TECHNICAL REPORT OF EFSA

Table of Public Comments on the EFSA Draft Nanotechnology Opinion[1]

Prepared by EFSA

(Question No EFSA-Q-2007-00228)

Issued on 5 March 2009

This technical report contains the comments received during the public consultation which was launched on 17 October 2009, and closed 1 December 2009 at 17:00 CET. At the deadline EFSA had received 208 submissions, from about 30 interested parties (individuals, non-governmental organisations, industry organisations and national assessment bodies). All comments received are tabulated with reference to the contributor and the section of the draft opinion to which the comment referred. There were a number of submitted comments received outside the electronic form which thus did not fulfil the EFSA submission criteria. However, those comments were still considered and have been manually inserted in the table below. Duplicate comments received from the same contributor appear only once and comments submitted by individuals on a personal capacity are listed anonymously. Comments submitted formally on behalf of an organization appear with the name of the organization. The line numbers mentioned in the comments refer to the draft opinion. A report on the outcome of the public consultation is published on the EFSA website http://www.efsa.europa.eu/EFSA/efsa_locale-1178620753812_1211902361968.htm.

[1] For citation purposes: Technical Report of EFSA on Table of Public Comments on the EFSA Draft Nanotechnology Opinion. *EFSA Technical Report* (2009) 236, 1-48

Section	Contributor	Comment
General comments	Bayer AG	We agree in general with the EFSA draft opinion which recognizes the risk assessment paradigm is applicable to ENM. The Risk Assessment methodology in place within the EU has proved reliable and adequate for a whole range of diverse chemical substances, and can be applied to nanomaterials. We welcome the case by case approach proposed by the authors. As with any new chemical substance, case-by-case considerations and expert judgment will play an important role in the risk assessment.
General comments	BLL	Concerning the above mentioned draft opinion BLL welcomes the initiative of EFSA and the consultation process. As a national member of CIAA BLL is supporting the general and specific comments CIAA made in its position paper. BLL appreciates very much that EFSA have mentioned in its draft opinion already the “Progress Report and Position Paper on Nanotechnology in Food Applications” dated March 2008. Meanwhile BLL has worked out a new version, published in September 2008 with new aspects concerning the progress in the field of definitions, in the field of food law and aspects of food technology. Please find the updated version attached. We ask you to verify and to change the document in your list of reference.
General comments	BEUC	This paper is a response to the EFSA’s public consultation on the ‘Draft Opinion of the Scientific Committee on the Potential Risks Arising from Nanoscience and Nanotechnologies on Food and Feed Safety’. EFSA draws attention to the gaps that exist in understanding the risks (human and environmental) posed by engineered nanomaterials (ENM) and highlights the need for more research which would be aimed at understanding how to assess and manage the risks they pose. EFSA also stresses the fact that a great deal of uncertainty exists around the potential risks that nanotechnologies’ applications may present. BEUC calls for: □ Agreement on a definition for engineered nanomaterials (ENM); □ More information to allow traceability & mandatory labelling of ENM; □ Creation of a European register of food/feed containing ENM; □ Further research on ENM to address the knowledge gaps which currently exist; □ Development of specific risk assessment methodologies for ENM; □ Application of the precautionary principle. The draft EFSA opinion focuses on engineered nanomaterials (ENM), describing their possible uses and applications in every step of the food chain (from production processes to agrochemicals, feed and food contact materials and food/feed ingredients). It highlights huge areas of uncertainty that surround nanotechologies and is clear in its recommendations that these knowledge gaps need to be addressed. While BEUC is aware that existing and foreseen applications of nanomaterials can offer benefits to consumers, we are also concerned about the potential for nanomaterials to have adverse effects (both short and long term) on human health and the environment which have so far not been adequately addressed at European level. In our view, knowledge gaps regarding the safety of nanotechnologies used in food should be addressed as a matter of urgency. It is clear that the following series of measures must be taken: [Comment Continue below]
General comments	BEUC	1. Agreement on a definition for engineered nanomaterials (ENM) It is clear that agreement on common definitions for what constitutes nanotechnology and ENM has to be reached in order to make it easier for information exchange on current and future commercial applications of ENM in the food and feed areas. We therefore call on EFSA, as a matter of urgency, to propose a common definition of what constitutes ENM. 2. Greater transparency on the use/potential uses of ENM According to the EU food industry, there are currently no foods using

		nanotechnologies or ENM on the market. The draft opinion highlights the serious need for information on what products using nanotechnologies are already on the market, in the pipeline or at the research stage. This would help in identifying those products which are most likely to be of greatest concern based on current understanding and would allow for an exposure assessment to be carried out. In addition, it must be ensured that any claims industry makes about their products containing/ not containing ENM are true. 3. Creation of a European register for food/feed products containing ENM In order to increase knowledge/information about the use of nanotechnologies in products, the opinion highlights the need for the creation of a register for food/feed products containing ENM at a European level. We would like to stress the importance of making the notification of the use of nanotechnologies mandatory for industry and to make the register available to the public. The register should also include those products which are available for purchase over the internet. In addition, we consider it crucial to require mandatory labelling of the nano-content of products with which consumers regularly come into direct contact such as food and cosmetic products. Consumer exposure to products notified should be examined and taken into account in the context of risk assessment and risk management measure developments.
General comments	BEUC	4. Further research on ENM EFSA draws attention to the gaps that exist in understanding the risks (human and environmental) posed by ENM and highlights the need for more research which would be aimed at understanding how to assess and manage the risks they pose. EFSA stresses the fact that a great deal of uncertainty exists around the potential risks that nanotechnologies' applications may present. It draws attention to the paucity of research and uncertainties regarding the characterisation, detection and measurement of ENM in addition to their toxicity, distribution, metabolism, absorption and excretion. We consider that, as a matter of urgency, more research should be undertaken on the potential risks for human health and the environment. The Commission and Member States need to increase research funding in order to ensure that uncertainties around health and environmental risks presented by some ENM are addressed. More studies are also required in order to determine the potential, long-term accumulation/persistence of ENM as current research on this area is extremely limited. The potential impact of ENM on human health and the environment should be examined via toxicological and eco-toxicological studies. 5. Specific Risk assessment EFSA concludes that existing risk assessment methods can be applied to ENM but must be performed on a case-by-case basis. It however states that 'due to the lack of sufficient data and information' it is difficult to give specific risk assessment guidance for the potential hazards of ENM. In our view EFSA needs to exercise caution when saying that the existing risk assessment paradigm is applicable for ENM whilst also stating that existing risk assessments may not be. EFSA must ensure clarity in their statements in order for them not to be misunderstood. We ask for consumer exposure to products containing ENM to be examined and taken into account in the context of risk assessment and risk management measure developments. We call for specific risk assessment methodologies to be developed and harmonised across the European Union. Mandatory guidelines for product development and marketing need to be defined and implemented. Above all, products containing nanomaterials should be subject to risk assessment to evaluate their potential effects on health and the environment before being put on the EU market. 6. Application of the precautionary principle Finally considering such a lack of certainty about the safety of ENM, we believe that the precautionary principle should be applied in the field of nanotechnologies, in particular in the food and feed product areas. A lack of complete knowledge and certainty about the safety of nanomaterials should not prevent regulators from taking precautionary actions. We regret that the

		opinion does not stress the need for regulators to start developing risk management measures and engage into risk communication even in the absence of a complete safety evaluation and risk assessment of nanoparticles. We believe that the EU needs to address the loopholes in regulations so that nano materials are included and there is clear guidance on how these regulations apply. In addition, the Scientific Committee should recommend ensuring that the use of ENM in food/feed is assessed and found to be safe before products can be placed on the EU market. Finally, the European Commission needs to ensure transparency and effective public engagement in order to examine public concerns, including areas of research considered as unacceptable, and ensure actions will be undertaken at an appropriate stage. For certain applications, product labelling requirements may be imposed to manufacturers to allow consumers to identify products which contain nanomaterials. Information and awareness-raising campaigns should be developed. Consumers have the right to receive clear and reliable information on the potential risks of nanomaterials threatening human health and the environment.
General comments	The Dr Hadwen Trust for Humane Research	The following comments are submitted in response to the public consultation on the Draft Opinion of the Scientific Committee on the Potential Risks Arising from Nanoscience and Nanotechnologies on Food and Feed safety and the Environment by the Dr Hadwen Trust for Humane Research. The Dr Hadwen Trust is the UK's leading medical research charity that funds and promotes exclusively non-animal techniques to replace animal experiments. Our vital work benefits humans with the development of more relevant and reliable science whilst also benefiting laboratory animals. We believe that excellence in medical research and testing can and should be pursued without animal experiments. Our organisation has 38 years' experience of funding high-quality, peer-reviewed and innovative research aimed both at advancing medical progress and replacing procedures on animals. We very much appreciate the opportunity to comment on this paper, and believe that as a research organisation dedicated to replacing animal tests (as well as the use of animals for other experimental purposes), our specific scientific expertise in the fields of toxicology and human health are relevant to this topic. We hope that our comments will be considered useful and constructive.
General comments	The Dr Hadwen Trust for Humane Research	We agree that the current uncertainties for risk assessment of nanotechnologies and their possible applications in the food and feed area, as well as in other areas of use, arise due to the presently limiting information in characterisation, detection and toxicology data. We are also aware of the lack of knowledge surrounding the current usage of engineered nanomaterials (ENM) and therefore exposure to such products is an area requiring immediate attention. Whilst recognising that the currently used risk-assessment paradigm is applicable for ENM our concerns regarding section 6 of the report (proposed guidance for risk assessment of ENM in food and feed area) centre around the acceptance of conventional toxicity testing methods to be used in identification of ENM hazards. We agree that additional issues specific to ENM need to be addressed due to the different properties displayed by ENM when compared to the bulk-form material and this will require testing method adaptation along with the treatment of ENM on a case-by-case basis. However, we do not agree that the current testing strategies are adequate for ENM and not represent the most scientifically robust methods to employ. As with the cosmetics sector, it will be extremely difficult in the food and feed industry to characterise ENM and current guidelines do not address ENM. Until methods are in place to properly determine the behaviour of ENM in living organisms and make careful and informed risk assessments, it would not be defendable for regulators and industry to assert that ENM in food or feed products

		are safe. We feel that it is more appropriate in the case of nanomaterials for companies to take a precautionary approach by avoiding exposing workers, consumers or the environment to these forms of substances. We do not believe clear commercial and societal drives to produce and market the many new and exciting nano-containing applications should overtake the fundamental requisite to protect human and environmental health and safety.
General comments	UK Food Standards Agency	• Our expert committess (ACNFP and COT) considered that the draft opinion generally provided a good summary of the available information, • Our commtitees strongly supported A case by case risk assessment based on functionality rather than size. The example cited to illustrate this was that there were 40 forms of single walled carbon nanotubes • It is disappointing that there is still such a paucity of basic toxicological data on nanomaterials • The drivers for the development of engineered nanomaterials could be more clearly portrayed to give a better appreciation of what types of product might reach the market • There should be recognition that a new generation of nanomaterials is in development which were intended to have specific biological properties, such as the ability to cross barriers • The opinion could more clearly acknowledge the importance of promoting public understanding in this area.
General comments	Nanotechnology Industries Association	General Comments o We, the NIA (in collaboration with the CIAA concerning some comments), agree with the general core of the EFSA Opinion, although it takes a very generic perspective, and would of course need to be specific to each application on a case-by-case basis. o While the European Commission requested 'an initial scientific opinion on the risks arising from nanoscience and nanotechnologies on food and feed safety and the environment', the EFSA Opinion focuses on 'engineer nanomaterials (ENMs)' only (see EFSA Opinion, line 188). The NIA welcomes this focus, and urges EFSA to ensure that this focus and appropriate terminology is maintained throughout the opinion. o All potential nano-applications should not be regarded as the same. What type of differentiation is EFSA recommending? Appropriate reflections are missing from the draft Opinion. o The paper is a literature review. It cannot, however, serve as instructions for the preparation of risk assessment dossiers, in the event of submission of applications. o We note the conclusion that the current usage levels of ENM in food and feed are not known. There is a misconception that the potential applications are already in use. This however appears to contradict the statement under 4.2.1, where it is said "In conclusion, significant consumer and animal exposure to ENM ingredients in food and feed is currently not likely within EU, though there may be exposure to nanoscale fractions within other materials. However, products are available via the Internet; this contribution to consumer exposure is not quantified". o We also welcome the conclusion that claims made concerning materials available in nanoform may not be true and cannot be routinely verified with the available analytical methods lines 271-274. o We note that the terminology "nanotechnologies" and "nanomaterials" are used interchangeably, while the Opinion addresses ENMs, only. The terminology should be consistent and appropriate throughout.
General comments	American Chemistry Council	December 1, 2008 The Nanotechnology Panel (Panel) of the American Chemistry Council: Comments on the European Food Safety Authority's (EFSA) Draft Opinion of the Scientific Committee on the Potential Risks Arising from Nanoscience and Nanotechnologies on Food and Feed Safety

