, it is unclear how it could be included as an element in a definition.

### Redox potential

The redox potential is a measure of the tendency of an entity to lose or acquire electrons. Species that readily acquire electrons (reduction) have a high relative redox potential (e.g. Au3+) while those that lose electrons have a low redox potential (e.g. Na). By definition, the redox potential of a species is measured against a reference potential or reference electrode (e.g. hydrogen, calomel or Ag/AgCl, KCl). Measurement of the redox potential is meaningful for nanomaterials which can participate in electron transfer or uptake. Coating of nanomaterials may also participate or inhibit the redox activity of the nanoparticle.

While the redox potential may be useful in determining how active a given nanomaterial would be in human and environmental oxidation-reduction processes, it is unclear how this could be included as an element in a definition.

### (Photo)catalysis.

Photocatalytically active materials are semiconductors in which electron-hole pairs are formed upon exposure to light that generate highly reactive free radicals on the material surface. Titanium dioxide is a semiconductor of this kind.

Photocatalytic activity is highly material dependent. Within materials, it is size dependent. It can be enhanced or completely switched off by treating the surface of the material or by introducing dopants. Thus, while photocatalytic activity is very relevant for risk assessment, it is not a property that all nanomaterials will have. Due to this, it is unclear how it could be included as an element in a definition.

### Radical formation potential

The photocatalytic decomposition of water on the surface of a $TiO_2$ nanoparticle results in the generation of free radicals on the surface which in turn react with organic matter. This is an example of the potential of a given nanomaterial to generate free radicals. However while extremely valuable for the purpose of risk assessment, it is currently unclear how this measurand could be standardized such that it would be meaningful to consider it as an element in a definition.

### Zeta potential (surface charge)

Particles suspended in a solution may be charged and surrounded by an ionic cloud. This cloud is called the electrical (or interfacial) double layer. The double layer consists of the inner charged layer at the particle surface and a polarized stationary layer with opposite charge around the surface. Simplified, it can be said that the zeta potential is the potential between the dispersion medium and the stationary layer of fluid attached to the dispersed particle. The zeta-potential varies with the pH and salinity of the liquid.

The apparent zeta potential of a given nanomaterial is controlled by the coating and nanomaterials with different cores and shells that have the same outer shell exposed to solution will have the same zeta potential. While the zeta potential is useful for the purposes of risk assessment, it is unclear how it could be included as an element in a definition.

### Water solubility

Solubility has already been included in one definition (Cosmetics Directive) in which it is associated with bio-persistence.

Of course, it is important to know the aqueous solubility and (bio)degradability of nanomaterials to assess their potential for accumulation. Insoluble, non degradable nanomaterials would have a high priority for risk assessment as (bio)persistence/accumulation may be associated with chronic hazardous effects.

The words "solubility" and "persistence" are often used to discuss the rate of 'degradation'. Complex multi-component systems often degrade to their constituent macromolecular/nanosized components. In principle, the stability of a nanomaterial drops with decreasing size.

There are IUPAC definitions for solubility, dissolution, solution, colloidal dispersion and colloid. However, these definitions pre-date recent advances in nanomaterial engineering and manufacturing. The current scientific literature appears to be unclear on what is implied when dissolution is used to describe what occurs when a nanomaterial is added to a solvent (IUPAC Gold Book. http://goldbook.iupac.org).

In some literature, nanomaterial solubility implies that the nanomaterial degrades to its molecular units in solution (e.g. Ag releasing Ag+ ions in solution) (Auffan et al., 2009). In other literature this would be termed degradation (Doty et al., 2005, Prasad et al., 2005). Dissolution has also been used to refer to individual nanomaterial units (nanoparticle, nanotube, aggregate etc.) dispersed in solution when the dispersion is indefinitely stable (Banerjee et al., 2005, Selvan et al., 2007, Lin et al., 2004). In the recent physics and chemistry literature, the terms solubility and dispersion are used interchangeably to refer to the latter situation.

This ambiguity is recognised in the current OECD Guidance manual for the testing of manufactured nanomaterials under the OECD Sponsorship Programme. This document states that "it must be recognised that solubility and dispersibility are not identical though the distinction can be difficult to recognise with nanomaterials". (ENV/JM/MONO(2009)20.)

The ambiguity on solubility creates potential interpretation problems when including solubility as an element in a definition. In addition, if water insolubility was added as an element, the final surface coating on a nanomaterial could determine whether it was considered to be a nanomaterial or not. For example, soluble nano-ZnO would not be considered to be a nanomaterial whereas insoluble coated nano-ZnO would be.

Octanol-water partition coefficient,

The octanol-water partition coefficient is only meaningful for nanomaterials that are considered to be water soluble. As can be seen in the preceding section, there is ambiguity on what is implied if a nanomaterial is described as being soluble.

While the partition coefficient is a useful parameter for risk-assessment, it is of limited use as an element of a definition as it is not applicable to a wide variety of nanomaterials.

In general, the properties that can be said to change with size are those which physicists call 'cooperative' such as electronic conductivity, light scattering (and its relation to absorbance), magnetism, and superconductivity. All these properties exhibit a characteristic 'dimension' which may be micrometres (for semiconductor 'quantum effects') or a few nm (for similar effects in metals). For these effects on metals and semiconductors (catalytic) reactivity depends on size, shape and atomic structure.

For certain nanomaterials a threshold around 20-30 nm was suggested to be associated with a change in physico-chemical properties (Auffan et al., 2009). For other compounds, changes in conduction bands and redox activity were observed at larger particle sizes (Gilbert and Banfield 2005). In addition, other physico-chemical properties were found to show a continuous effect when related to size increase or decrease (Wei et al. 2004, Siow et al. 2004, Herzer 1995).

In conclusion, various properties may change when sizes are decreasing depending on chemical composition. For various characteristics these changes may occur at different sizes. Although no specific characteristic can be attributed to a specific size the fact that such changes occur is evident. Most physico-chemical parameters mentioned may be important to know for risk-assessment purposes. However, whether they each individually show sufficient discrimination to have universal applicability to all nanomaterials appears doubtful.

### 3.3.6. Organic and inorganic nanomaterials

In recent literature, nanomaterials have been described in terms of ‘soft’ and ‘hard’ (Nalwa 2009). There is no reported strict definition or explanation for the terms. The terms attempt to differentiate nanomaterials broadly on the basis of their chemical nature and likely behaviour in the body.

Nanomaterials regarded as ‘soft’ are often biodegradable and non-biopersistent[6]. Those produced from natural organic materials in the form of self-assembled, processed, or encapsulated nanostructures are termed ‘soft’. Examples of ‘soft’ nanomaterials are liposomes, nano-emulsions, and biopolymer based nano-encapsulates.

Engineered nanomaterials that are largely inorganic and insoluble are termed ‘hard’. Those regarded as ‘hard’ are non-biodegradable and potentially biopersistent. Examples of ‘hard’ nanomaterials are metals, metal oxides, and carbon materials (e.g. fullerenes, nanotubes, fibres).

It should be realized that exceptions to these categories exist especially when we consider the more complex nanomaterials including a multitude of coatings. In many cases the terms 'hard' and 'soft' are oversimplifications for complex hybrid materials. The terms are mainly used for applications where nanomaterials are meant (or likely) to be in an intimate contact with a living organism, such as food, medicine and cosmetic products. However, the terms should not be confused with the terms ‘soft metal’ and ‘hard metal’ that are already used in chemistry – which refer to the mechanical strength of different metals.

In conclusion, although various descriptions are used for 'hard' and 'soft' nanomaterials, the most common use is a distinction based on the origin of the nanomaterials being organic or inorganic compounds, respectively. In general the inorganic (hard) nanomaterials such as metals and metal oxides are non biodegradable and potentially biopersistent. In this respect, the distinction between hard and soft nanomaterials is not absolute as certain persistent nanomaterials may be organic (carbon nanomaterials) and certain oxides may be (bio)degradable.

As a result of the above rationale, terms like 'hard' and 'soft' as markers of potential biopersistence would not be useful in the definition.