		EFSA has launched a public consultation on its draft scientific opinion relating to nanoscience, nanotechnologies, and food and feed safety. This draft opinion focuses on engineered nanomaterials (ENM) that could be introduced into feed and food. It elaborates on approaches to risk assessment and as such is not an assessment of any specific application of ENM. In addition, the opinion appropriately states that it is generic in nature and is not in itself a risk assessment of nanotechnology, as risk assessments are likely to vary on a case by case basis depending on the material and application being evaluated. The Committee is applauded for providing a comprehensive overview of the current state of the science for risk assessment of nanomaterials in food and food-based applications. The Panel agrees with the Committee's recognition that the currently used risk assessment paradigm, which includes hazard identification, hazard characterization, exposure assessment, and risk characterization, are considered to be applicable for the assessment of engineered nanomaterials. Accordingly, current toxicity testing approaches used for conventional materials are a suitable starting point for the assessment of nanomaterials. We also recognize that these studies may need to be complemented with additional endpoints and with a more comprehensive material characterization. Efforts are underway through the Organization of Economic Cooperation and Development's (OECD) Working Party on Manufactured Nanomaterials to further explore the appropriateness of toxicology guideline studies for the assessment of nanomaterials. The data generated through this international effort should be invaluable in providing additional testing guidance for assessing nanomaterials. The Committee highlighted that available data on oral exposure to specific ENM and the associated potential hazard assessment are limited and that the majority of the data available are for other routes of exposure. Furthermore, the oral studies cited indicate that many ENMs were characterized only to a limited extent. The Panel agrees with the Committee that additional nanomaterial characterization will be a vital component of future toxicological evaluations, and the Panel strongly supports adequate material characterization as part of a toxicology study. Endpoints likely to be important for material characterization may include, but are not limited to: evaluation of size and size distribution, shape, purity, surface area and surface composition.
General comments	Scientific Committee of the Belgian Federal Agency for the Safety of the Food Chain	The document highlights the potential risks of ENM in food. At present, it is difficult to detect and quantify ENM. Moreover, only little is known about the dietary exposure to ENM and its impact on human health. This causes the document to be hypothetical and vague at some places. Nevertheless, it is necessary to carry out this exercise. The section on toxicology and toxicokinetics on the other hand, is more comprehensive and elaborated with concrete literature data. The document treats rather too unilaterally some aspects of nanoscience and nanotechnology. Primarily toxicology and toxicokinetics are discussed, two very important aspects that should be addressed when food safety of ENM is considered. Nanotechnology however, also offers many solutions to improve food safety such as, for example, a faster, better and cheaper diagnosis with low detection limits for all kinds of undesirable substances that may be present in food, which is not elaborated in the document. This argues for a title like "potential risks and benefits of nanoscience and nanotechnology on food and feed safety."
General comments	TNO Quality of Life, BU Quality & Safety, Zeist	We (toxicologists and risk assessors from TNO Quality of Life, BU Quality & Safety, Zeist, The Netherlands) would like to thank the EFSA Scientific Committee for preparing this draft opinion. There is a great need for a scientific opinion on the risk assessment of ENMs, clear definitions and recommendations on test strategies. We have some general and some specific comments on the draft opinion. These comments are submitted in the related sections.
General comments	The UK Government Chemist	As UK Government Chemist, it is part of my remit to provide advice on the dependencies between analytical science, policy, standards and regulation. It is reassuring to see that the Committee is acutely aware of the need to improve characterisation, detection and measurement of engineered nanomaterials (ENM), with the spotlight on complex matrices such as food, feed and biological tissues. By recognising the underpinning requirement for robust analytical methods, this important Opinion should

		help toward a wider adoption of realistic risk assessment approaches by the nanotechnology community.
General comments	CIAA	General comments •We agree with the general core of the EFSA Opinion, although it takes a very generic perspective, and would of course need to be specific to each application on a case-by-case basis. •All potential nano/applications should not be regarded as the same. What type of differentiation is EFSA recommending? Appropriate reflections are missing from the draft Opinion. •The paper is a literature review. It cannot however serve as instructions for the preparation of risk assessment dossiers, in the event of submission of applications. •We note the conclusion that the current usage levels of ENM in food and feed are not known. There is a misconception that the potential applications are already in use. This however appears to contradict the statement under 4.2.1, where it is said “In conclusion, significant consumer and animal exposure to ENM ingredients in food and feed is currently not likely within EU, though there may be exposure to nanoscale fractions within other materials. However, products are available via the Internet; this contribution to consumer exposure is not quantified”. •We also welcome the conclusion that claims made concerning materials available in nanoform may not be true and cannot be routinely verified with the available analytical methods lines 271-274 •We note that the terminology "nanotechnologies" and "nanomaterials" are used interchangeably. As the Opinion is only addresses ENMs, this should be consistent throughout.
General comments	Stora Enso Consumer Board	Nanotechnology has raised a big research interest and many projects on new nano solutions are going on. Same time consumers and green NGOs are concerned about the possible healthy risks of nanotechnology. Whenever there starts new projects on nanotehcnology and solutions, risk analysis and consumer health aspects should be included to the research. In addition, definitions on nanosized particles and nanotechnology needs clarification. Nanosize particels do not necessarily have any nanotechnological properties or contain any features of nanotechnology.
General comments	Bayerisches Staatsministerium für Umwelt und Gesundheit	Die "draft scientific opinion" der EFSA ist eine Zusammenstellung des bisherigen, begrenzten Kenntnisstandes zum Einsatz von technisch hergestellten Nanomaterialien in der Lebensmittelproduktion und deren Auswirkungen auf den Organismus. Es werden Schlussfolgerungen auf sehr allgemeiner Ebene gezogen und Empfehlungen für weitere Forschungsaktivitäten gegeben. Das Bayerische Staatsministerium für Umwelt und Gesundheit stimmt dem Entwurf uneingeschränkt zu. Ergänzend wird darauf hingewiesen, dass aufgrund der unzureichenden Erkenntnisse zum tatsächlichen Ausmaß des Einsatzes von Nanomaterialien in Lebens- und Futtermitteln dringender Handlungsbedarf dahingehend gesehen wird, von Seiten staatlicher Kontrollbehörden auf dem Markt befindliche Lebens- und Futtermittel auf das Vorhandensein von technisch hergestellten Nanomaterialien zu untersuchen. Hierzu müssen auf EU-Ebene zeitnah validierte Routinemethoden für den Nachweis und die quantitative Bestimmung von Nanomaterialien in Lebens- und Futtermitteln entwickelt und etabliert werden.
General comments	RIVM (National Institute for Public Health and the Environment)	Throughout the text, the distinction between the potential risks arising from the use of nanotechnology in general and the potential risks arising from ENM (engineered nanomaterial) containing products is not always clear. Since the use of nanotechnology in the production of food and feed, leads to different risks than the presence of ENM in food and feed products, making this distinction is crucial when talking about risks. Furthermore, the distinction between ENM that dissolves and ENM that persists within the GI-tract (as is made in L827-835) is also very important in risk assessment and should therefore also be included in the summary and recommendations. The same holds true for ENM which are intended to increase the bioavailability of incorporated substances.

		The distinction between these different ENM is also very important with respect to legislation and regulatory issues. ENM that persist within the GI-tract should be regarded as a new chemical identity, while ENM that dissolve or ENM that is intended to increase the bioavailability of incorporated substances, may be regarded as a new formulations of existing chemical identities. More reference should be made to the implications of the scientific knowledge on the potential risks of nanotechnology and ENM containing products for legislative and regulatory issues.
General comments	ILSI Europe aisbl	ILSI Europe welcomes EFSA's draft Opinion on nanotechnology. Important progress is made, e.g. distinguishing nanomaterials from dissolved chemicals and focusing on engineered nanomaterials (ENM) rather than natural nanomaterials. We also agree with the draft opinion that the current risk assessment paradigm is applicable for ENM. As the line below is not included in the sections referred to in the web comment form, it is included under General Comments: Line 152 "… that existing toxicological and eco toxicological methods may not be sufficient to address all of the issues arising from nanoparticles … " We tend to disagree, as we have many methods at our disposal that can be built into standard protocols, can be enhanced, and can be applied case by case as needed. Moreover, as argued above, this is not unique to ENM, but rather a function of the toxicological methods.
General comments	DSM Nutritional Products Ltd	As the same key issues are raised at numerous points throughout the EFSA Opinion; DSM Nutritional Products Ltd comment here on the key issues raised throughout the document as a whole, rather than on specific individual sections. In our comments we have attempted to draw together the key issues, and have made reference to the various points at which they appear in the EFSA text. We believe that EFSA were requested by the Commission to undertake 4 main tasks. Due to the character limitation of the field for submitting comments, we have provided our input sequentially in relation to these tasks. For a single document containing the comments please contact Comments Submitted by DSM Nutritional Products Ltd. DSM Nutritional Products Ltd welcomes the opportunity to comment on the Draft Opinion. The safety associated with exposure to nanoform materials is an extremely important issue. We are grateful for this initiative by EFSA and the Commission, and would like to take this opportunity to suggest that greater clarity is introduced into the document on a number of issues. EFSA have provided an opinion on the need for specific risk assessment approaches, for technologies, processes and applications of nanoscience and nanotechnologies in food and feed. We understand that in order to undertake this task EFSA were requested by the Commission to perform the following: 1. Identify the nature of potential hazards 2. Assess the appropriateness of risk assessment methods 3. Assess the potential risks associated with actual & foreseen applications 4. Provide guidance on the data needed for risk assessment

General comments	Deutsche Umwelt- u. Gesundheits-Initiative (DUGI) e.V.	Dear Ladies and gentleman, we from DUGI e.V. (Deutsche Umwelt- und Gesundheits-Initiative e.V.) / German Environmental and Health-initiative e.V., Non-Govermental, non-profit organisation, to push all activities for a sustainable future, is STRICTLY against ALL Nano-Technologie in Food and medicin !!!
General comments	VHUE e.V.	General comments: Dear Sirs, the german BfR has edited a study on "Nanotechnologie - Fortschritt mit Risiken, Anwendungen im Lebensmittelbereich" in March, 2006. Dr. Rainer Gürtler, the autor, mentioned on page 14: "that for example Siliciumdioxid-nanparticles may cause disfunctions of the cell; the replication as well as the transcription of DNA were reduced." For further information please see the document itself, being present in the internet ! Siliciumdioxid nanoparticles are used in nearly all medicine products ! Without knowing exactly what happens afterwards. I was really very concerned ! Yours sincerely, VHUE e.V. Association for the Support of Environmental sick People www.umweltbedingt-erkrankte.de
General comments	Private person	At this stage, I think it is very important to address the uncertainties and potential problems/risks and the research needs, however without “painting the devil on the wall” and thereby contributing to a similar attitude to “nano-” as one may see to “genetically modified –“ among segments of the population. I find the document well balanced. In particular, an aspect like food allergy not mentioned explicitly may be considered taken care of by the reference to “effects on the immune system” in the Opinion.
Summary	Bayer AG	Summary – lineS 50-52 “There may also be additional toxic effects caused by ENM that are not readily detectable by current standard protocols.” This statement is not substantiated by scientific facts and should be deleted from the summary:
Summary	Nanotechnology Industries Association	Summary o Line 15: Comment: The definition of ‘Engineered Nanomaterials’ (ENMs) should be included up front. o Line 24: Opinion text ‘Formulation at the nanosize changes the physico-chemical properties of a material [...]’ should read ‘Formulation at the nanosize may change the physico-chemical properties of a material […]’ o Line 28: Opinion text ‘Current uncertainties for risk assessment of nanotechnologies and its possible applications in the food and feed area […]’ should read ‘Current potential uncertainties for risk assessment of ENMs and their possible applications in the food and feed area […]’ o Line 30: Opinion text ‘[…], detect and measure ENM in food/feed and biological matrices […]’ should read ‘[…], detect and measure some ENMs in food/feed and biological matrices […]’ o Line 42: Comment: The available data on oral exposure to specific ENMs exists for existing substances! ENMs are not a substance class! o Line 60: Comment: Appropriate risk assessment should be conducted on a substance in the food and feed area. ENMs are not a substance, nor a substance class!