### 3.3.7. Nanocomposites

According to IUAPC (2007), a nanocomposite is a composite in which at least one of the phase domains has at least one dimension of the order of nanometres. A composite is in turn defined as multi-component material comprising multiple, different (non-gaseous) phase domains in which at least one type of phase domain is a continuous phase.

Usually, nanocomposite is understood to refer to the combination of a bulk matrix and nanophase(s) (Ajayan et al. 2003). Examples include ceramic/metal nanoparticle composites, metal intercalated graphite, nanoparticle polymer composites, carbon nanotube/polymer composites etc. Certain properties of the nanocomposite are usually designed to be superior to that of the bulk matrix. For example, small amounts of carbon nanotubes can dramatically improve the conductivity and tensile strength of rubber. So, composite materials may have incorporated internal or external structures at the nanoscale to convert nanospecific characteristics to that composite. This has to be considered when describing a “nano”- material, structure and/or composite, even when the ultimate size of the material/structure/composite is much larger.

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[6] The term biopersistence may have different meanings. In relation to food, it will mean persistence in the body, whereas in other applications it may also mean environmental persistence. However, the main tenet is that biological systems can not deal with a biopersistent material through normal processes, e.g. digestion, metabolism, excretion, elimination etc.

It should be noted that such nanocomposites (e.g. car tyres with incorporated carbon black) would never normally be considered to be nanomaterials. However, as nanocomposites have an internal structure on the nanoscale, they would be considered to be nanomaterials according to many of the proposed definitions included in Annex I.

Certain nanomaterials and composite materials may have incorporated internal or external structures at the nanoscale to confer nanospecific characteristics to that composite. An example of such more complex nanomaterials are liposomes, which may be loaded with drug particles or metal particles that are used in medicinal and cosmetic applications, and can also be considered as nanocomposites. The next-generation hybrid nanomaterials are already in development and are typically based on the concept of a hierarchical assembly of many components (e.g. quantum dot superlattices, dendrimers, polymers) (Love et al. 2005, Daniel and Astruc 2004, Schmidt and Bodmeier 1999). These could also be considered to be nanocomposites. Any definition should be over-arching to ensure that it does not quickly become obsolete. Therefore, a reference to the internal structure should be included within the definition as this would then also capture complex assemblies provided the internal structure was within the specified range.

The manufacturing of nanocomposites has been foreseen by the definition of SCENIHR (2007) that defines a nanomaterial as "*being composed of discrete functional parts, many of which have one or more dimensions of the order of 100 nm or less*" (SCENIHR 2007). The definition of nanostructure mentions specifically the possible presence of internal and/or external functional parts (SCENIHR 2007). So, the definition of nanomaterial includes any material/structure/composite containing external or internal structures at the nanoscale.

In conclusion, including the internal structure at the nanoscale in a definition would include nano-composites provided the internal structure is within the specified size range. As the external dimensions of nanocomposites would be typically larger than 100 nm, most nanocomposites would not be considered to be nanomaterials with a definition based solely on external dimension. However, nanocomposites such as car tyres incorporating carbon black or tennis rackets incorporating carbon nanotubes should not be considered as nanomaterials. This should be addressed in the definition and exclusions for such types of nanocomposites should be considered.

It should be realized that during the production phase, when such nanomaterials are added to the product, and at the end of the life-cycle of these composite products (waste), there may be a risk associated with the nano-specific character of the materials used.

### 3.3.8. Persistence

Persistence can be defined as the property of continuation of existence of a chemical/material. Persistence or accumulation is considered a risk factor for hazardous effects in the long-term. Persistence is used primarily in a risk assessment context, to define chemicals or materials that are retained in the body or in the environment, although it could also be applied to durable products. Insoluble, non degradable nanomaterials would have a high priority for risk assessment as (bio)persistence/accumulation may be associated with chronic hazardous effects. In this respect persistence can be considered as the opposite of solubility or (bio)degradability (see 3.3.5).

With respect to the persistence of nanomaterials, a distinction needs to be made between:

- a nanomaterial characteristics which remains constant during the life-cycle
- a nanomaterial that changes during its life cycle but remains a nanomaterial

- a nanomaterial that is changed to a form no longer considered to be a nanomaterial but nonetheless one or more of its components is able to persist. This would include metals and Persistent Organic Pollutants (POPs). It should be noted that the classical criteria used to define POP's is lipid solubility (or a high octanol water partition coefficient) and steric hindrance of metabolism. Neither of these criteria is particularly relevant if the material is a solid.

At present there is very limited information to identify the most critical properties that are likely to lead to bioaccumulation or bio-magnification. However, ready uptake, bio-stability and poor clearance are likely to be the driving factors.

In conclusion, features associated with solubility (and degradability) of nanomaterials are very important for the risk assessment of nanomaterials in view of the possibility for persistence and accumulation both in man and the environment. These features include size and shape, water solubility, surface charge and surface reactivity. However, these features cannot be translated into an element of a definition as they are part of the characterisation of a nanomaterial and can change for each individual nanomaterial depending on chemical composition, surface modification and environment of the nanomaterial. Also, if persistence was included as an element of a definition, it would presume that information on which to base a decision on whether a given material was persistent or not was available. This will not be the case. In addition, it would link any defined nanomaterial with hazard and not all nanomaterials will have an associated (specific) hazard.

### 3.3.9. Manufactured versus natural

Three main categories of nanomaterials (nano-sized particles) can be distinguished, i.e. naturally occurring nanomaterials (e.g. gas-phase condensation products, ash, minerals, colloids), man-induced nanomaterials (by products of human activities like ultrafine particles from high-temperature processes such as combustion and industrial processes) and engineered or manufactured nanomaterials.

Many natural or synthesized materials may be used to produce nano-sized particles by size reduction (grinding, milling). In contrast there are carbon black and silica that are also extremely high production volume chemicals (millions of tons/pa) that have been chemically synthesized using bottom up methods for more than 60 years. All these materials will eventually gain access the environment, resulting in exposure of plants, animals and humans beyond the usual context of their normal presence.

Assessing the risks of all newly designed top-down and bottom-up nanosized materials is an exercise that needs to be performed carefully on a case-by-case basis. It is then, only then, possible to conclude, in the context of each proposed use, whether there are specific risk assessment needs. Within this context both the words "manufactured" and "engineered" are used to indicate the production of newly developed nanomaterials. Although differences are defined between engineered and manufactured nanomaterials (ISO/TS 80004-1:2010), the word "engineered" nanomaterials generally includes "manufactured" nanomaterials.

In order to designate specifically purpose-made nanomaterials, the term "manufactured" may be used. However, this term may not be sufficient to identify all produced nanomaterials. The meaning of "manufactured" must also include the processing of materials with the purpose to obtain materials at the nanoscale.

In conclusion, based on their origin, three types (natural, man-induced, manufactured) of nanoscale materials can be distinguished. As a result, a general definition should cover all these three types of nanoscale materials, the distinction being provided by the use of the qualifiers natural, man-induced and/or manufactured.

## 3.4. Conclusions

The various definitions for “nanoscale” and “nanomaterial” proposed so far have mainly sought to identify an inclusive size range that could be used to bring an increased understanding of the terminology that uses the prefix “nano”. Although ISO has taken a more elaborate approach recently using a general material hierarchy to define a series of core terms such as “nanoscale”, “nano-object” and “nanostructured material”, in past discussions SCENIHR preferred to use a more general framework based on existing terminology linked to an understanding of “nanoscale” with the aim of avoiding the proliferation of unnecessary terms. While both have their merits, it is clear that neither approach actually addresses the uncertainties that are associated when definitions of “nanomaterial” are requested for use as “enforceable terms” in a specific regulatory setting.

In addition to defining “nanoscale” in the context of “nanomaterial” it is also important to consider the key issues relating to both the interpretation and the uncertainties associated with any inclusive size range proposed as indicative of “nanoscale”.

This opinion considers several criteria in terms of their possible implications relating to the interpretation of the definitions of nanoscale and nanomaterial. It should be noted that in the metric system the “nanoscale” is the range below 1 micrometre (μm) and above 999 picometre (pm). Many previous reports (see Annex 1) have recommended an upper limit for a nanomaterial of approximately 100 nm and a lower limit of approximately 1 nm. Therefore, for one or more external dimensions, a working range for the nanoscale of 1 to 100 nm was used here to illustrate some of the important considerations relating to the definition of a nanomaterial and its measurands.

The key factors discussed were as follows.

**Size**

When considering any definition for nanoscale and nanomaterial it is evident that size is the predominant factor. Consideration was given as to what the term “size” refers to, as well as to the possible specification of upper and lower cut-off limits. However, within a regulatory context, size alone as measurand might not be sufficient as any size mentioned should be controllable and enforceable. This assumes that adequate methodologies are available. Several techniques are available for measuring at the nanoscale (i.e. below 1 μm), but the measurements obtained using these techniques are not always comparable. Therefore, not only is the size itself important but also the methodology used. For any accurate determination of size and size distribution at least two complementary methodologies should be used. Any nanomaterial should be described by its size and size distribution including the methodologies used for the measurement.

Size is the universal parameter that applies to all nanomaterials. It is evident that the size distribution is also an element that should be considered. Size was taken to refer to one or more external dimensions as this will capture most nano-objects such as plates or sheets, fibres and nanoparticles. However, solely referring to size as “one or more external dimensions” will not capture aggregates and agglomerates of primary particles nor, critically, more complex multi-component nanomaterials that are used in medicinal and cosmetic applications as their external dimension is likely to be larger than a specified upper size limit. Thus, a reference to the internal structure is also usually added to capture aggregates, agglomerates and complex assemblies. Based on the discussions, it seems that validated standard methods to determine size and its corresponding distribution are required.

In terms of the size limit, upper and lower cut-offs were considered. A lower size limit of 1 nm for one or more external dimensions is complicated by the fact that many molecules would then also be included and some nanoparticles may be excluded. Therefore, in any definition of a nanomaterial, molecules need to be generally excluded.

However, around 1 nm, the distinction between molecules, nanoclusters and nanoparticles is unclear. Therefore, exceptions should be made to allow for inclusion of certain specific entities such as fullerenes, dendrimers, graphene, clusters and complex hybrid molecular structures. In addition, there are tubes and fibres which may have a diameter below 1 nm and a length above 100 nm.

The upper size limit for one or more external dimensions of 100 nm is complicated by the potential exclusion of aggregates, agglomerates and multi-component assemblies that would have external sizes greater than this. The inclusion of a reference to "internal structure" with the same specified range as the external dimensions will include such materials within the scope of definition. However, this would also include nanoporous materials that are not usually considered to be nanomaterials. Therefore, some additional criterion is needed to take such materials into account.

Within a regulatory context, it seems useful to specify an upper and lower limit to facilitate a uniform interpretation. Equally, there seems to be a need for validated standardised methods to determine size and its corresponding distribution as these would ensure comparability of results. Also, the definition should aim at being over-arching so as to also include next-generation nanomaterials and avoid quickly becoming obsolete.

## Size distribution

The size distribution should be taken into account when defining and describing a nanomaterial. When only a fraction of the material has a size within the size range of the definition/description, the conditions for determining whether and when such a material will be considered a nanomaterial should be clearly described. This may be by allowing a part (certain %) of the number size distribution to be below a certain threshold or by using the information on the size distribution itself.

Using the number size distribution, materials might be defined as being a nanomaterial when more than 0.15% of the material has a size below 100 nm. The 0.15 % is derived from the mean plus/minus three times Standard Deviation (indicating 99.7% of the data set of measured nanoparticles). Assuming that the distribution is log-normal, this criterion is fulfilled if the mean of the $\log_{10}$-transformed sizes minus three times the Standard Deviation (of the $\log_{10}$-transformed sizes) is smaller than 2 (which is the $\log_{10}$ of 100 nm). However, it is recognised that nanomaterials may have other size distributions (e.g. bi-modal). The fraction of the material with a size below 100 nm can either be estimated from the measured size distribution in a sample or be measured.

The size distribution of a material should be presented as size distribution based on the number concentration (i.e. the particle number) and not on the mass concentration of a nanomaterial product as a small mass concentration may contain the largest number fraction.

To ensure comparability of results, validated standard methods are needed to determine both size and its distribution.

## Volume Specific Surface Area

The volume specific surface area (VSSA) is a complementary qualifier (criterion) to distinguish nanostructured materials from non-nanostructured materials based on its integral material surface area per material volume. For dry, solid materials, including agglomerates and aggregates, the VSSA can be estimated from the specific surface area as determined by the BET-method and the density. A limitation of the BET-method is that it is only applicable to powders and/or dry solid materials and not to nanomaterials embedded in solids and suspensions. Other methods exist that can be used for the analysis of both powders and particles dispersed in liquids but most still remain difficult to use on a routine basis. More developments are needed in the area of analysing powders and particles in liquids. For determination of the VSSA, the density of the material has to be known. In most cases, the density of the bulk material can be used. However, when there is no bulk equivalent of the nanomaterial, the density of the nanomaterial itself should be determined. It should be noted that not all nanomaterials are easily amenable to VSSA determination by the BET method (e.g. dispersions, nanocarriers etc.). Expressing the surface area related to the volume instead of mass allows for an additional criterion independent of the density and size or number size distribution of the nanomaterial. A VSSA above $60 m^2/cm^3$ would indicate an average size below 100 nm and thus a high nanomaterial content.

It should be noted that materials with a VSSA below $60\ m^2/cm^3$ may still contain a fraction with a VSSA above $60\ m^2/cm^3$ in view of the size distribution within the preparation.

### Surface modifications

Surface modifications were considered in the context of including specific properties as elements of the definition. There is a multitude of possibilities for the application of coatings and surface modifications of nanomaterials. Both deliberately applied and environmentally acquired coatings can have a major impact on nanomaterial interaction with biological systems. The coating and core together control the properties of a given nanomaterial and it is not useful to look at either the properties of the core or coating in isolation as they may not be representative of how the nanomaterial will behave in a given environment. Thus, each combination of a nanomaterial with a coating has to be considered as an individual case for safety evaluation. The variability in coatings on nanomaterials prohibit the feasibility of including elements based on specific surface properties within a definition as these properties may vary with coatings.

### Other physico-chemical characteristics

Several properties from the OECD WPMN list of physico-chemical characteristics that are considered to be relevant for the characterisation of nanomaterials for toxicological testing were evaluated as possible discriminators for the identification of a nanomaterial. These were crystalline phase, photocatalytic activity, zeta potential, redox potential, radical formation potential, water solubility and the octanol-water partition coefficient.

It was concluded that while all of these properties are very useful for risk assessment, none of them appears to meet the criterion of universal applicability required for a definition.

Solubility and degradability are highly relevant for the risk assessment of nanomaterials. Like any other material, nanomaterials can be degraded either mechanically, chemically or by dissolution; in fluids, they can form agglomerates or stable dispersions depending on solvent chemistry and surface coating. As for the other properties listed here, they crucially affect the behaviour of a nanomaterial of concern but they are not sufficiently over-arching to be included in a definition or to serve as a criterion for the definition of “nanomaterial”. They may however be valid criteria to assess the effect or the lifetime of a certain nanomaterial in different environments and its potential to release free nanoparticles.

It was also considered whether nanomaterials could be differentiated based on persistence or whether they are organic or inorganic. It was concluded that persistence is important for risk assessment, but, if it were included as an element of a definition, the definition would rely on information that may not be available. In addition, it would link any defined nanomaterial with the potential for a chronic hazardous effect while this would not be valid for all nanomaterials. Defining nanomaterials as either organic or inorganic was also considered to be relevant for risk assessment but this criterion would fail to capture hybrid nanomaterials.

In general, some of the properties that can be said to change with size are those which physicists call “cooperative” such as electronic conductivity, light scattering (and its relation to absorbance), magnetism, and superconductivity. All these properties exhibit a characteristic “dimension” which may be micrometres (for semiconductor “quantum effects”) or a few nm (for similar effects in metals). For these effects on metals and semiconductors (catalytic) reactivity depends on size, shape and atomic structure.

Various physico-chemical properties may change with decreasing size, depending on chemical composition. For the various characteristics discussed these changes may occur at different sizes. Although no specific characteristic can be attributed to a specific size, the fact that such changes occur is evident.