		Background as provided by the European Commission o Line 150: The word “bulk” is not appropriate in this context; the opposite of nanoscale is macroscale (see also throughout the Opinion)
Summary	American Chemistry Council	The Committee noted that there are currently a limited number of standardized reference materials (SRM) for ENMs, especially those with precise and reproducible detection and quantification of ENM in food feed and biological tissues. The silica SRM is noted from the Joint Research Center, Institute of Reference Materials and Measurements. It is important to note that the National Institute of Standards and Technology (NIST) is developing reference materials for environment, health, and safety (EH&S) research and metrology which, to date include gold nanoparticles at 10, 30 and 60 nanometers in diameter. The NIST group is developing additional reference materials which will assist in those conducting EHS assessments for a variety of ENMs. The Panel appreciates the opportunity to comment on the EFSA draft opinion on the potential risks arising from the use of nanomaterials in food and feed. We urge the Committee to encourage the adoption of adequate material characterization profiles as a component of all toxicological studies.
Summary	Scientific Committee of the Belgian Federal Agency for the Safety of the Food Chain	Line 15: It is too much a simplification to consider food and feed together (this is better presented on Line 196). Basic aspects (applications and potential impacts) for food and feed are not necessarily similar. For example, accumulation of nanoparticles in the organs of a chicken probably influences only little the health of the animal during his lifespan whereas human consumption of the chicken meat could affect human health in the long term. Carry-over (metabolization of ENM in animals, bioconcentration/-transfer, …) is an important topic to discuss. (On line 960 information generation of carry-over of ENM along the food chain is however, addressed as a recommendation.) Line 17 / line 876: it is not clear what is meant with ‘generic’? Rephrase or remove from sentence: “This opinion is generic in nature and is in itself not a risk assessment….”
Summary	CIAA	line 15: The definition of what ENM are should be included up front 24: Formulation at the nanosize MAY change the physico-chemical properties 28: for risk assessment of ENM 30: detect and measure “some of the” ENM 40: specific physicochemical properties 42: The available data exists for existing substances! ENM is not a substance class! 51: It is not clear why pharmacological endpoints would need to be addressed. This is not mentioned anywhere else in the document 60: risk assessment of a substance in the food and feed ENM is not a substance, nor a substance class!
Summary	EFFAT	After line 65 It widely recognised that scientific knowledge of real impact in of ENM in the food and feed or in food contact material for consumers, workers and the environment is lacking. Products incorporating or in contact with ENM are already manufactured and consumed without the necessary understanding of hazards and without a proper information of workers and consumers of presence of ENM in the food or in the FCM.
Summary	University of Bergen	In lines 29-32 of the summary of this opinion, the committee highlights the uncertainties involved in assessing the risks of nanotechnologies. These uncertainties relate to all aspects of engineered nanomaterials (ENMs) - characterisation, detection, measurement and toxicology. This essentially means that the opinion of the committee is that we don"t know what ENMs look

		like, have no good way of finding out where they are, can''t tell the quantities in which they might be present, don''t know all the characteristics that might make them harmful and can''t say if they will in fact be harmful. Despite this complete and utter lack of relevant information (and indeed the additional lack of agreed methods by which to gather such information), the opinion concludes in lines 37-39 that "the currently used risk assessment paradigm" is applicable. This is incomprehensible! When we do not have information or methods by which we can characterise, detect, measure, and/or understand toxicological properties, there is absolutely no way in which we can reasonably claim to be able to calculate risk. The paradigm is so completely flawed in this case as to be useless! While it is certainly the dominant paradigm for decision-making, for ENMs we must recognise its limitations and pursue creative alternatives. What we are doing in this case is not assessing risk, but negotiating uncertainty and this requires a different paradigm for decision-making. For nanotechnologies, we must understand that decision-making is as much about imaginaries as it is about information. In the face of extremely limited information by which we might be able to judge risks, we make decisions and move forward based on particular ideologies and images of the future, and it is these that need to be opened up for transparent negotiation and debate. It is a grave mistake to pretend that in this case decisions are being made on sound scientific assessments of risks, because our incomplete knowledge will inevitably lead to unintended consequences and the public will hold decision makers accountable for these if they claim to have made decisions based on an assessment of the risks. If, however, we are able to own the fact that decisions are made based on particular priorities, values, beliefs and visions for the future, the accountability for any unintended consequences becomes shared among those who share the pursued visions and values. Rather than claiming that decisions can be made on scientific risk assessment, we would do much better to have public negotiations around our socio-technical imaginaries and make decisions based on clearly articulated and socially negotiated desires for the future.
Summary	ILSI Europe aisbl	Line 34 - 36 (also line 872, line 894): ‘The limited database on ENM assessments should be considered in the choice of appropriate uncertainty factors in the risk characterisation step". It is not clear what this refers to - does this mean uncertainties in the exposure assessment, rather than uncertainties in extrapolation of animal data to human (for which various uncertainty factors exist for current risk assessment procedures). Lines 45-52: we do not agree with the sentence “However, the adequacy of currently existing toxicological tests to detect all aspects of potential toxicity of ENM has yet to be established.” The lack of adequacy of currently existing toxicological tests to detect all aspects of potential toxicity is related to the tests rather than to the materials tested. That is, if a test would not be adequate to detect ENM toxicity, it would also not be adequate to detect similar toxicity of non-nanoform materials. In its current form, the sentence suggests that ENM are more dangerous than other materials, which is not necessarily true. Therefore we would like this statement, and related statements in the draft opinion, to be modified to take account of this point. The document makes the argument for case by case risk assessment, hence toxicology, which makes sense, but then says that the adequacy of current tests is not yet established using (line 51) current standard protocols. That is exactly the reason that the protocols will need to be enhanced, e.g. the use of electron microscopy as well as routine. That is the meaning of case by case, so it is a mistake for the opinion to dwell so much on standard protocols. It might be useful to indicate in the Opinion which points to the best of the current knowledge need to be added to or emphasised in the standard protocols, such as toxicokinetics studies to specific organs such as the lymphatic system and the lungs. Also attention should be given to bioaccumulation potential, e.g. accumulation of certain degradation-resistant ENM in secondary lysosomes, which in cells with a long survival such as neurons or hepatocytes might lead to chronic toxicity.
Summary	DSM Nutritional	1.2 Hazard Studies

	Products Ltd	We agree with the statement that existing knowledge on chemicals cannot be fully extrapolated to ENM (789), but disagree that currently existing toxicity tests may not detect aspects of toxicity (47+853), and should only be seen as a 'starting point' (797). It is important to note that there is a great deal of experience with these methods in the case of non-nanoform materials, which were validated in some cases using materials of different structure from those that are routinely tested. Current testing methods were designed to identify unknown toxicities, and are fully applicable given that practical aspects such as dosimetry are properly considered. The EFSA Opinion states that 'adequacy of the existing toxicological tests….has yet to be established' (46+853), and that 'additional toxic effects caused by ENM….not readily detectable' (50); whilst not suggesting alternative methods. We agree that a 'case-by-case' approach should be taken, as indeed is the case with all risk assessments. We also agree that specific properties of ENM should be considered (900). However we are concerned that the Opinion might lead to an overly conservative approach and could be more practical. We believe the value of the existing methods to identify and characterise unknown toxicological hazards should be clarified: "Established toxicological test protocols are designed to investigate unknown toxicities. These tests are adequate to investigate the toxicity associated with nanoform materials given that exposure and dosimetry are carefully considered and a wide range of potential end-points are included."
Summary	VdMi	Background as provided by European Commission line 117 to 129: sounds like science fiction and should be added as an annex for interested parties. 150: The word "bulk" refers to material, which is transported by a lorry or in a silo. The opposite of nanoscale is macroscale (see also line 259) 164: Commisson asked for a risk assessment of ENM and not of the science cluster nanotechnology.
Summary	VdMi	line 28: for risk assessment of ENM 30: detect and measure "some of the" ENM 40: specific physicochemical properties 42: The available data exists for existing substances! ENM is not a substance class! 60: risk assessment of a substance in the food and feed ENM is not a substance, nor a substance class!
Assessment	American Chemistry Council	The Panel also agrees that current risk assessments for ENMs related to the food and feed areas should consider the specific properties of the ENM in addition to the properties of the non-nano form of the same material. The Committee has stated that generally it is not possible to satisfactorily extrapolate scientific data from non-nano materials and apply it to ENM. Consequently specific case by case risk assessments should be performed when assessing the safety of the ENM, based on data from relevant safety tests applicable to the particular ENM use. While the Panel agrees that information on the non-nano form alone many not be sufficient to evaluate the nanomaterial form, such information has value in assessing hazard and providing guidance to the overall hazard assessment. For example, if a particle elicits a specific toxicological response in its bulk form, this hazard-potential may also be relevant for the nano-form. This information may help prioritize the studies necessary to evaluate the nano sized material. In addition, although the focus of most nanotoxicology research has been on the increased toxicity of the nano-form, there are instances where the nano-form does not display increased reactivity or hazard

		potential. In these instances there may be a greater opportunity to extrapolate to the data on the bulk form of the material. The Committee opinion noted the difficulty of assessing the actual application of nanomaterials in the market. The Panel recognizes this difficulty which can be attributed in part to the use of “nano” as a marketing term for various products and applications. Therefore, care needs to be taken when making estimates as to the actual volume of these materials in the consumer market.
Assessment	CIAA	background as provided by the Commission line 117 to 129:sounds like science fiction and should be added as an annex for interested parties. 150:The word “bulk” refers to material, which is transported by a lorry or in a silo. The opposite of nanoscale is macroscale (see also line 259) 164:Commisson asked for a risk assessment of ENM and not of the science cluster nanotechnology.
Assessment	DSM Nutritional Products Ltd	1. Identify the nature of potential hazards 1.1 Scope We believe there are areas of confusion in the scope of the Opinion. For example, paragraphs 484 and 844 discuss issues that are not relevant to the risk assessment of nano-scale materials per se; namely exposure assessment to bioactive molecules. "Exposure assessment to bioactive molecules should not be part of an Opinion dealing with risks associated with physical size. If necessary this issue should be dealt with more comprehensively elsewhere." We believe EFSA should better define what is within and outside the scope of the definition of ENM. Not doing so could lead to unwarranted conservatism. Hazard is dependent on the material being assessed. In light of this fact it is important to note that nanoforms of different materials will not elicit the same effects, and therefore should be considered separately. We therefore welcome the recognition that the specific properties of ENM should be considered as an additional factor, further to those arising from the inherent properties of the same material in non-nanoform dimensions. ENM have been defined as any material deliberately created that is nanoscale in at least one dimension, generally but not rigorously defined as 100 nm or less. The properties of ENMs within living organisms, that are not seen with other forms of the same material, are unusual kinetic behaviour and an ability to illicit an inflammatory response in high dose studies (possibly due to oxidative properties). This response is due either solely or partly due to an increased number of molecules available for contact with the biological system (surface area), in addition to surface molecules with covalent bonds that are at strained angles or otherwise in a higher than normal energetic state. In all cases it is solid persistent particles (insoluble or not dispersible into molecular components) that are associated with these newly observed effects. Particles that are not recalcitrant to degradation following absorption, have not been shown to present these effects, and would not be predicted to do so.

		The Opinion states (336) that a dissolved chemical of the same material does not present the effects potentially associated with the nanoform. However, included in the scope of the Opinion are nano-sized and nanoencapsulated ingredients (288), and there is a recognition that nanotechnologies may be used to improve nutrient delivery (217). It is possible that some novel forms of minerals may be persistent when given in the nanoform, all other nutrients are soluble or dispersible in biological systems. When delivering a nutrient to a person or animal, it is the intention to deliver it systemically or make it available to the intestinal contents. Therefore carrier systems are also designed to be dispersible. As the Opinion states that ENM are absorbed to a limited extent from the GI tract (631), it is unlikely that a nutrient mix designed to be dispersible will cross into the circulation without first being degraded. In the case that this did occur systemic half-life of the intact particle would be very limited due to physiological processes. In the guidance (830) it is stated that the demonstration of disappearance of nanostructure in the lumen of the GI tract, is required to allow a risk assessment to be based on the chemical, and not its structure. If this cannot be proven kinetic studies are required. However this presents a clear difficulty as kinetic studies cannot usually differentiate between nanoform and dissolved material in any complex matrix (e.g. gut lumen or blood). These issues would be resolved if the guidance was clarified as follows: "Nano-sized and nanoencapsulated nutrients that are designed to readily disperse into their molecular constituents should not be assumed to present the unique hazards associated with persistent solid nano-materials."
1. Introduction to the opinion	UK Food Standards Agency	Given the broad mandate from the Commission, it might be useful to begin by describing the broad range of nanotechnology applications in food, before explaining why EFSA decided to focus only on ENM (engineered nanomaterials).
1. Introduction to the opinion	Nanotechnology Industries Association	Assessment • 1. Introduction to the Opinion o Line 188: Comment: The introduction states that 'this opinion focuses on engineered nanomaterials (ENM) that are deliberately introduced into the food chain […]'. This definition of the Opinion's scope is important and should be followed throughout; this has not bee the case, as a large part of the Opinion discusses the use of ENMs in 'Food Contact Material'. (see below, line 268-317) o Line 190: Comment: If 'size' is considered a nanoscale property, many 'natural' products will fall under the definition of 'deliberately engineered to have nanoscale properties' (cf. homogenisation processes, extraction/purification processes, etc.). Clarification is needed on EFSA's definition and use of the term 'natural', as well as 'engineered'.
1. Introduction to the opinion	Cefic Food Contact Additives	General comments: CEFIC FCA, European Chemical Industry Council and Plastics Europe welcome the Draft Opinion on the Potential Risks Arising from Nanoscience and Nanotechnology as an opportunity to start discussing with the European Food Safety Agency on the new

potential issues raised by the new nano-structured materials in the context of food and feed.

As a first comment we would like to recommend to EFSA and competent authorities in general to use the “nano” definition outlined in ISO TS 27682. This will certainly ease understanding and communication. Indeed, the terms used in the draft are different, and may bring some misunderstanding into the issue.

CEFIC does not recognise the nano-structured materials as a new class of substance for the following reasons:
-The physicochemical properties of a nano-structured material are highly dependant on its chemical substance.
-The potential toxicological and eco-toxicological properties are also extremely dependant on the chemical substance. Data exist on substance. They can be used to evaluate the nano-structured form of this very chemical substance.
As a consequence, a substance-by-substance approach should be the recommended approach.

To our knowledge the nano-structured material used in food/ feed are mainly integrated into the matrix and we consider that testing should be performed on commercially available materials and not on products developed for research specifically.

We need adequate standards and test methods developed at international level and therefore would recommend the draft opinion be completed by a specific action plan. As long these new methods to measure and evaluate the potential risks of the nano-structured materials are not tested and validated, we believe that the available OECD test methods should be applied to the nano-structured materials.

Specific comments:
p 2 – line 49-50 (see also Conclusions p 22 – line 910 - 911: There may also be additional toxic effects caused by ENM that are not readily detectable by current standard protocols…
The document will significantly improve when it is made clearer that the above statement is to be considered a hypothesis. No discussion is provided in the document itself on this hypothesis, and therefore its inclusion in conclusions and summary gives it unwarranted emphasis. Up till now no effects were reported in the peer reviewed literature that suggests that new mechanisms of toxicity do exist for nanoparticles. A main difference with bulk particles seems to be a difference in availability for uptake and translocation in the body.
p 10 - line 380-381 : .....dynamic "corona" .....Formation of protein corona around the Engineered Nano Material (ENM) is presented as an important hypothesis potentially influencing ENM kinetic behaviour (e.g. an influence on uptake and distribution). No indication is given whether these "corona" also exist around bulk material particles, and what experimental evidence specific to nanoparticles is available in support of this hypothesis.
p 10 - line 400 : It is important to measure the ENM in the matrix.....The aim should be to measure ENM in a number of selected standard matrices or simulants (as is done in food contact today but not in food as far as I know). This to avoid that every food type needs to be tested individually.
p10 line 444 : Due to the size of ENM – with a molecular mass well above 1000 g/mol, quite large in comparison with a molecule – the diffusion rates will be extremely low so that it can be reasonably expected that no migration of ENM from FCM to food occurs.
The reference FSA, 2008 is not listed in the reference list.

For further information please contact

1. Introduction to the opinion	Scientific Committee of the Belgian Federal Agency for the Safety of the Food Chain	Line 193: It is not clear why 'environmental' contamination of food/feed or the impact on plant health is not considered(*) whereas ENM contamination by pesticide and fertilizer residues is (both are more or less 'indirect' sources). Maybe some more clarification is needed why the first areas are not treated, whereas the latter can be treated together with direct ENM application on food/feed. (*) Notice that contrary to what is stated, the environmental aspect is glanced at in e.g. lines 459 – 464 where exposure due to environmental release is briefly discussed, and at point 5 (lines 775 - 786). Line 199: a more obvious second potential route of human exposure is the consumption of food of animal origin (that were feed with ENM containing feedstuff) rather than the carry over of ENM from feed to food (see also comment to Summary, line 15). It needs to be well explained what is meant exactly with 'carry over'.
1. Introduction to the opinion	FAO	Line 195: why not consider nanotechnolgy applications like new water purification systems (filters removing pathogens/contaminants) and soil cleaning systems (removal or catalysation of oxidation of contaminants), which might impact the food chain and indeed have some food safety implications?
1. Introduction to the opinion	AQUANOVA AG	line 190-192/line 254-258 The distinction between nanoscale organic formulations (like micelles) and nanoparticle made of inorganic materials is not clear enough. So we would kindly ask you for addition the scientific opinion according to the following BfR explanation "What is the link between liposomes, micelles or vesicula and nanotechnology?" (http://www.bfr.bund.de/cd/24949): "Organic compounds like liposomes, micelles or vesicles are used in foods to encapsulate other substances like vitamins or flavourings, to transport them around the body and release them in a targeted manner. As the size of these "transport containers" is frequently in the nanometre range, they are also called nanocapsules. However, in contrast to inorganic, insoluble nanoparticles, their nanoscalability does not lead to any new properties or, by extension, to any new biological effects. Hence, the use of nanoscale organic compounds is not classified as nanotechnology in the narrower sense by BfR. Organic substances like beta-cyclodextrin or polysorbates are frequently used for the capsule membrane. They are toxicologically tested and assessed, and are approved as food additives (E 459 and E 432 up to E 436)." Please see also the definition for engineered nanomaterials of the American Chemistry Council (http://www.americanchemistry.com/ s_acc/bin.asp?SID=1&DID=5090&CID=654&VID=109&DOC=File.PDF): "Proposal: An Engineered Nanomaterial is any intentionally produced material that has a size in 1, 2, or 3-dimensions of typically between 1-100 nanometers. It is noted that neither 1 nm nor 100 nm is a "bright line" and data available for materials outside of this range may be valuable. Buckyballs are also included even though they have a size <1 nm. Exclusions: 1. Materials that do not have properties that are novel/unique/new compared to the non-nanoscale form of a material of the same composition. 2. Materials that are soluble in water or in biologically relevant solvents. Solubility occurs when the material is surrounded by solvent at the molecular level. The rate of dissolution is sufficiently fast that size is not a factor in determining a toxicological endpoint. 3. For those particles that have a particle distribution such that exceeds the 1-100 nm range (e.g. 50-500 nm) if less than 10% of the distribution falls between 1-100 nm it may be considered as non an Engineered Nanomaterial. The 10% level may be on a mass or surface area basis, whichever is more inclusive. 4. Micelles and single polymer molecules." We are also missing the classification of the nanomaterials regarding the Novel Food Regulation (EC) No. 258/79. Please see the statement of the Official inspection of foodstuffs exemplary for our NovaSOL Q10 product:

		"Regarding the solubilisates in nanoscale dimension by the firm Aquanova, Darmstadt: Coenzyme Q 10 (ubiquinone) is a substance insoluble in water. In the product NovaSOL Q Coenzym Q 10 the water-insoluble ubiquinone is packed into micelles by adding amphiphilic substances. Since the micelle"s surface is polar, the homopolar character of ubiquinone is covered, and no aggregation (even in water) with other homopolar substances is possible. In this way the coenzyme Q10 is present in a colloidal solution. Biological cellular membranes constitute a double layer of lipid nearly impervious for polar substances. The transport of larger polar molecules takes places nearly exclusively over membrane proteins specific for the substrate. As a result, the polar micelles are very probably not transported over the cellular membranes of the intestine. Hence the intake is done only after the ubiquinone is again present in its free form. According to my opinion the packing of coenzyme Q10 in micelles does not lead to any modified metabolism in the intestine. Hence this does not concern, despite the unusual production procedure, a novel food pursuant to Regulation (EC) No. 258197 Article 1 Para. 2 Letter f."
1. Introduction to the opinion	Federal Institute for Risk Assessment	Line 188: Please check: "This opinion focus on …"
1. Introduction to the opinion	ILSI Europe aisbl	Lines 188-192 " "Natural" nanoscale materials (e.g. micelles) will be considered if they have been deliberately used or engineered to have nanoscale properties, or used e.g. to encapsulate bioactive compounds". Existing food additive such as lecithins (E 322), allowed generally for use in food at GMP levels, and polysorbates (e.g. E 433) allowed for some specific applications and among others for use in dietary supplements, would fall under the scope of this statement. They are deliberately (as all food additives) used to emulsify or solubilise food ingredients, e.g. a bioactive substance, by means of forming a nano-sized structure, i.e. micelles in case of polysorbate use and vesicular structures in case of lecithins. This can also be viewed as a micro-encapsulation process. In our opinion the sentence should not only be rephrased, but even more, the issue of such classical food additive applications which are presently authorised and since long times known to form nanoscale structures in food applications, should be considered somewhere in more detail in the document. It may otherwise be a source of confusion.
1. Introduction to the opinion	DSM Nutritional Products Ltd	2. Assess the appropriateness of risk assessment methods 2.1 Exposure Assessment We welcome the recognition that dosimetry for nanoparticles is complex as weight of dose may not be the most important parameter. In this respect the guidance given in paragraph 807 is helpful. We agree that due to the difficulties in analysing some ENM in the food matrix a conservative approach should be taken by assuming that all ENM added remains present in nanoform in the food prior consumption, unless it is positively identified to have dissolved / degraded. 2.2 Risk Assessment Paradigm We agree that the established process of identifying and characterising hazard, and comparing against exposure to characterise the risk is fully applicable to the assessment of ENM. We do not agree however with the suggestion that new additional uncertainty factors may be applied (873, 36 & 895). There is a long history in toxicology of using such factors as a surrogate for rational scientific assessment of risk (mainly by risk managers). Given appropriate modification of test methods and a rigorous approach to investigating the same product that will be marketed, there is no need to create and enshrine additional uncertainty factors in toxicological practice. "Instead of applying factors with no scientific basis; during the case-by-case approach to risk assessment, it can be decided if the margin of safety between observed effects and intended use is appropriate to ensure safety."
1. Introduction to the opinion	Food Safety Authority of Ireland	Line 188 "This opinion focus on..." should read "This opinion focuses on..."

2. Introduction to nanotechnologies in the food and feed area	Nanotechnology Industries Association	• 2. Introduction to nanotechnologies in the food and feed area o Line 210: Clarification is required on the meaning of the word 'lower' in '[…] with the common theme that they all involve
2. Introduction to nanotechnologies in the food and feed area	TNO Quality of Life, BU Quality & Safety, Zeist	Comment line 208-210: In this section nanotechnologies are defined as …a broad assemblage of processes, materials, and applications that span physical, chemical, biological, engineering and electronic sciences with the common theme that they all involve manipulation of substances at a size range in the (lower) nanoscale. This definition is interpreted as that materials/processes are used to manipulate material to nanoscale, which has specific accompanying beneficial effects. According to this definition, these processes are intentionally performed to obtain a nanoscale product with the accompanying properties. However, it may be possible that a manufacturer unintentionally produces a nanoscale product, for example as by-product in industrial processes. This product does not fall within the definitions set in the draft opinion. However, to our opinion it remains unclear whether a nanoscale product in this case will be considered an ENM and needs a specific risk assessment strategy. Comment line 231 – 267 "Terms used in the opinion": In the draft opinion several terms are described, but to our opinion no sound definition for ENMs is provided. Therefore, in case of a product safety or regulatory assessment, it is difficult to decide whether the material in question is to be regarded as an ENM and needs a specific risk assessment strategy. For example: "…nanoscale refers to a dimension of the order of 100 nm and below. Since the changes in characteristics that are seen on reducing dimensions do not occur uniquely at the 100 nm size, it is important that some latitude is allowed in this definition with respect to the meaning of "the order of" and it is recognised that there will be various borderlines. Generally, we are in the order of 100 nm or less, but there are size-related effects that can appear at larger size." In this part some space for interpretation is given on whether a material needs to be considered as an ENM. It is mentioned that size-related effects can appear at a larger size than 100 nm, but it is not mentioned whether materials below 100 nm can also be regarded as not-nanoscale. Furthermore: "…Food and feed may contain components that have internal structures that individually could be present at the nanoscale, e.g. naturally occurring molecules, micelles or crystals. However, as said above, natural components are considered as ENM within the context of this opinion, only if they have been deliberately used or engineered to have nanoscale properties or used e.g. to encapsulate bioactive compounds." Within the context above it is not clear what is exactly meant with 'nanoscale properties'. Finally: "Engineered nanomaterial … such that it is composed of discrete functional and structural parts…". Within this part of the definition for ENM it is noted that 'parts' can be interpreted as a component/particle in a larger structure (e.g. coating) as well as a physical part of the subject/particle. Taken this into account, guidance is needed what is meant by 'discrete functional and structural parts'. Considering the above, a clear definition of an ENM is needed.
2. Introduction to	FAO	Line 230:

nanotechnologies in the food and feed area		Given public concerns about new technologies applied to the food sector, it appears essential that safety challenges concerning the nanotechnologies are addressed early on, alongside public involvement in decision making about governance issues and research activities.
2. Introduction to nanotechnologies in the food and feed area	CIAA	line 188: This opinion focus on engineered nanomaterials (ENM) that are deliberately introduced into The concept of 'deliberately introduced' is important and should be applied throughout the report where appropriate. (see also for food contact material) 189: the food chain. Such ENM range from food contact material, ingredients and additives, to 191: (e.g. micelles) will be considered if they have been deliberately used or engineered to have Clarification needed on 'natural' nanoscale materials (e.g. micelles) 192: nanoscale properties, or used e.g. to encapsulate bioactive compounds. If one considers 'size' a nanoscale property, homogenised milk will fall under the definition of 'deliberately engineered to have nanoscale properties'. – clarification is needed. 210: Manipulation of substances at a size range in the (lower) nanoscale. This is subject to interpretation because both the size range is not defined and the lower nanoscale region is not specified. Clarification is neede heer. 211:Due to the small size of ENM no new unique properties arise. The properties are known, only the applications might be new. 221 to 226:It is not clear whether the focus is on nanotechnologies or ENM. This report should focus on ENM solely. The properties of manufactured nanomaterials depend on the technology used of the specific producer; i.e. the properties are product and production specific. Snow crystals change their form dependent on the temperature. 231:Terms used in the opinion The terminology used throughout the report like non-nanoform, non-nanoscale line 472, equivalent chemical counterpart line 917-919, equivalent non-nanoforms line901, conventional chemical, macroscale line 335 material and on the other hand nano-sized line 288,626 nanoscale, naturally occurring variant is not consistent line 409 nanoform line556. line 238:The ISO publication is a technical document and has been recently published as DIN ISO CEN TS 27687 without the chapter on nanostructured materials. The terms listed in the annex of this publication are not fully accepted by the standardisation bodies. 239-243: need further clarification or examples where it is not practical to use the prefix nano. As it is, it is confusing and subject to interpretation. 257: Change to "if they have been deliberately used or engineered to have properties/characteristics that are different from bulk materials. Clarification needed on 'natural' nanoscale materials (e.g. micelles), or engineered to have nanoscale properties, or used e.g. to encapsulate bioactive compounds. 259-261:macroscale material and dissolved chemicals are compared to nanoscale material throughout the opinion but the differences in properties between macroscale (beyond nanoscale) and dissolved chemical (smaller than nanoscale) is not indicated. 262-267:agglomerate and aggregate are defined but no relationship to nanomaterials is given at this point
2. Introduction to nanotechnologies in the food and	Federal Institute for Risk Assessment	Line 210: Reference may be made to ISO/TS 27687 Nanotechnologies - Terminology and definitions for nano-objects - Nanoparticle, nanofibre and nanoplate". Line 230: … before they are placed on the market.