It may be important to know most of the physico-chemical parameters mentioned above for risk-assessment purposes. However, whether they each individually show sufficient discrimination to generally identify the wide variety of nanomaterials is doubtful.

### Composite nanomaterials

A nanocomposite is a composite in which at least one of the phase domains has at least one dimension of the order of nanometres. For example, complex nanomaterials such as liposomes loaded with metal particles that are used in medicinal and cosmetic applications may also be considered as nanocomposites. The inclusion of “internal structure” as an element of the definition is needed to include nanocomposites into the definition of nanomaterials.

However, nanocomposite also refers to a multi-phase material where at least one phase has dimensions of the order of nanometres (e.g. nanoparticles) and one phase is a continuous phase (bulk phase, e.g. polymer matrix). This interpretation of nanocomposite is specific for materials that include a bulk phase material (e.g. car tyres that have incorporated carbon black). It was noted that the inclusion of “internal structure” as an element of the definition would also mean that those nanocomposites would be defined as nanomaterials. Exclusion criteria would have to be developed to avoid considering macroscopic composite objects as nanomaterials. However, it should be recognised that the production and waste disposal of such products at the end of their life cycle could give rise to risks associated to the use of nanomaterials as internal structures in such composites.

### Manufactured, natural, by-product of human activity

Based on their origin, three main types of nanoscale materials (natural, by-products of human activity, and engineered/manufactured) can be distinguished. As a result, a general definition should include all three types of nanoscale materials, the distinction being provided by the use of the words natural, by-products of human activity or manufactured (engineered).

In order to designate more specifically purposely made nanomaterials within regulations, the terms “engineered” or “manufactured” may be used. However, the term manufactured may not be sufficient to identify all produced nanomaterials with a specific nano-risk for man and environment. The modification (processing) of materials (substances, minerals) by decreasing the size from bulk material into nanoscale materials may also pose an additional risk. As a result, the meaning of “engineered” or “manufactured” has to include the processing (grinding or milling resulting in size reduction) of materials with the purpose to obtain materials at the nanoscale.

In conclusion, a working definition based on size referring to one or more external dimensions or an internal structure with a specified upper and lower limit (100 and 1 nm, respectively) seems credible. Size is universally applicable to all nanomaterials and is the most suitable measurand. For regulatory purposes, the number size distribution should also be considered using the geometric mean size and its geometric standard deviation to refine the definition. Alternatively, a specific fraction of the number size distribution might be allowed to be within the specified size ranges of the definition.

For dry solid powders the volume specific surface area (VSSA) can be used as an additional criterion to identify a nanomaterial.

The size range specified should capture most nanomaterials although some challenges remain in terms of inclusions and exclusions. Such challenges can be posed by molecules around the specified lower limit, as well as by nanoporous materials and nanocomposites with respect to the internal structure.

Validated standardised methods for measuring size and its distribution would be needed to ensure comparability of results.

At the moment, no scientific data are available to indicate that a specific size associated with specific properties due to the nanoscale can be identified for nanomaterials in general. There is no scientific evidence in favour of a single upper limit. Although an upper limit of 100 nm is commonly used, there is no scientific evidence to qualify the appropriateness of this value. The use of a single upper value might be too limiting for the classification of nanomaterials and a differentiated approach might be more appropriate. This approach could be based on a relatively high upper threshold for which it is assumed that the size distribution at the lower end will most likely be above the lower, more critical threshold. The lower threshold of 100 nm would be the critical one for which extensive nano-specific information has to be provided in order to perform case-by-case risk assessment.

As there is no scientific evidence to qualify the appropriateness of the 100 nm cut-off, it is important to consider the whole nanoscale metric (1-999 nm). This could be enabled by applying a tiered approach using intermediate thresholds. For example, 500 nm could be used as high upper threshold and 100 nm as low upper threshold. In this case:

Category 1: size > 500 nm

> If the size (e.g. mean, median, etc.) of the material is above 500 nm it is assumed that the size distribution at the lower end will most likely be above the designated lower threshold of 100 nm. This should be confirmed by determination of the size distribution. Thus, the need for further evaluation regarding possible nano-specific properties may be of lower priority and for the moment classical risk assessment should be performed taking into consideration the particulate nature of the material.

Category 2: 500 nm> size >100 nm

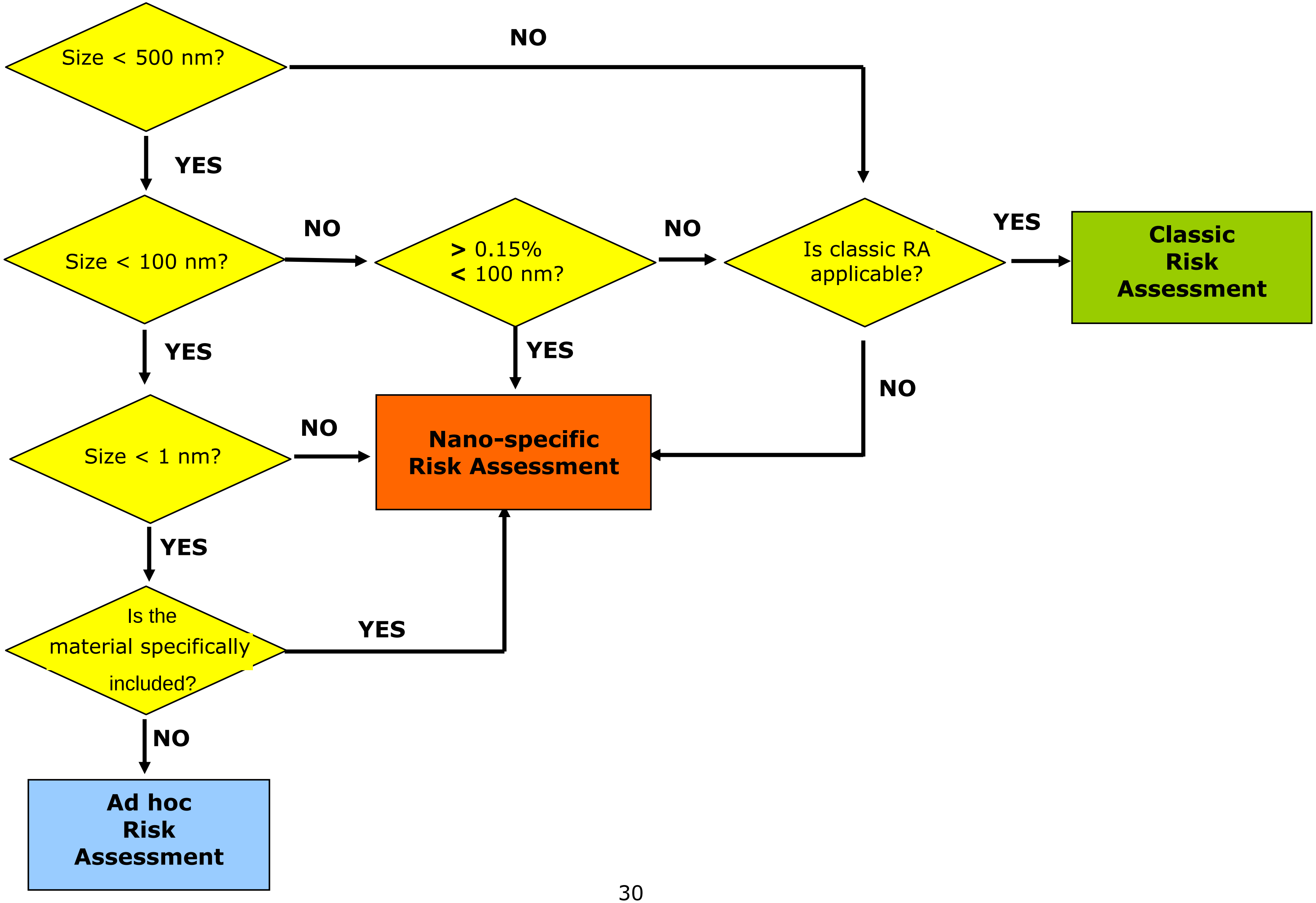
> When the size is <500 nm it is more likely that part of the size distribution will be lower than 100 nm and that a material may be considered a nanomaterial. Therefore a more detailed characterisation and nano-specific risk assessment will be necessary.
>
> A nanospecific risk assessment should be undertaken if the characterisation demonstrates that >0.15% (or any specified percentage) of the number size distribution is <100 nm. If these characteristics are not met, the need for further evaluation regarding possible nano-specific properties may be of lower priority and thus for the moment classical risk assessment should be performed taking into consideration the particulate nature of the material.