feed area		Line 244-249: As in line 230, reference may be made to ISO/TS 27687 Nanotechnologies - Terminology and definitions for nano-objects - Nanoparticle, nanofibre and nanoplate“. Line 262-267: When distinguishing between agglomeration and aggregation, it may be help-ful for the unaware reader, to point out that agglomerates can deagglomerate, while aggrega-tion is usually irreversible. Various contradicting definitions of these terms exist, potentially causing misunderstanding between scientists from different disciplines.
2. Introduction to nanotechnologies in the food and feed area	RIVM (National Institute for Public Health and the Environment)	L218-220: Is it possible to include some examples to clarify what is meant by “the intersection between food medicines and cosmetic sectors”? L262-267: From this text the distinction between agglomerates and aggregates in terms of surface or volume ratio is not very clear. Please clarify.
2. Introduction to nanotechnologies in the food and feed area	ILSI Europe aisbl	The first paragraph misses the point that most food and feed and biochemical, physiological and pharmacological reactions take place in liquid phase at the nano level. This really is the control for ENM. In this opinion ENM sometimes refers to particulate material that indeed needs to be considered as a new situation, but sometimes ENM refers to soluble phase reactions that are part of normal physiology. It is important for the opinion to be very clear on what is meant by ENM. This is demonstrated in line 223 where the opinion mentions “the lack of knowledge about the potential effects and impacts of nano-sized materials…” – it is assumed that what it meant here is not all nano-sized materials but only insoluble particles, and this clarification should be provided.
2. Introduction to nanotechnologies in the food and feed area	DSM Nutritional Products Ltd	3. Assess the potential risks associated with actual & foreseen applications This was not explored in any detail in the Opinion. There is a short discussion of the concerns of public (224-230), this is an important issue that was not requested in the ‘terms of reference’ from the Commission. "The discussion of consumer concerns is not relevant in a scientific opinion and should be dealt with more comprehensively elsewhere. However in writing their Opinion, EFSA should have in mind that it will be interpreted by interest groups outside the food industry or scientific community." There is a short description of studies thus far published on oral and inhalation exposure to nanoform materials. The section on non-oral exposure is poorly written (702). The information presented should either be removed or elaborated such that it is useful to the document. At present adverse effects are presented when neither treatment conditions nor the basic nature of the ENMs are described. This could be highly misleading to the reader. "The section on non-oral exposure should be clarified as to treatment conditions and nature of the ENM used or removed."
2. Introduction to nanotechnologies in the food and feed area	VdMi	line 211: Due to the small size of ENM no new unique properties arise. The properties are known, only the applications might be new. 221 to 226: The focus of this chapter is unclear, whether the focus is on nanotechnologies or ENM. This report should focus on ENM solely. The properties of manufactured nanomaterials depend on the technology used of the specific producer; i.e. the properties are product and production specific. Snow crystals change their form dependent on the temperature.
Terms used in the opinion	Nanotechnology Industries Association	• Terms used in the Opinion o Comment: The terminology used throughout the report is not consistent (cf. ’non-nanoscale’ (line 472), ‘equivalent chemical counterpart’ (line 917-919), ‘equivalent non-nanoforms’ (line 901), ‘conventional chemical’ (line 826), ‘macroscale’ line 335, ‘conventional macroscale material’ (line 675), ‘naturally occurring variants’ (line 409) etc.). o Line 238: The International Standards Organisation (ISO) publication is a technical specification document and has been

		recently published as DIN ISO CEN TS 27687 without the chapter on nanostructured materials. Where possible, official standards should use in opinions and guidance. o Line 244-249: The NIA generally supports the definition used in this opinion (i.e. the combination of size (~100nm) and functionality), but further clarification is necessary.
Terms used in the opinion	FAO	line 243: Even if not in the scope of this scientific opinion, I would point here the need for harmonized nomenclature for nanoparticles. There is in fact a growing international recognition that some particles greater than 100nm exhibit similar behaviour to nanomaterials, and will be therefore important to consider different approaches for avoiding arbitrary size limits in order to promote effective management of risks. Additionally it would be important to consider possible extra criteria in the definition (it is not only the small size that matters, but also the added novel characteristics or properties of the new substances).
Terms used in the opinion	AQUANOVA AG	line 190-192/line 254-258 The distinction between nanoscale organic formulations (like micelles) and nanoparticle made of inorganic materials is not clear enough. So we would kindly ask you for addition the scientific opinion according to the following BfR explanation “What is the link between liposomes, micelles or vesicula and nanotechnology?” (http://www.bfr.bund.de/cd/24949): “Organic compounds like liposomes, micelles or vesicles are used in foods to encapsulate other substances like vitamins or flavourings, to transport them around the body and release them in a targeted manner. As the size of these “transport containers” is frequently in the nanometre range, they are also called nanocapsules. However, in contrast to inorganic, insoluble nanoparticles, their nanoscalability does not lead to any new properties or, by extension, to any new biological effects. Hence, the use of nanoscale organic compounds is not classified as nanotechnology in the narrower sense by BfR. Organic substances like beta-cyclodextrin or polysorbates are frequently used for the capsule membrane. They are toxicologically tested and assessed, and are approved as food additives (E 459 and E 432 up to E 436).” Please see also the definition for engineered nanomaterials of the American Chemistry Council (http://www.americanchemistry.com/ s_acc/bin.asp?SID=1&DID=5090&CID=654&VID=109&DOC=File.PDF): “Proposal: An Engineered Nanomaterial is any intentionally produced material that has a size in 1, 2, or 3-dimensions of typically between 1-100 nanometers. It is noted that neither 1 nm nor 100 nm is a “bright line” and data available for materials outside of this range may be valuable. Buckyballs are also included even though they have a size <1 nm. Exclusions: 1. Materials that do not have properties that are novel/unique/new compared to the non-nanoscale form of a material of the same composition. 2. Materials that are soluble in water or in biologically relevant solvents. Solubility occurs when the material is surrounded by solvent at the molecular level. The rate of dissolution is sufficiently fast that size is not a factor in determining a toxicological endpoint. 3. For those particles that have a particle distribution such that exceeds the 1-100 nm range (e.g. 50-500 nm) if less than 10% of the distribution falls between 1-100 nm it may be considered as non an Engineered Nanomaterial. The 10% level may be on a mass or surface area basis, whichever is more inclusive. 4. Micelles and single polymer molecules.” We are also missing the classification of the nanomaterials regarding the Novel Food Regulation (EC) No. 258/79. Please see the statement of the Official inspection of foodstuffs exemplary for our NovaSOL Q10 product: “Regarding the solubilisates in nanoscale dimension by the firm Aquanova, Darmstadt: Coenzyme Q 10 (ubiquinone) is a substance insoluble in water. In the product NovaSOL Q Coenzym Q 10 the water-insoluble ubiquinone is packed into micelles by adding amphiphilic substances. Since the micelle"s surface is polar, the homopolar character of ubiquinone is covered, and

		no aggregation (even in water) with other homopolar substances is possible. In this way the coenzyme Q10 is present in a colloidal solution. Biological cellular membranes constitute a double layer of lipid nearly impervious for polar substances. The transport of larger polar molecules takes places nearly exclusively over membrane proteins specific for the substrate. As a result, the polar micelles are very probably not transported over the cellular membranes of the intestine. Hence the intake is done only after the ubiquinone is again present in its free form. According to my opinion the packing of coenzyme Q10 in micelles does not lead to any modified metabolism in the intestine. Hence this does not concern, despite the unusual production procedure, a novel food pursuant to Regulation (EC) No. 258197 Article 1 Para. 2 Letter f.“
Terms used in the opinion	ILSI Europe aisbl	We consider it would be useful to clearly define what are considered dissolved chemicals, nano scale, micro scale and macro scale materials. It might be useful to add that dissolved chemicals represent the control or default state; that nano/sub-nano is normal in biochemistry, physiology and pharmacology; and that ENM of concern relates to particulate materials.
Terms used in the opinion	Food Safety Authority of Ireland	Line 248 "Generally, we are in the order of 100nm or less..." Some word are missing here. The use of microscale, macroscale and non-nanoscale terminology throughout the document should be reviewed as they are used inconsistently and therefore may serve to confuse the reader to believe they mean different things when they may not.
Terms used in the opinion	Friends of the Earth (Europe, Australia, Germany), EEB	The regulation of nanotechnology in food and feed and packaging warrants a precautionary approach. To address our concerns, we recommend that the EFSA: ▪ Define manufactured nanoparticles and nanoscale food and feed components as all ingredients and additives that are added to food or feed or packaging, including as processing aids, which: ▪ measure <0.3 -300nm in one or more dimension, or that have a structure that exists at this scale, or ▪ in which particle size is important to achieving the technological function or may relate to a difference in toxicity ▪ Soluble manufactured nanoparticles and nanoscale food or feed components to be included in nanoparticle definitions, disclosure and safety testing requirements. ▪ Define as nanoparticles agglomerates and aggregates whose primary particles are nanoscale or which possess nano-structures and subject them to nanoparticle-appropriate risk assessment and exposure metrics. EFSA itself stated: “It can be assumed that ENM agglomerates break up under certain conditions that occur in food, feed, the gastro intestinal tract and biological tissues.”
Terms used in the opinion	VdMi	244 to 249: The term nano is a scientific measure for 10 to the minus 9. 100 nm would already mean 10 to the minus 7. The range for nano-related effects should not be further expanded and the use of “nano” overstressed.
3. Application of nanotechnologies in the food and feed area	Nanotechnology Industries Association	• 3. Application of nanotechnologies in the food and feed area o Line 279-283: Comment: This category does NOT fall under the opinion’s remit of ‘deliberately introduced into the food chain’ (see Opinion, line 188) o Line 292-293: Comment: This category does NOT fall under the opinion’s remit of ‘deliberately introduced into the food chain’ (see Opinion, line 188) o Line 296-308: Comment: None of these statistics fall into the report’s remit and merely confuse the matter. o Line 309: Comment: The PEN inventory lists products with nano-claims, only. PEN does not test if the claims are true.
3. Application of nanotechnologies in the food and feed area	Scientific Committee of the Belgian Federal Agency for the Safety of the Food Chain	Line 307: “..due to the fixed or embedded nature of ENM in plastic polymers, they are not likely to provide any significant exposure to the consumer”. This seems to be a sweeping conclusion, because “Size (of ENM) potentially reduces the effectiveness of barriers … “ (line 155), “only a few studies have investigated the possible migration of ENM from FCM ...”(line 439). Since one of the main short term applications of ENM for food is FCM, special attention should be paid to the exposure

		through FCM. This issue is briefly mentioned later in the document, but should perhaps be discussed sooner in the document. Line 316: it should be explained what is meant by "nanotechnology processes"? (examples are nanofiltration and -catalysis in zeolites with nanopores, which can improve a production process but not necessarily give rise to in ENM food.)
3. Application of nanotechnologies in the food and feed area	FAO	line 308: Another interesting area of research in the nanotechnology applications is the so called 'Synthetic biology'. That is the name given to a new area of work that combines biotechnology, genetic engineering with nanotechnology, informatics and engineering. It is likely to be some time before artificial organisms capable of self-replication are developed, although critical breakthroughs in the quest to develop synthetic life are being achieved. Synthetic biology has potential applications throughout agricultural and food production systems. The ETC Group reports that Amyris Biotechnologies is developing synthetic microbes to produce nutraceuticals, vitamins and flavours for use in food processing (Amyris Biotechnologies 2006; ETC Group 2007). Codon Devices is also developing synthetic biology applications for agriculture, including efforts to improve the efficiency and control of genetic engineering of plants. For a detailed introduction to the area of synthetic biology see ETC Group (2007). This is a research area that needs to be considered in the food safety context.
3. Application of nanotechnologies in the food and feed area	FAO	line 295: By providing new tools for gene manipulation, nanotechnology is also likely to expand the genetic engineering of crops. Nanobiotechnology now appears to offer a new suite of tools to manipulate the genes of plants or animals by using nanoparticles, nanofibres and nanocapsules, rather than using viral vectors, to carry foreign DNA and chemicals into cells. These nanomaterials can transport a much larger number of genes as well as the chemicals that trigger gene expression. Theoretically, the use of nanotechnology also offers greater control over the release of DNA at the target site. These possible applications of nanotechnology need to be carefully evaluated in the food safety context .
3. Application of nanotechnologies in the food and feed area	CIAA	line 270-276: We welcome the observations that the claimed nanoscale character of applications cannot be verified in many cases 294 - 295: Other indirect applications of nanotechnologies in the food and feed area, such as the development of nanosized agro-chemicals, pesticides, or veterinary medicines. We would welcome clarification and explanation of the 'exposure scenario' considered for this opinion on 'Potential Risks …'.
3. Application of nanotechnologies in the food and feed area	RIVM (National Institute for Public Health and the Environment)	L313-315: "Based on information from EU food industry organizations, there is currently no food ready for marketing, which is produced with use of nanotechnologies or from ENM". Information from several internet sites indicates that there are several food items which are produced with the use of nanotechnologies or from ENM available on the market.
3. Application of nanotechnologies in the food and feed area	Friends of the Earth (Europe, Australia, Germany), EEB	Inconsistencies: On page 8 is written: "The fixed or embedded nature of ENM in plastic polymers, that are not likely to provide any significant exposure to the consumers." But on page 11: "(…) few studies indicates that some ENM may migrate while others do not. Migration is likely dependent on the type of ENM and FCM and no general conclusion can be drawn." As other chemicals nanomaterials could migrate from FCM. This combined with line 317, page 8 statement "The current status of FCM or uses of nanotechnology processes are more uncertain and such applications may be available on the EU market" actually suggest that exposure to the consumer already does exist. EFSA also does not examine the safety challenges of next generation nano food packaging that are designed to interact with the food they contain, releasing colour, odour, nutritional or flavour nanocapsules over time. Many Nano-food products are already on European market. BUND and Friends of the Earth published a list in March 2007 with currently available products in the food-sector. This list contains: nano additives, dietary supplements, FCM and packaging. You will find the whole report with the product list