Category 3: 100 nm> size >1 nm

> The material is considered a nanomaterial and nanospecific risk assessment has to be performed. A VSSA above the threshold (e.g. >60 $m^2/cm^3$) may be used as an additional qualifier to indicate a size below 100 nm.

Figure 1 provides a schematic description of this approach.

**Figure 1: The use of size to identify an approach for the risk assessment of nanomaterials**



## 4. OPINION

This opinion has tried to address the complexity and the uncertainties and provide advice on the essential scientific elements for a working definition for the term "nanomaterial" for regulatory purposes using specific examples that may be applicable to certain classes of nanomaterial.

With the expected increase in the applications of nanotechnology there is an urgent need to identify by clear unequivocal descriptions what can be considered a nanomaterial and what should not be. This need to identify a nanomaterial and nanotechnology products comes from the uncertainty regarding the risk assessment and safety evaluation of nanotechnology products or, more specifically, the nanomaterials themselves.

It should be stressed that "nanomaterial" is a categorisation of a material by the size of its constituent parts. It neither implies a specific risk nor does it necessarily mean that this material actually has new hazard properties compared to its constituent parts. However, size will influence biodistribution (and distribution kinetics) in an organism or in an ecosystem and thus has an impact on the risk assessment of nanomaterials. One should also consider whether the definition should aim at being over-arching so as to include next-generation nanomaterials and avoid quickly becoming obsolete. Although the toxicological profile of the chemical components of a given nanomaterial may be well known, there may be cases where its specific properties raise concerns about their specific potential to harm humans and the environment.

In general, the risk assessment methodology currently used is also applicable for nanomaterials as it allows for sufficient flexibility. Although most of the existing toxicological and ecotoxicological methods for hazard identification are likely to be appropriate, they may not be sufficient to address all the hazards of nanomaterials. Therefore not all currently applied methodologies for hazard identification or exposure assessment of chemicals may be applicable to nanomaterials. The assays may need to be supplemented by additional tests, or replaced by modified tests, as it cannot be assumed that current scientific knowledge has elucidated all the potential adverse effects of nanoparticles.

Based on current knowledge, the Committee was invited to provide advice on the essential elements of a science-based working definition of "nanomaterials" and, specifically, to identify the most appropriate metrics to define materials at the nanoscale, taking into account:

- Reported size ranges and other relevant characteristics and corresponding metrics: The size ranges and other relevant characteristics (e.g., specific surface area, shape, density, spatial arrangements, aggregation, agglomeration, etc.) and corresponding metrics of materials reported as "nanomaterials" in the scientific literature;
- Characteristics: A first indication of possible characteristics and associated mechanisms that alone or in various combinations may lead to different properties;
- Physico-chemical properties: The physical and chemical properties that materials may show as a result of being at nanoscale or having a nanoscale structure;
- Threshold(s): The threshold(s) at which properties identified above may be expected to occur (the threshold(s) may be "below" or "above" depending on the relevant characteristic(s) and associated metric(s)).

There is sufficient evidence that there can be a change in some properties of the material at the nanoscale which is, for instance, due to the increase in surface-to-volume ratio. These nanospecific properties raise concerns over their potential to cause harm to humans and the environment. The chemical reactivity of nanoparticles often relates to the surface area. Consequently, the chemical reactivity per mass dose increases for smaller particles of the same type. This effect may or may not be associated with an increase in biological activity or toxicity. It is this uncertainty that warrants the careful evaluation of possible risks associated with nanotechnology products. However, although a change in properties may occur with a decrease in size at the nanoscale, and indeed many nanomaterials are specifically produced for such properties, it is at the moment not possible to identify a specific size or size range for nanomaterials at which a specific property in general would change or appear, nor a specific property that in general is introduced with size.

Several international and national organisations have proposed definitions for the nanoscale and nanomaterials. In most of the definitions proposed, the size refers to one or more external dimensions or an internal structure within a specified size range. The typical upper limit for the nanoscale is 100 nm, while the lower limit is either 1 nm, around 1 nm, or not indicated. The definition of the nanoscale as having a size between approximately 1 and 100 nanometre is commonly accepted, although: (i) there is no scientific evidence in favour of a single upper limit; (ii) there is no scientific evidence to qualify the appropriateness of the 100 nm value; (iii) the term "approximately" or "of the order of" cannot be used in a regulatory context; iv) it does not take into account that a nanomaterial will comprise a range of sizes; and v) the methodology needs to be adequate to support the applications of the definition. However, it is recognised that a number of measurement methods are currently available without being validated and specifically developed for nanomaterials. Some definitions have also included a reference to specific properties or nano-specific properties without identifying these properties.

This opinion provides advice on the essential scientific elements of a working definition for the term “nanomaterial” for regulatory purposes. The major question for both regulators and manufacturers is to identify when a material or product can be considered a nanomaterial. It should be noted that in the metric system the “nanoscale” is the range below 1 micrometre (µm) and above 999 picometre (pm). Criteria relevant for the discrimination between nano and non-nano are discussed using a working definition for nanoscale being approximately 1 to 100 nm. Any material with one or more internal or external dimensions in the nanoscale is then considered a nanomaterial. The feasibility of including specific properties as criteria was assessed.

With an overarching definition using the range of 1 nm to 100 nm, even with the addition of “engineered” or “manufactured”, such a terminology would include biological materials that are commonly used and processed and thus can be considered to be “engineered” or "manufactured" e.g. in the food and pharmaceuticals industry. Therefore, appropriate adaptations might be necessary within a regulatory context dealing with areas such as food/feed and pharmaceuticals.

When considering any definition for “nanoscale” and “nanomaterial” it is obvious that size is the predominant feature. Size, when used to refer to one or more external dimension will capture most nano-objects such as plates or sheets, fibres and equidimensional nanoparticles. However, within a regulatory context, any size mentioned should be controllable and enforceable. This requires that adequate validated methodologies are available. Several techniques are available for measuring at the nanoscale (i.e. below 1 µm). However, the measurements obtained using these techniques are not always comparable/consistent. Thus, not only is size itself important, but also the method used to measure it. Any nanomaterial should be described by its size and number size distribution including the methodologies used for the measurement.

Solely referring to size as “one or more external dimensions” will not capture aggregates and agglomerates of primary particles, and importantly, more complex multi-component nanomaterials that are used in medical and cosmetic applications as their external dimension is likely to be larger than a specified upper size limit. Therefore, the size of the primary particles should be determined for the identification of a nanomaterial. In terms of the size limit, the lower and upper cut-offs of 1 and 100 nm, respectively, were considered. However, a lower size limit of 1 nm for one or more external dimensions is complicated by the fact that many molecules would then also be included and some nanoparticles may be excluded. The upper size limit for one or more external dimensions of 100 nm is also complicated by the potential exclusion of aggregates, agglomerates and multi-component assemblies that would have external sizes greater than this. The inclusion of a reference to “internal structure” with the same specified range as the external dimensions will include materials with external dimensions above the upper size limit within the scope of definition. This would also include nanoporous and nanocomposite materials. Although this would include materials normally not recognised as nanomaterials, the production and waste disposal of such products at the end of their life cycle could give rise to risks associated to the use of the primary nanoparticles as internal structures in such composites.

To distinguish a nanostructured material from a non-nanostructured material, the volume specific surface area (VSSA) can be a complementary criterion, based on its integral material surface area per unit volume. For the calculation of the VSSA, the density of the material has to be known. For dry, solid materials, including agglomerates and aggregates, the VSSA can be estimated from the specific surface area as determined by the BET-method and the bulk density, a well known parameter. A limitation of the BET-method is that it is only applicable to powders and/or dry solid materials and not to nanomaterials embedded in solids and suspensions. Other methods exist that can be used for the analysis of both powders and particles dispersed in liquids but most still remain difficult to use on a routine basis. More developments are needed in this domain. In addition to the size considerations, a VSSA above $60 m^2/cm^3$ would indicate a nanomaterial with a size below 100 nm. Expressing the surface area related to the volume instead of mass allows for an additional criterion independent of the density of the nanomaterial. The VSSA is an integrated expression of the total surface area. Therefore, if a sample has a VSSA lower than $60 m^2/cm^3$, this does not exclude the possibility that a fraction of the material may have a size below 100 nm.

When describing a nanomaterial, data on the size distribution should be taken into account. It should be noted that a nanomaterial sample may have a particular size distribution (e.g. normal, log-normal, bi-modal). When only a part of the material has a size within the size range of the definition or description it should be clear whether and when such a material will be considered a nanomaterial. This may be by allowing a part (certain %) of the number size distribution to be below a certain threshold or by using the information on the size distribution itself. Using the number size distribution, materials might be defined as being a nanomaterial when more than 0.15% of the material has a size below 100 nm. The 0.15 % is derived from the mean plus/minus three times the Standard Deviation (indicating 99.7% of the data set of measured nanoparticles). The size measurements should be performed on the primary particles and their agglomerates/aggregates at optimal dispersed state. However, different distribution thresholds might be required for specific areas of application.

For nanomaterials the size distribution should be based on the number fraction and not on mass fraction, as a minimal fraction of the mass could contain large numbers in the low size range, while a low number of large sized particles would represent most of the mass. Also for dose-response effects, the hazard is associated more with the number of particles or the total surface area rather than the mass.

As size is a key element of a definition, there is a need to develop reliable and ultimately validated standardised methods to determine size and its corresponding distribution to ensure comparability in results.

There is a multitude of possibilities for the application of coatings and surface modifications to nanomaterials. Purposely applied and environmentally acquired coatings can have a major impact on nanomaterial interaction with biological systems. The coating and core together control the properties of a given nanomaterial and it is not useful to look at either the properties of the core or of the coating in isolation as they may not be representative of how the nanomaterial will behave in a given environment. So, each combination of a nanomaterial with a coating has to be considered as an individual case for safety evaluation. The variability in coatings on nanomaterials prohibits the feasibility of including criteria based on surface properties within a definition as these properties may vary with coatings.

Several properties from the OECD Working Party on Manufactured Nanomaterials (WPMN) list of physico-chemical characteristics considered relevant for the characterisation of nanomaterials for toxicological testing, were evaluated as possible discriminators for the identification of a nanomaterial. These were crystalline phase, photocatalytic activity, zeta potential, redox potential, radical formation potential, water solubility and the octanol-water partition coefficient. It was concluded that while all of these properties are very useful for risk assessment, none appears to meet the universal applicability criterion required for a definition.

Features associated with solubility and degradability are highly relevant for the risk assessment of nanomaterials in view of the possibility for persistence and accumulation both in man and the environment. These features include size, shape, hydrophilicity/hydrophobicity, surface charge and surface reactivity. Like any other material, nanomaterials can be degraded either chemically or by dissolution. In fluids, they can form agglomerates or stable dispersions depending on solvent chemistry and their surface coating. While these properties affect the behaviour of nanomaterials crucially, they are not sufficiently over-arching to be included in a definition or to serve as a criterion for defining nanomaterials. They are part of the characterisation of a nanomaterial and can change for each individual nanomaterial depending on chemical composition, surface modification and the immediate environment of the nanomaterial. They may however be valid criteria to assess the effect or the lifetime of a certain nanomaterial in one or the other environment and its potential to release free nanoparticles.

Most physico-chemical parameters mentioned may be important to know for risk-assessment purposes. Whether they each individually show sufficient discrimination to generally identify the wide variety of nanomaterials is doubtful.

The differentiation of nanomaterials on the basis of whether they are inorganic or organic was also considered. Certain persistent nanomaterials may be organic and certain oxides may be (bio)degradable. It would also not capture hybrid nanomaterials with inorganic and organic components. Thus, terms like inorganic nanomaterial as marker for potential biopersistence would not be a useful criterion to include in a definition.

Certain nanomaterials and composite materials may contain internal or external structures at the nanoscale that were incorporated to confer nanospecific characteristics. The internal structure with a size at the nanoscale would be an element to include in a definition, as then nano-composites will be included in the definition of a nanomaterial. As the external dimensions of nanocomposites would be typically larger than 100 nm, a definition based solely on external size would not consider most nanocomposites to be nanomaterials.

There are nanocomposites consisting of one bulk phase to which nanomaterials are added. It was noted that the inclusion of "internal structure" as an element of the definition means that those nanocomposites would be defined as nanomaterials. Exclusion criteria would have to be developed to avoid considering macroscopic composite objects as nanomaterials. However, it should be recognised that the production and waste disposal of such products at the end of their life cycle could give rise to risks associated to the use of nanomaterials as internal structures in such composites.

Based on their origin, three types of nanoscale materials (natural, by-products of human activity, engineered) can be distinguished. As a result, a general definition should include all three types of nanoscale materials, the distinction being provided by the use of the words natural, by-products of human activity or manufactured (engineered).

In order to designate more specifically purposely made nanomaterials within regulations, the terms "engineered" or "manufactured" may be used. When considering the purposely made nanomaterials, the meaning of "engineered" or "manufactured" also needs to include the processing (e.g. grinding or milling resulting in size reduction, chemical processing) of materials to obtain materials at the nanoscale.

In conclusion,

- whereas physical and chemical properties of materials may change with size, there is no scientific justification for a single upper and lower size limit associated with these changes that can be applied to adequately define **all** nanomaterials;

- there is no scientific evidence for a single methodology (or group of tests) that can be applied to **all** nanomaterials ;

- size is universally applicable to define all nanomaterials and it is the most suitable measurand. Moreover, an understanding of the size distribution of a nanomaterial is essential and the number size distribution is the most relevant consideration;

A defined size range would facilitate a uniform interpretation. For regulatory purposes the number size distribution should also be considered for further refinement of the definition. Alternatively, a specific fraction of the number size distribution might be allowed to be within the specified size ranges of the definition. For dry powders, the volume specific surface area (VSSA) may be added to the size to identify nanomaterials. In addition, the definition should include both external and internal nanostructures.

For the lower limit of the definition of nanomaterials, the size of 1 nm is proposed. However, around 1 nm, the distinction between molecules, nanoclusters and nanoparticles is unclear. In general molecules should be excluded. However, exceptions should be made to allow for inclusion of certain specific entities such as fullerenes, dendrimers, graphene, clusters and complex hybrid molecular structures. In addition, there are tubes and fibres which may have a diameter below 1 nm and a length above 100 nm.

At the moment, no scientific data are available to indicate that a specific size associated with specific properties due to the nanoscale can be identified for nanomaterials in general. There is no scientific evidence in favour of a single upper limit. However, an upper limit of 100 nm is commonly used. There is no scientific evidence to qualify the appropriateness of this value. The use of a single upper value might be too limiting for the classification of nanomaterials and a differentiated approach might be more appropriate. This approach could be based on a relatively high upper threshold for which it is assumed that the size distribution at the lower end will most likely be above the lower, more critical threshold. The lower threshold of 100 nm would be the critical one for which extensive nano-specific information has to be provided in order to perform case-by-case risk assessment.

As there is no scientific evidence to qualify the appropriateness of the 100 nm cut-off, it is important to consider the whole nanoscale metric (1-999 nm). This could be enabled by applying a tiered approach using intermediate thresholds. For example, 500 nm could be used as high upper threshold and 100 nm as low upper threshold. In this case:

Category 1: size > 500 nm

If the size (e.g. mean, median, etc.) of the material is above 500 nm it is assumed that the size distribution at the lower end will most likely be above the designated lower threshold of 100 nm. This should be confirmed by determination of the size distribution. So, the need for further evaluation regarding possible nano-specific properties may be of lower priority and thus for the moment classical risk assessment should be performed taking into consideration the particulate nature of the material.