		here: www.foeeurope.org/activities/nanotechnology/Documents/Nano_food_report.pdf Furthermore plant protection product with nanoscale ingredients are on the market. See: Geohumus (www.geohumus.com) and Nano-Argentum 10 (www.nanosys.ch). But it is certainly possible that more products are available as there is no obligation for labelling or registration. To close the knowledge gap regarding the application of NT in the food and feed area it is necessary that an obligatory register of nano-products be established. This register must be available for the public. Furthermore, in order to allow consumers to make an informed choice, product labels should indicate where manufactured nanomaterials have been added, or where nanoformulation has been used in product manufacture.
3. Application of nanotechnologies in the food and feed area	Food Safety Authority of Ireland	Line 306: "A contributing factor to the rapid commercial developments in the FCM area appears to be the expectation that, due to the fixed or embedded nature of ENM in plastic polymers, they are not likely to provide any significant exposure to consumers". I am not sure how much this expectation contributes to the popularity of FCM as a commercial outlet for ENM compared to its market potential, and its mention in this report may somehow legitimise this misconception.
3. Application of nanotechnologies in the food and feed area	VdMi	line 277 to 295: Chaudry made a market survey of future market chances for industry. Whether those forecasts become true depends on the market.
4. Prerequisite for risk assessment of ENM in food and feed	UK Food Standards Agency	• The draft opinion contains an apparent contradiction relating to the adequacy of the current risk assessment paradigm in assessing nanomaterials for safety. Section 4 suggests that the current risk assessment paradigm is appropriate, while this view appears to be contradicted in other sections which indicate that additional factors need to be considered when assessing nanomaterials for safety. The draft may benefit from more clearly highlighting that, although the basic risk assessment paradigm is appropriate for nanomaterials, the toxicity studies underpinning the risk assessment need to be tailored based on the properties and toxicokinetics of individual nanomaterials.
4. Prerequisite for risk assessment of ENM in food and feed	Nanotechnology Industries Association	• 4. Prerequisite for risk assessment of ENM in food and feed o Comment: This chapter does NOT give guidance; it's a literature review. o Line 319-332: Comment: The wording "nanocharacteristics" (line 325) and "unexpected effects" (line 330) are postulates and not facts with the intention to boost studies of interested parties. o Line 324-325: It is unclear what is meant by 'additional safety concerns' in the sentence 'The traditional RA paradigm is considered an appropriate starting point to address additional safety concerns that may arise due to [...]'. Does this mean new or additional toxicological studies? Will appropriate guidelines be made available?
4. Prerequisite for risk assessment of ENM in food and feed	ILSI Europe aisbl	Line 319. We consider that the definition of risk assessment is not the "evaluation of the potential" but the "probability".
4. Prerequisite for risk assessment of ENM in food and feed	VdMi	line 319 to 332: The wording "nanocharacteristics" (line 325) and "unexpected effects" (line 330) are postulates and not facts with the intention to boost studies of interested parties.
4.1. Physico-chemical	TNO Quality of Life, BU Quality & Safety,	Specifically for food/feed applications, other toxicological issues play a role. Exposure to food/feed components usually happens orally. In contrast to dermal and inhalatory exposure, oral exposure inevitable occurs in a fluidic aquatic environment. Under

characterization of ENM, stability in food and feed matrices, and analytical tools	Zeist	influence of physiological circumstances (gastric fluids, saliva etc) nano-structured components in food/feed may dissolve, which eradicates their nano-specific characteristics. In such situations, to our opinion the toxicological profile of the non-nano (bulk) counterpart will apply. Therefore, to our opinion one of the key elements in risk assessment of ENM for food and feed applications is to know if (and how fast) nano-sized materials dissolve in a couple of key physiological fluids. When nano-materials appear (partly) insoluble, a next important step would be to obtain characteristics of nanomaterials in key physiological fluids. In the draft opinion it was already stated “it is important to measure the ENM in the matrix, as properties of ENM may depend on the surrounding matrix”. From our experience we would like to mention that such measurements are notoriously difficult. Especially detection appears to be difficult in this respect; Methods that are useful for detection, such as radio-labeling or electron microscopy, only work for some materials or at some concentrations and are time-consuming and expensive. Although detection and measuring of ENM appears difficult, to our opinion, it is a key requirement to obtain reliable hazard indications. Firstly, detection is important to identify the nano-associated characteristics of a substance in order to establish dose-response relationships. Secondly, detection is important to obtain toxicokinetics data, an important requirement to ultimately calculate risks.
4.1. Physico-chemical characterization of ENM, stability in food and feed matrices, and analytical tools	CIAA	This chapter does NOT give guidance; it’s a literature review. line 319 to 332:The wording “nanocharacteristics” (line 325) and “unexpected effects” (line 330) are postulates and not facts with the intention to boost studies of interested parties. 325: additional safety concerns and lines329 special considerations Does this mean new or additional toxicological studies? If so are new guidelines to conduct these studies going to be available?
4.1. Physico-chemical characterization of ENM, stability in food and feed matrices, and analytical tools	Food Safety Authority of Ireland	Line 337: "... novel application..." should read; "...novel applications..."
4.1.1. Characteristics of ENM	UK Food Standards Agency	• The draft opinion does not discuss naturally occurring nanomaterials, and food itself is very complex at the nanoscale It might be useful to consider whether discussion of these nanomaterials and nanostructures could provide a useful context for examining engineered nanomaterials, • The distinction between soluble (biodegradable) and insoluble (biopersistent) nanomaterials needs to be more clearly highlighted. It should be noted that most of the available data are for “hard” biopersistent nanomaterials while there may be less potential for biodegradable nanomaterials to accumulate in tissues and organs, • The opinion should mention the difficulty of detecting in vivo the lipids used for nanoencapsulation as they are identical to naturally occurring lipids • The opinion may benefit from more clearly highlighting that 100 nm is not a rigid cut off point, several members had gained

		this impression from the opinion, and that factors such as surface charge can play a more important role in terms of toxicity, The example of titanium dioxide was provided where particles of the same size behave differently depending on whether they are positively or negatively charged.
4.1.1. Characteristics of ENM	Nanotechnology Industries Association	• 4.1. Physico-chemical characterization of ENM, stability in food and feed matrices, and analytical tools • 4.1.1 Characteristics of ENM o Line 367: Comment: The NIA cannot support the generalising comment saying 'Almost all types of ENM catalyze reactions, mainly oxidation reactions.' (cf. coated TiO2 particles are engineered (i.e. coated) to scavenge free radicals and prevent oxidative reactions); at least, it requires clarification. o Line 371-375: Comment: 3 types of particle sizes need to be considered: (a) primary particles (hardly ever free), (b) aggregates: Collection of primary particles tightly bound together by strong forces (> 100 nm), agglomerates: Collection of aggregates that are bound together by physical forces (e.g. short-range Van der Waals attraction) (particle size >> 100nm)
4.1.1. Characteristics of ENM	RIVM (National Institute for Public Health and the Environment)	L347: It is stated that for "nanoencapsulates and for assessing the sites of distribution and/or accumulation, the lipophilicity/hydrophobicity is important". It is not clear why the focus is on nanoencapsulates. Information on the hydrophobicity may be important for many different types of ENM.
4.1.1. Characteristics of ENM	ILSI Europe aisbl	Line 359. We recommend tightening the wording, it is stated, "what makes ENM special is that as the size of the particles decreases the surface area increases dramatically..." What is meant is the surface area proportion increases dramatically otherwise a very tiny particle would have a surface area bigger than the world! The subsequent statement that ultimately the properties of the surface molecules will dominate needs clarification, as for a pure substance the surface molecules and the internal molecules are the same, it is their reactivity that may differ.
4.1.1. Characteristics of ENM	VdMi	line 350 to 351: This phrase does not describe the chemical and physical state of any nanoparticulate matter at the point of sale. There the bonds are saturated and particles agglomerated and aggregated. Isolated nanoparticles cannot easily be transported at high concentrations, which makes them commercionally not of interest. 359: ENM is not a substance class. Every chemical substance is different.
4.1.2. Properties of ENM in food, feed and biological tissues	UK Food Standards Agency	• A clear emphasis needs to be placed on the possibility that a small change in formulation of a nanoproduct could result in a large change in the properties of that product e.g. special attention was needed to effects on mutable surface properties, • There could be a need to move from ingredient to product testing, as a consequence of formulation effects that change physical characteristics. This would have implications in terms of increased animal use and cost
4.1.2. Properties of ENM in food, feed and biological tissues	Nanotechnology Industries Association	• 4.1.2. Properties of ENM in food, feed and biological tissues o Line 377: Comment: It is not clear if the properties of ENM agglomerates and dispersed particles or free form particles are the same in relation to interaction with biological tissues.
4.1.2. Properties of ENM in food, feed and biological tissues	The UK Government Chemist	378-391 Many techniques are available at various stages of development to investigate both kinetic and equilibrium binding effects, particularly interactions with proteins. Examples include ion mobility spectrometry, surface plasmon resonance, and analytical ultrasound. However, these would need to be validated on a case-by-case basis. Given the "hard-corona" protein effect it may be possible to develop lower cost (e.g. ELISA) screening methods for ENM-protein adducts, especially if the structures of the free and bound biomolecules are sufficiently different.
4.1.2. Properties	Friends of the Earth	Line 379, add at the end of the sentence "and can also cross biological barriers, such as the blood brain."

of ENM in food, feed, and biological tissues	(Europe, Australia, Germany), EEB	
4.1.2. Properties of ENM in food, feed and biological tissues	FAO	line 391: Consideration of the nanoparticle stability within the food in relation to different storage and use conditions (e.g. from freezer to microwave)
4.1.2. Properties of ENM in food, feed and biological tissues	CIAA	line 377-385:t is not clear if the properties of ENM agglomerates and dispersed particles or free form particles are the same in relation to interaction with biological tissues. 378:ENM can interact with proteins and others. If ENM “react” with those substances has to be proven by experiments. 382 to 391:This is a very hypothetic example as no product has been proposed to the market.
4.1.2. Properties of ENM in food, feed and biological tissues	RIVM (National Institute for Public Health and the Environment)	L376-391: This section is on the properties of ENM with biomolecules like proteins in food, feed and biological tissues. In addition to the interactions like protein binding, the ENM may also dissolve in these matrices, either as ions or as neutral dissolved molecules. This should be addressed as well in this section.
4.1.2. Properties of ENM in food, feed and biological tissues	ILSI Europe aisbl	We consider the part on protein corona effects rather speculative. If mentioned at all, this should be related also to areas outside of nanotechnology.
4.1.2. Properties of ENM in food, feed and biological tissues	VdMi	line 378: ENM can interact with proteins and others. If ENM “react” with those substances has to be proven by experiments. 382 to 391: This is a very hypothetic example as no product has been proposed to the market.
4.1.3. Analytical tools for detection, quantification and characterisation of ENM in food and feed matrix	Nanotechnology Industries Association	• 4.1.3. Analytical tools for detection, quantification and characterization of ENM in food and feed matrix o Line 408-411: Comment: This is NOT a fact, but a temporary lack of routine detection and tracking equipment; the NIA is leading a consortium funded by the nanotechnology industry and the UK Government for 3 years, in order to characterise ZnO and CeO2 nanoparticles, develop detection and tracking equipment (using isotope tracking) and test their ecotoxicology and environmental fate (i.e. this project forms part of the OECD Sponsorship Programme of Manufactured Nanomaterials). It is anticipated that prototypes of detectors will be developed that allow the isotope tracking of these and other suitable particles in different media.
4.1.3. Analytical tools for detection, quantification and characterisation of ENM in food and feed matrix	Scientific Committee of the Belgian Federal Agency for the Safety of the Food Chain	Line 410 and line 420: “some ENM cannot be distinguished from naturally occurring variants of the same material; one such example is engineered nanoscale SiO2.” Does it makes a difference on human health whether it concerns ENM or naturally occurring materials? Are natural materials as harmful or do they behave differently?
4.1.3. Analytical tools for detection, quantification and	The UK Government Chemist	401-2 However, where the objective is food chain surveillance or quantifying exposure, more conventional extraction-separation-detection protocols may be valid. A chemical ‘fingerprint’ can be conclusive in these contexts. 404 Suggest ‘chemical analytical (and ideally, in situ chemical probe) tools’. Validation of high throughput sampling/sectioning