Category 2: 500 nm> size >100 nm

When the size is <500 nm it is more likely that part of the size distribution will be lower than 100 nm and that a material may be considered a nanomaterial and that therefore a more detailed characterisation and nano-specific risk assessment will be necessary.

A nano-specific risk assessment should be undertaken if the characterisation demonstrates that >0.15% (or any specified percentage) of the number size distribution is <100 nm. If these characteristics are not met, the need for further evaluation regarding possible nano-specific properties may be of lower priority and thus for the moment classical risk assessment should be performed taking into consideration the particulate nature of the material.

Category 3: 100 nm> size >1 nm

The material is considered a nanomaterial and nano-specific risk assessment has to be performed. A VSSA above the threshold (e.g. >60 $m^2/cm^3$) may be used as an additional qualifier to indicate a size below 100 nm.

For specific areas and applications, modifications of any overarching definition may be needed due to specific requirements regarding risk assessment for regulatory purposes.

## 5. MINORITY OPINION

None

## 6. COMMENTS RECEIVED DURING THE PUBLIC CONSULTATION

A public consultation on this opinion was opened on the website of the EU non-food scientific committees from 12 July to 15 September 2010. Information about the public consultation was broadly communicated to national authorities, international organisations and other stakeholders.

In total, 94 contributions were received of which seven were from public authorities, thirty nine from businesses, seven from trade associations, four from academia, eight from individuals, fourteen from NGOs and fifteen others (e.g. private or international organisations). Several submissions notably those from industry and the NGOs were identical

Each submission was carefully considered by the Working Group and the scientific opinion has been revised to take account relevant comments. The literature has been updated with relevant publications. The scientific rationale and the opinion section were clarified and strengthened in certain respects. The opinion, however, remained essentially unchanged.

## 7. ABBREVIATIONS

| | |
|---|---|
| **AFM** | Atomic force microscopy |
| **BET Method** | Nitrogen absorption methodology named after Brunauer, Emmett and Teller |
| **BSI** | British Standards Institute |
| **CAS** | Chemical Abstracts Service |
| **CMC** | Chemistry, manufacturing and controls |
| **CNRS** | French National Centre for Scientific Research |
| **D** | Diameter |
| **DLS** | Dynamic light scattering |
| **ECDC** | European Centre for Disease prevention and Control |
| **ECHA** | European Chemicals Agency |
| **EFSA** | European Food Safety Authority |
| **EMA** | European Medicines Agency |
| **EU** | European Union |
| **FDA** | (United States) Food and Drug Administration |
| **FDA-CDER** | FDA Center for Drug Evaluation and Research |
| **FERA** | The Food and Environmental Research Agency |
| **IUPAC** | International Union of Pure and Applied Chemistry |
| **ISO** | International Organization for Standardization |
| **MAPP** | Manual of Policies and Procedures |
| **µm** | Micrometre |
| **nm** | Nanometre |
| **NP(s)** | Nanoparticle(s) |
| **NRCWE** | National Research Centre for the Working Environment |
| **OECD** | Organisation for Economic Co-operation and Development |
| **OECD WPMN** | OECD Working Party on Manufactured Nanomaterials |
| **pm** | Picometre |
| **POP(s)** | Persistent organic pollutant(s) |
| **ρ** | Bulk density |
| **REACH** | Registration, Evaluation, Authorisation and Restriction of Chemicals |
| **RIVM** | National Institute for Public Health and the Environment |
| **SCCS** | Scientific Committee on Consumer Safety |
| **SCENIHR** | Scientific Committee on Emerging and Newly Identified Health Risks |
| **SCHER** | Scientific Committee on Health and Environmental Risks |
| **SD** | Standard deviation |
| **SSA** | Specific surface area |
| **TEM** | Transmission electron microscopy |

| | |
|---|---|
| **VSSA** | Volume specific surface area |
| **$\mathrm{VSSA}_{\mathrm{fr}}$** | VSSA fractionated by size |

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ANNEX I

## Selected existing definitions relevant for nanomaterials

### ISO TC 229 Nanotechnologies

ISO ISO/TS 27687:2008 Nanotechnologies- Terminology and definitions for nano objects—nanoparticle, nanofibre and nanoplate.

The document lists various terms and definitions related to particles in the area of nanotechnologies. The definition described in the document for the nanoscale is:

> *Nanoscale: Size range from approximately from 1 nm to 100 nm.*

This definition is accompanied by two notes:

*Note 1: Properties that are not extrapolations from a larger size will typically, but not exclusively, be exhibited in this size range. For such properties the size limits are considered approximate.*

*Note 2: The lower limit in this definition (approximately 1 nm) is introduced to avoid single and small groups of atoms from being designated as nano-objects or elements of nanostructures, which might be implied by the absence of a lower limit.*

Currently a new series of technical specifications is being prepared dealing with the various subjects within nanotechnology such as: core terms, nano-object, nanomaterial, carbon nano-object, nanostructured material, bio/nano interface, nanoscale measurement and instrumentation, medical health and personal care applications, and nanomanufacturing processes. These subjects will be described in the ISO/TS 8004 series dealing with the vocabulary of nanotechnologies.

### OECD

Although not formally published in an OECD document, the OECD Working Party on Manufactured Nanomaterials (WPMN) has published a description of a nanomaterial on its website.

The OECD states *"A nanomaterial or a nanoparticle is usually considered to be a structure between 0.1 and 100nm (1/1,000,000 mm)"*[7]. At the nanoscale, the physical, chemical, and biological properties of materials may differ in fundamental and often valuable ways from the properties of individual atoms and molecules or bulk matter.

In a working document a more elaborate definition is published: OECD WPMN Guidance for the use of the OECD Database on Research into the Safety of Manufactured Nanomaterials. Ver.1 October 2008.

***"Manufactured nanomaterials:*** *Nanomaterials intentionally produced to have specific properties or specific composition, a size range typically between 1 nm and 100 nm and material which is either a nano-object (i.e. that is confined in one, two, or three dimensions at the nanoscale) or is nanostructured (i.e. having an internal or surface structure at the nanoscale)."*

This definition is based solely on size.

### SCENIHR

In its "Opinion on the scientific aspects of the existing and proposed definitions relating to products of nanoscience and nanotechnologies" (SCENIHR 2007) the SCENIHR describes some basic principles relating to the nanotechnologies and the nanoscale.

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[7] http://www.oecd.org/about/0,3347,en_2649_37015404_1_1_1_1_1,00.htm

*"(1) …. taking into account the need to avoid the promulgation of unnecessary terms and the requirements that it should be based on sound principles of lexicology. In view of the mandate of SCENIHR, this framework has been developed in the context of risk assessment procedures. Most of the concepts and behaviour patterns seen at the very small dimensions associated with nanotechnology are not new, and can be described by the existing terminology used at larger scales. It is recognised that it is impossible to stop individuals producing new words and definitions, but it is crucial that a new language is not adopted unnecessarily by the scientific community, and that on those occasions where it is required, it is consistent with established terminology.*

*(2) … Secondly, many of the terms used in nanoscience are based on commonly used words such as 'substance', 'matter' and 'material' and terms in nanoscience should not conflict with the general meaning of such words.*

*(3) The majority of terms that need to be considered in the context of nanoscience and nanotechnology are those that start with the prefix 'nano-', which specifically means a measure of $10^{-9}$ units, the nature of this unit being determined by the word that follows. There is absolutely no need to change the meaning of any scientific term, such as metre or material just because it is pre-fixed by 'nano-'.*

*(4) The majority of terms used in nanotechnology are broadly self-explanatory There are, however, some situations in which explanations are required in the development of a suitable framework for this terminology, especially for risk assessment purposes. "*

A clear statement was made that according to the metric system nanoscale actually means a size between 1 and 999 nm, being the size above picometre ($10^{-12}$) and below micrometre ($10^{-6}$). For the purpose of risk assessment of nanotechnology products, however, the nanoscale was limited to sizes of the order of 100 nm or less.

*Nanoscale: A feature characterised by dimensions of the order of 100 nm or less.*

In the opinion a framework was described involving a hierarchy of terms, principal of which is "nanoscale", which is considered here to be characterised by dimensions of the order of 100 nm or less. The framework builds on this concept of the nanoscale and develops a series of definitions, appropriate for risk assessment purposes, based on considerations of size, shape and properties. Key words defined in this framework include nanomaterial and nanoparticle, with particular emphasis on the limits to the nanoscale, the features that characterise a nanomaterial, the distinction between different geometric shapes at the nanoscale, and the potential for harm of released discrete free particles and/or their decomposition products. Within this scheme an engineered nanomaterial is defined as:

*Engineered nanomaterial: Any material that is deliberately created such that it is composed of discrete functional parts, either internally or at the surface, many of which will have one or more dimensions of the order of 100 nm or less.*

### EU Legislation containing a definition of "nano"

There is currently one EU regulation (Regulation EC/1223/2009 on Cosmetic Products) that includes a definition of nanomaterials. This text has foreseen a specific article (Article 2.3) to make a change in the definition possible depending on scientific and/or international developments.

Article 2.1.k of Regulation EC/1223/2009 on Cosmetic Products

*(k) "nanomaterial" means an insoluble or biopersistent and intentionally manufactured material with one or more external dimensions, or an internal structure, on the scale from 1 to 100 nm.*

Article 2.3 of Regulation EC/1223/2009 on Cosmetic Products

*In view of the various definitions of nanomaterials published by different bodies and the constant technical and scientific developments in the field of nanotechnologies, the Commission shall adjust and adapt point (k) of paragraph 1 to technical and scientific progress and to definitions subsequently agreed at international level. That measure, designed to amend non-essential elements of this Regulation, shall be adopted in accordance with the regulatory procedure with scrutiny referred to in Article 32(3).*

### EU legislation referring to size

In addition to a specific reference to nanomaterials, legislation also exists in which (non nanospecific) sizes of solid substances are included. The determination of size is specifically mentioned for solid substances/preparations when used as feed additives for animal nutrition. Such is the case for Commission Regulation (EC) No 429/2008 of 25th April 2008 on detailed rules for the implementation of Regulation (EC) No 1831/2003 of the European Parliament and of the Council as regards the preparation and the presentation of applications and the assessment and the authorisation of feed additives:

Annex II Section II**:** IDENTITY, CHARACTERISATION AND CONDITIONS OF USE OF THE ADDITIVE; METHODS OF ANALYSIS

2.1.5. Physical state of each form of the product

*For solid preparations data on particle size distribution, particle shape, density, bulk density, dusting potential and the use of processes which affect physical properties shall be provided. For liquid preparations, data for viscosity and surface tension shall be given. Where additive is intended to be used in water, the solubility or extent of dispersion shall be demonstrated.*

The statement is built upon scientific terms which are defined elsewhere.

### EU Legislation under discussion

Legislation for the regulation of novel foods is currently being prepared and has already been through a first reading of the European Parliament.

The European Parliament adopted the first reading of the legislation by a resolution on the 25th of March 2009, where Council agreement was obtained in June 2009.

European Parliament legislative resolution of 25th March 2009 on the proposal for a regulation of the European Parliament and of the Council on novel foods and amending Regulation (EC) No XXX/XXXX [common procedure] (COM(2007)0872 – C6-0027/2008-2008/0002(COD).

Article 3.2.f

*(f) "engineered nanomaterial" means any intentionally produced material that has one or more dimensions of the order of 100 nm or less or is composed of discrete functional parts, either internally or at the surface, many of which have one or more dimensions of the order of 100 nm or less, including structures, agglomerates or aggregates, which may have a size above the order of 100 nm but retain properties that are characteristic to the nanoscale."*

This text is partly adapted from the SCENIHR definition, but the adaptation may create some ambiguities because several aspects dealing with size have been included within this one definition (e.g. both nanomaterials below ("below" is included in the text above with the phrase "*or less*") and above 100nm). As in the existing legislation for cosmetic products, and also within the proposed legislation on novel foods, the possibility is included for a revision of the definition of "nano" based on scientific and/or international developments.

Article 3.3

*"In view of the various definitions of nanomaterials published by different bodies at international level and the constant technical and scientific developments in the field of nanotechnologies, the Commission shall adjust and adapt point (c) of paragraph 2 to technical and scientific progress and with definitions subsequently agreed at international level. That measure, designed to amend non-essential elements of this Regulation, shall be adopted in accordance with the regulatory procedure with scrutiny referred to in Article 14(3)."*

This definition combines size and non-specified properties that are characteristic to the nanoscale.

**Miscellaneous**

- FDA Nanotechnology. A report of the U.S. Food and Drug Administration Nanotechnology Task Force. July 25, 2007.

The FDA did not provide a clear definition on nanotechnology in its 2007 report, but opted for a broader approach to the subject.

*"The Task Force has not adopted a precise definition for "nanoscale materials, "nanotechnology" or related terms to define the scope of its work. The Task Force concluded that it would be most productive to take a broadly inclusive approach in identifying potentially relevant studies, data and other information.*

*The Task Force believes the FDA should continue to pursue regulatory approaches that take into account the potential importance of material size and the evolving state of the science. Moreover, while one definition for "nanotechnology," "nanoscale material," or a related term or concept may offer meaningful guidance in one context, that definition may be too narrow or too broad to be of use in another. Accordingly, the Task Force does not recommend attempting to adopt formal, fixed definitions for such terms for regulatory purposes at this time. As the FDA learns more about the interaction of nanoscale materials with biological systems and about general concepts that can inform the agency's judgement, it may become productive to develop formal, fixed definitions, appropriately tailored to the regulation of nanoscale materials in FDA-regulated products."*

- FDA Center for Drug Evaluation and Research (CDER) June 2010 MAPP 50159

The FDA-CDER has recently published a manual for chemistry, manufaturing and controls (CMC) reviewers within the Office of Pharmaceutical Science of the FDA. In this manual, designated Manual of Policies and Procedures (MAPP) 50159, a definition of a nanomaterial is included, describing a nanomaterial/nanoscale material as any material with at least one dimension smaller than 1,000 nm. The aim is to identify nanotechnology products and to enter the information obtained on the various nanomaterials into a nanotechnology database that may ultimately be used to develop policy regarding nanotechnology products. The use of the definition is limited to this Manual only dealing with reviews of pharmaceutical products.

- Reflection paper on nanotechnology-based medicinal products for human use EMEA/CHMP/79769/2006

In the EMEA (now EMA) reflection paper, nanomedicine has been defined as follows:

*"Nanomedicine is defined as the application of nanotechnology in view of making a medical diagnosis or treating or preventing diseases. It exploits the improved and often novel physical, chemical and biological properties of materials at nanometre scale*.

*Nanotechnology is a broad term, which covers a wide range of methods, tools and possible applications. There are a variety of definitions reported in literature, each generated for different purposes. For the purpose of this document, the definitions are based on those provided in the UK Royal Society and Royal Academy of Engineering report, the European Science Foundation foresight study on nanotechnology and the Vision paper and Basis for a strategic research agenda for Nanomedicine by the European Technology Platform on Nanomedicine:*

*Nanotechnology is defined as the production and application of structures, devices and systems by controlling the shape and size of materials at nanometre scale. The nanometre scale ranges from the atomic level at around 0.2 nm (2 Å) up to around 100 nm."*

This definition is solely based on size.

- Health Canada Interim Policy Statement on Health Canada's Working Definition for Nanomaterials

Health Canada considers any manufactured product, material, substance, ingredient, device, system or structure to be nanomaterial if:

*a. It is at or within the nanoscale in at least one spatial dimension, or;*

*b. It is smaller or larger than the nanoscale in all spatial dimensions and exhibits one or more nanoscale phenomena.*

*"For the purposes of this definition:*

*The term "nanoscale" means 1 to 100 nanometres, inclusive; The term "nanoscale phenomena" means properties of the product, material, substance, ingredient, device, system or structure which are attributable to its size and distinguishable from the chemical or physical properties of individual atoms, individual molecules and bulk material; and,*

*The term "manufactured" includes engineering processes and control of matter and processes at the nanoscale."*

This definition is based on size but the size range can be exceeded if there is evidence of non-specified "nano-scale phenomena".