characterisation of ENM in food and feed matrix		and imaging approaches is needed to power up the sensitivity and specificity of in situ analytics. 406-8 In vivo tracking of ENM is a challenge felt keenly by environmental toxicologists (personal communication). Radiolabelling is only an option at the risk assessment stage; it may be appropriate to encourage the development of self-indicating ENM, e.g. incorporating reactive tag technologies (some variants can also provide information about particle integrity). 412-5 In particular, the development of matrix-based reference materials will require careful planning and experimentation to ensure that the interactions of ENM with food, feed and biological tissue are representative of real samples. 419-422 Suggest 'other tools. Effective chemical analysis depends on steps being taken to minimise artificial losses during the preparatory steps and optimise analytical detection limits. If ENM contain elements which also are endogenous, or are taken up with natural food (such as SiO2), specialised techniques and further method development will be needed to quantify the amount of ENM'. (In the case of SiO2, possible approaches include isotope ratio mass spectrometry, silane skeleton analysis, and differentiation based on surface chemistry.) 424 Suggest 'species exists, but a focus on characteristic chemical structures may be needed to determine whether it is in nanoform.'
4.1.3. Analytical tools for detection, quantification and characterisation of ENM in food and feed matrix	FAO	line 401: Additionally, the tendency of nanoparticles to aggregate, often as a result of the drying stage during the synthesis process or on exposure to different media, is of particular importance to the characterisation of nanoparticles, while most material characterisation techniques focus on the pristine particle (as-synthesised) in powder form, following purification and/or drying.
4.1.3. Analytical tools for detection, quantification and characterisation of ENM in food and feed matrix	CIAA	line 394-427: this section merely highlights the difficulties of analysing nanomaterials and complications added by the lack of means to distinguish between naturally occurring and ENM 404: If the levels of detection are so low then are they relevant for risk assessmnet? We could develop more sensitive methods for detection of bulk material/soluble particles too.n 425:Detection methods are available, but depend on the chemical element to be determined. ENM cannot be detected as a substance class.
4.1.3. Analytical tools for detection, quantification and characterisation of ENM in food and feed matrix	Federal Institute for Risk Assessment	Line 400-401: To facilitate reading, it may be specified that: "It is important to measure the ENM in the relevant matrix, …" and "… than to analyse in simple model matrices."
4.1.3. Analytical tools for detection, quantification and characterisation of ENM in food and feed matrix	RIVM (National Institute for Public Health and the Environment)	L392-427: This section addresses the analytical tools for detection, quantification and characterization of ENM in food and feed matrices. However, only the advantages and limitations of electron microscopy and analytical tools that analyze elements rather than particles such as ICP-MS are discussed. Obviously, more techniques exist that should be addressed regarding the title of this section. In addition, the losses of an element during the preparatory steps for ICP-MS analysis are discussed, but more importantly, preparatory steps can affect the particle characteristics (aggregation, agglomeration, charge, etc) before analysis.
4.1.3. Analytical tools for detection, quantification and characterisation of	ILSI Europe aisbl	Line 395 refers to pristine ENM, how is this different from ENM?

ENM in food and feed matrix		
4.1.3. Analytical tools for detection, quantification and characterisation of ENM in food and feed matrix	Food Safety Authority of Ireland	Line 395: the word "pristine" is used two times in this document but is not defined anywhere that I could find. Line 404: "However, the detection by EM is only possible if the number of ENM is sufficiently high to find a detectable number of ENM in the matrix...." should read: "However, the detection by EM is only possible if the number of ENM is sufficiently high in the matrix...."
4.1.3. Analytical tools for detection, quantification and characterisation of ENM in food and feed matrix	Food Safety Authority of Ireland	Line 395: the word "pristine" is used two times in this document but is not defined anywhere that I could find.
4.1.3. Analytical tools for detection, quantification and characterisation of ENM in food and feed matrix	VdMi	line 425: Detection methods are available, but depend on the chemical element to be determined. ENM cannot be detected as a substance class.
4.2. Exposure to ENM from food and feed	Nanotechnology Industries Association	• 4.2. Exposure to ENM from food and feed o Line 433: Comment: the term 'nanotechnology' is not appropriate in this context (see above).
4.2. Exposure to ENM from food and feed	TNO Quality of Life, BU Quality & Safety, Zeist	Chapter 4.2 gives an overview of information available regarding the sources of ENM exposure. It is concluded in the draft opinion that significant consumer and animal exposure to ENM ingredients in food and feed is currently not likely within the EU, though there may be exposure to nanoscale fractions within other materials. However, as indicated in section 4.2.1., there is a (great) lack of data regarding possible sources of exposure. Therefore, it is not clear what the basis is for the conclusion stated in section 4.2.1 (line 465-468).
4.2. Exposure to ENM from food and feed	CIAA	line 433:The use of "nanotechnology" is a too wide expression for the identification of risks; better use ENM.
4.2. Exposure to ENM from food and feed	EFFAT	Line 432 After the word Consumers, add: ", and workers in the processing facilities,"
4.2. Exposure to ENM from food and feed	Friends of the Earth (Europe, Australia, Germany), EEB	Line 433, please add "…due to the current limited knowledge on the availability". EFSA has stated clearly earlier in the document that there is huge uncertainty on what products and in what amounts are available on the market in Europe, concerning nano applications in food and feed
4.2. Exposure to ENM from food and feed	ILSI Europe aisbl	It might be considered to comment on other routes of exposure than oral, inhalation and dermal exposure could be very important not only for manufacturers but also for those involved in handling of packaging etc.
4.2. Exposure to	VdMi	line

ENM from food and feed		433: The use of “nanotechnology” is a too wide expression for the identification of risks; better use ENM.
4.2.1. Sources of exposure	Bayer AG	Section 4.2.1 Sources of Exposure Due to the size of ENM – with a molecular mass well above 1000 g/mol, quite large in comparison with a molecule – the diffusion rates will be extremely low so that it can be reasonably expected that no migration of ENM from Food Contact Materials to food occurs. (See Brandsch; 4th International Symposium on Food Packaging, “Nanomaterials in contact with Foods”; 2008-11-19/21, Prague) [Note: The reference FSA, 2008 is not listed in the reference list.]
4.2.1. Sources of exposure	Nanotechnology Industries Association	• 4.2.1. Sources of exposure o Line 444: Comment: Any consideration of potential ‘release of ENM (or their residues) into food/feed through wear of food/feed processing machines with coatings containing ENM’ needs to be put into context of potential ‘wear and tear’ of regular (i.e. non-ENM containing) processing equipment. o Line 460: Comment: The NIA strongly disagrees with the use of the word ‘inevitably’ in the context of ‘Production and widespread use of ENM in consumer products (e.g., electronics, medicines, packaging materials) will inevitably result in environmental release of these particles over the product life-cycle (Nowack and Bucheli, 2007).’
4.2.1. Sources of exposure	Scientific Committee of the Belgian Federal Agency for the Safety of the Food Chain	Line 458: “residues of nano-formulated pesticides ... are currently not likely as no nano-formulated .. are currently available in the EU”. This statement seems to be too strong; Quid the import of foodstuff from outside the EU? Quid undeclared applications? (cfr. line 433) Line 465: The entire section on "sources of exposure is characterized by uncertainty and lack of evidence that the described phenomenon occurs. On the other hand, the conclusion “significant consumer and animal exposure to ENM ingredients in food and feed is currently not likely within EU, though there may be exposure to nanoscale fractions within other materials. However, products are available via the Internet; this contribution to consumer exposure is not quantified.“ is too vigorously.
4.2.1. Sources of exposure	CIAA	line 444 – 445: There may be release of ENM (or their residues) into food/feed through wear of food/feed processing machines with coatings containing ENM. We would appreciate clarification on this statement. 446:“Nanotechnology devices” is too much science fiction for such a report; better eliminate last phrase. 460: packaging materials) will inevitably result in environmental release of these particles The term ‘inevitably’ does not seem appropriate could this please be clarified. 466 to 468:“ ’though there may be exposure to nanoscale fractions with other materials” and the last phrase should be completely eliminated. Every likelihood should not be taken into account.
4.2.1. Sources of exposure	Federal Institute for Risk Assessment	Line 455-458: At least one agroproduct presumably containing ENMs is currently available in the EU. Nanoargentum 10, a plant care product intended for „all plants and vegetables“ (reg-istered and legally sold in Germany) is claimed to contain 10 ppm

		colloidal metallic silver. Residues on treated food and feed plants appear likely. It must be noted, however, that no data has been made publicly available demonstrating the nanoscale properties of the active ingredient. In addition, it has been claimed by third parties, that the Syngenta products Primo MAXX and Banner MAXX represent nanoemulsions (ENM acc. to EFSA definition in chapter 2) of trinexapac ethyl and propiconazole, respectively. Both are approved for treatment of greens by NL and UK CAs, resp. However, residues in food and feed products appear unlikely based on the intended use, and no data demonstrating the nanoscale of both formu-lations is currently available to the german CA. The following wording is proposed …and veterinary products are currently not likely as no nano-formulated pesticides or fertilizers and veterinary drugs are currently commercially available in the EU. But, in particular individual cases the presence of ENM in pesti-cides, fertilizers, strengthening agents for plants and veterinary drugs placed on the market cannot be totally ruled out due to the lack of respective information of actual size and size distribution of so-called micro- or nanoemulsions. In principle, human ex-posure … Line 459-464: It may be noted that use of nanoscale TiO2 or SiO2 in paints and surface treatments could also contribute to food and feed contamination with ENMs via environ-mental routes / sewage. This contribution may comparatively substantial, considering the relative quantities used for these applications (Schmid & Riediker, 2008).
4.2.1. Sources of exposure	Friends of the Earth (Europe, Australia, Germany), EEB	Different sources of exposure are described. EFSA itself mentioned that information on exposure (commercial use) is extremely limited and all available information comes from industry. Therefore the conclusion that “significant consumer and animal exposure is currently not likely within the EU” is neither comprehensible nor justifiable.
4.2.1. Sources of exposure	RIVM (National Institute for Public Health and the Environment)	L451-452: “Exposure assessment from applications in feed for the target animal (e.g., food-producing species) would follow the same lines as for human exposure assessment”. Please explain which lines are followed for the exposure assessment for humans as well as animals. L465-467: “significant consumer and animal exposure to ENM ingredients in food and feed is currently not likely within EU, though there may be exposure to nanoscale fractions within other materials”. Please explain what kind of materials (e.g. consumer products or medicines?). L467-468: “However, products are available via the Internet; this contribution to consumer exposure is not quantified”. Please specify what kind of products (e.g. food and feed products or consumer products?)
4.2.1. Sources of exposure	EFFAT	Line 465 As the information of the use of ENM in food/feed and FCM (line 430) and as it may be that products incorporating undeclared ENM are already marketed and distributed (line 433), it is not possible to estimate whether exposure is significant or not. In such a sentence significant would only refer to a quantitative vision and not a qualitative appreciation of the hazards. Moreover, not only consumers and animals are exposed to ENM. Agriculture (e.g. use of pesticide) and foodworkers (during the processing) are under threat, if not properly protected. Consumers are exposed to ENM by ingestion only when they buy the products. Workers are exposed to ENM (e.g. by inhalation, skin contact, ingestion etc...) all day, week, month (years?) long. We therefore suggest the following change: "In conclusion, in a quantitative perspective, significant, consumer, agriculture worker, foodworker and animal exposure to..."
4.2.1. Sources of exposure	Food Safety Authority of Ireland	Line 466: "...within EU..." should read: "...within the EU..."
4.2.1. Sources of	VdMi	line

exposure		446: “Nanotechnology devices” is too much science fiction for such a report; better eliminate last phrase. 448 to 464: As long as there is no application seen for the next future, this chapter is not necessary. 466 to 468: “ ’though there may be exposure to nanoscale fractions with other materials” and the last phrase should be completely eliminated. Every likelihood should not be taken into account.
4.2.1. Sources of exposure	MDCTec Ltd.	Lines: 438 to 443 Additional Information availalbel. To be published in Food Additives & Contaminants Title: "Nanomaterials in contact with food - consumer exposure through interaction and interfaces." Summary: Exposure of consumers with nanoparticles from food packaging through interaction and interfaces is presented and discussed in detail based on two technological approaches. Organoclay nanocomposites are well represented in food packaging applications. The mobility of the organoclay nanoparticles in plastic is investigated and the contamination probability of food through interaction estimated respectively. The second technological approach considered and well represented in food packaging applications are nanoscale layers applied at the interface contact material / food. Mechanical and chemical stability of these layers are the main criteria for their safe use in food contact applications. Difficulties in quantification of nanoparticle contaminations in foods or food simulants and the feasibility of chromatographic techniques will be discussed. Literature (1) Reichert, P.; Nitz, H-J.; Klinke, S.; Brandsch, R.; Thomann, R.; Mülhaupt, R. „Poly(propylene)/organoclay nanocomposite formation: Influence of compatibilizer functionality and organoclay modification.“ Macromol. Mater. Eng. 2000, 275, 8-17 (2) Reichert Peter, "Polyolefin-Nanocomposite auf der Basis von organophilen Dreischichtsilikaten" Inaugural Dissertation, 2000 (3) Field Flow Fractionation, Technical Documentation, Postnova Analytics GmbH, Max-Planck-Str. 14, 86899 Landsberg am Lech, Germany
4.2.2. Estimations of dietary exposure	Nanotechnology Industries Association	• 4.2.2. Estimations of dietary exposure o Line 477- 484: Comment: This paragraph implies an extremely large number of additional tests that need to be done in order to estimate dietary exposure covering encapsulated bioactive compound, nanocarrier system and the free form of the nanomaterial. Clarification is needed. o Line 491-496: Comment: This paragraph implies additional tests to show the effects of change in structure of the primary ENM. Clarification is needed.
4.2.2. Estimations of dietary exposure	TNO Quality of Life, BU Quality & Safety, Zeist	Section 4.2.2 gives an overview of the estimation of dietary exposure. From this section it appears that no estimations of dietary exposure to ENM have been performed up till know. This section gives an overview of the issues that might be important when using data to perform an exposure assessment for ENM. To our opinion it might be valuable to take into account some form of tiered approach to estimate the exposure to ENM, ranging from screening to very detailed assessments. This will influence the use of the data and the variability’s and uncertainties to be taken into account. For example, at the level of screening, information regarding processing might not be necessary.

4.2.2. Estimations of dietary exposure	The UK Government Chemist	[Sorry - previous attempts didn"t define Chapter/Section correctly for this comment.] 480 Define primary and secondary particles? 480 'currently' is correct in this context – the analytical problems are challenging but steady progress can be anticipated.
4.2.2. Estimations of dietary exposure	FAO	line 483: Dietary exposure to nanoparticles can result not only from ingestion of nanoparticles in food, or from food contact materials including packaging. More indirect exposure can arise from ingestion of food from animals such as fish and shellfish (i.e., molluscs and crustaceans), that have taken up nanoparticulate matter, as part of the human diet. Many food products contain considerable amounts of anthropogenic (naturally occurring) nanoparticles, such as silica (or even traces of titanium dioxide), and that makes estimation of exposure to deliberately added nanoparticles even more difficult.
4.2.2. Estimations of dietary exposure	CIAA	line 477- 484: implies an extremely large number of additional tests that need to be done in order to estimate dietary exposure covering encapsulated bioactive compound, nanocarrier system and the free form of the nanomaterial. 491-496: imply additional tests to show the effects of change in structure of the primary ENM. It is not clear if the changes refer to physical morphology and changes due to interactions with different biological components referred to in line 492
4.2.2. Estimations of dietary exposure	RIVM (National Institute for Public Health and the Environment)	L475: "The current food consumption databases can be used". Please consider adding the following text: "to estimate the consumption of food and feed products containing ENM".
4.3. Toxicokinetics of ENM	The Dr Hadwen Trust for Humane Research	We wholly agree with the recommendations into furthering the currently limited knowledge and understanding of ENM behaviour and toxicokinetics. However, we do not support the assumption that "…the toxicological properties of substances, including ENM, will have to rely on in vivo studies". Until revised and specific test guidelines for nanomaterials exist, toxicity testing would have to be carried out according to already existing guidelines and/or by corresponding test methods. For these reasons the Dr Hadwen Trust recommends that for non-essential, non-medical applications (including cosmetic and household products, sporting equipment, textiles, food, feed and paints), ENM manufacture and use is prohibited immediately until relevant nano-specific safety testing and risk assessment protocols are in place. In a recent publication by the Royal Commission on Environmental Pollution it was acknowledged that "...the scientific basis to fully understand all properties and risks of nanomaterials is not sufficiently available at this point in time" . In accordance with this the Dr Hadwen Trust further believes that animal testing of nanomaterials is scientifically highly questionable. We would prefer to see an acknowledgement that, in concordance with the mention of in vitro methods that are not yet validated, existing animal tests are not validated for this application, and greater emphasis to be placed on the development, validation and use of non-animal test methods. Animal tests have limited value because of their inherent uncertainties . These include the difficulties of extrapolating test data between species, genders and breeds of animals including humans (due to anatomical, physiological, biochemical, metabolic and pharmacological differences). There are major uncertainties in interpreting information from high-dose animal tests with single chemicals in ways that are relevant to low-dose human exposures to chemical cocktails. There are also problems with mimicking human routes of exposure in animal tests, and with scaling up from small animals with a short lifespan to larger humans who may be exposed to chemicals over decades. Even for data-rich chemicals, these uncertainties delay regulatory decision-making, prolonging risks of damage to human health and the environment.
4.3. Toxicokinetics	TNO Quality of Life,	It is noted that the draft opinion mainly focus on the differences between ENM and its macro scale equivalent, whereas a

of ENM	BU Quality & Safety, Zeist	comparison of the ENM with the (dissolved) chemical remains, to some extend, underexposed. A random check of some of the references used (e.g. De Jong, 2008; Kim, 2008; Wang, 2007; Zhang, 2004) showed that upon oral exposure to nano particles, not the particles but the metal ion was detected in tissues by e.g. ICP-MS techniques. As such, a discrimination between ENM absorption followed by distribution of these particles over the body, or (absorption of) the dissolved chemical released from the particle (e.g. as free soluble or protein bound component), cannot be made. Therefore, to our opinion it remains unclear if the measured increase of the analysed compounds in the respective tissues or an observed increase in toxicity in some of the studies is related to systemically available ENM or to a release of chemicals thereof (e.g. as a result of the substantial bigger surface area of ENM compared to its macro scale equivalent). Therefore, for the toxicokinetics and toxicity of the ENM, a comparison with relevant counterparts might be considered of relevance (e.g. in case of a metal containing ENM, kinetic and toxicological information of a soluble metal equivalent might be taken into account as well). It might be assumed that in case of a release of chemicals from the ENM, because of the assumed more gradual release of the chemical from the ENM, a more worst case estimate might be found compared to the insoluble ENM. However, information on the relative release might provide a more realistic risk assessment.
4.3. Toxicokinetics of ENM	CIAA	This section is rather meaningless unless the examples specifically compare to bulk materials of different sizes. For example the ranges of uptake given under absorption of "2-200" times greater and "15-150" times greater this does not really add anythingexept that it is shown that not all ENMs are equal....in the same way that not all bulk materials are equal either.
4.3.1. Absorption	Nanotechnology Industries Association	• 4.3. Toxicokinetics of ENM • 4.3.1. Absorption o Line 477- 484: Comment: Substances as used by Kreyling are not the reality of substances likely to be marketed. There is no 'model ENM'.
4.3.1. Absorption	CIAA	line 516 to 523:Model substances as used by Kreyling are not the reality of substances likely to be marketed. There is no model ENM.
4.3.1. Absorption	Federal Institute for Risk Assessment	Line 516-519: The sentence describing the findings of Szentkuti may be potentially misinter-preted by the unprepared reader as relating to passage across the whole GI barrier. It might be stressed at the end of the sentence, that the findings relate to „this outer part of the bar-rier“ or „the mucos barrier separating lumen and cellular epithelium“. Line 522-523: The study by Jani et al., 1994, found an exceptionally high level of absorption of 12 % at a dose of 12.5 mg/kg bw/d TiO2-NP (500nm) administered repeatedly for 10 days to rats. It may be appropriate to cite data from other studies, such as that by Wang et al., 2007, which showed (at 14 days post application) liver residues corr. to 0.0001, 0.004 and 0.0001 % of a single high dose of 5 mg/kg bw TiO2 NP with average sizes of 25, 80 and 155 nm, respectively, administered to mice. Other results implicate, that an effective elimination is unlikely to be the main reason for such low tissue residues (e.g. Fabian et al., 2008 i.v. ad-ministration).
4.3.1. Absorption	RIVM (National Institute for Public Health and the Environment)	Section 4.3.1 (L502-530): This section addresses the absorption of ENM in the gastro intestinal tract. The difference between “absorption” and “translocation” is not clear. In line 516 “diffusion” seems to be more appropriate than “absorption”.
4.3.1. Absorption	ILSI Europe aisbl	Line 503. This sentence appears ENM to mean insoluble particulates. This needs to be made clear as it also relates throughout the whole of the toxicokinetics Section, 4.3.
4.3.1. Absorption	Food Safety Authority of Ireland	Line 511: "...the epithelium is denending on their..." should read: "...the epithelium depends on their..."
4.3.1. Absorption	VdMi	line

		516 to 523: Model substances as used by Kreyling are not the reality of substances likely to be marketed. There is no model ENM.
4.3.1. Absorption	VdMi	line 425: Detection methods are available, but depend on the chemical element to be determined. ENM cannot be detected as a substance class.
4.3.2. Distribution	TNO Quality of Life, BU Quality & Safety, Zeist	Regarding distribution, due to their small size ENM may enter cellular structures that are inaccessible for bulk compounds. Previous reports already mentioned increased cellular uptake by ENM when compared to 'micro'-sized materials. When, for instance, ENM acquire the ability to cross the blood-brain barrier, novel toxicological profiles may arise. Novel biodistribution kinetics could be assessed using ADME studies. To our opinion, analyzing biodistribution of representative ENM would be very helpful to identify changes that may affect hazardous responses. The technical challenge for these studies is to reliably detect/measure ENM (see also comment in section 4.1). It would be useful to screen ADME features for different (groups of) ENM for which application is anticipated. ADME results can than be correlated with key characteristics (in relevant matrices), such as: size distribution, hydrophobicity, agglomeration/aggregation, particle number, shape. This would help to define toxicological parameters for nanomaterials in food/feed, and could ultimately enable toxicological profiling using read- across techniques.
4.3.2. Distribution	Federal Institute for Risk Assessment	Line 549-550: Again, the cited figure of 6 % recovery of 50 nm polystyrene particles in se-lected organs from Jani et al. (1990) appears very high in comparison to other data. Evalua-tion of the study by Hillyer and Albrecht (2001) on Au-NP (4 to 58 nm) cited in lines 545-547, shows that these authors recovered only between 0.002 and 0.0002 % of the dose adminis-tered with the drinking water in 9 major organs representing 21 % of bw. Please refer also to comment on line 522-523 made above.
4.3.2. Distribution	RIVM (National Institute for Public Health and the Environment)	L546: It is stated that the distribution to organs increases with smaller particle size. It is unclear if the distribution, i.e. the exposure at organ level, is expressed as mass of ENM per mass of tissue. In that case, when expressed as surface area of ENM per mass of tissue the increase in distribution with smaller particle size is even greater. Although it is unclear how a concentration is best expressed, the difference should be discussed. The same holds for details of other studies in this section in which the translocation or distribution of a certain ENM is expressed as a percentage of a dose. Does the percentage refer to a percentage of the number of particles, the mass of particles or the surface area? Section 4.3.2 (L531-593): The placental barrier is identified as a barrier that requires special attention for ENM. In addition, also the blood-brain barrier and blood-testis barrier should be addressed. Can the ENM cross these barriers and reach the vulnerable tissue behind? It would be interesting to investigate whether in vitro tests can predict if certain ENM can cross these barriers.
4.3.3. Metabolism (biotransformation)	ILSI Europe aisbl	Line 595. Does this refer to particulate or soluble materials?
4.3.3. Metabolism (biotransformation)	Food Safety Authority of Ireland	Line 595: "There is no information regarding biotransformation of ENM after oral administration". Does this cover animal and human studies?
4.3.5. Conclusion on Toxicokinetics	FAO	line 646: What about information on possible relation between nanoparticles bio-interactions and allergenicity?
4.3.5. Conclusion on Toxicokinetics	FAO	line 643: What about information on the possibility for nanoparticles to evade the protective blood-brain barrier and enter the brain?
4.3.5. Conclusion on Toxicokinetics	CIAA	line 621 to 646:There should be listed the effects seen with specific substances and not postulates on ENM in general.
4.3.5. Conclusion	RIVM (National	L621. It is stated that toxicokinetic studies on ENM following oral exposure have been performed mainly on metals and metal

on Toxicokinetics	Institute for Public Health and the Environment)	oxides. However, already several studies on nanoformulated drugs have been published in the public scientific literature, including information on the toxicokinetics of the ENM. L646. This is a very important conclusion. If the clearance of some ENM from tissues is indeed very slow there is a great potential for accumulation and toxicity after chronic exposure. Therefore, an important recommendation should be that not only information about the toxicity after chronic exposure should be obtained (L965-967), but also information about the toxicokinetics (especially clearance and tissue distribution) after chronic, repeated exposure.
4.3.5. Conclusion on Toxicokinetics	ILSI Europe aisbl	Line 621. The conclusion mentions that the bulk of the preceding section was based on metals and metal oxides, this should be made clear much earlier in the text.
4.3.5. Conclusion on Toxicokinetics	VdMi	line 621 to 646: There should be listed the effects seen with specific substances and not postulates on ENM in general
4.4. Toxicity of ENM	The Dr Hadwen Trust for Humane Research	With a new field such as nanomaterials, the full range of potential toxicities is not known. Using standard animal toxicity tests, which are little more than 'black box' methods, would risk overlooking novel unwanted effects. Human cell-based assays, in contrast, would allow the study and elucidation of a range of molecular and cellular mechanisms of toxicity. For example, human cell culture assays can be used to monitor the oxidative stress responses of cells exposed to nanoparticles. There are a number of non-animal techniques currently being developed that represent a potential for nanomaterial safety testing. For example, perfusable 3D cell-matrix chambers for testing nanoparticle permeability and transport through tissues ; and the HμREL device , which allows the toxicity of nanomaterials to be tested on several cell types in a multi-chambered microchip with a microfluidic channel, represent promising in vitro methods. Human cell culture techniques have provided useful information on specific cellular responses to nanomaterials by measuring chemical responses or responses at the DNA level using biomarkers and genomic techniques . The feasibility of analysing in vitro nanomaterial activity in a general, systemic fashion has also been demonstrated using a multidimensional profiling approach with multiple cell types and assays reflecting different aspects of cellular physiology . The data are then clustered using computational methods to identify nanomaterials with similar patterns of biological activity across a broad sampling of cellular contexts, as opposed to sampling from a single assay. This approach yields robust and detailed structure-activity relationships. Additionally, interesting alternative tests are already being developed by EU-funded Joint Research Centre projects such as Nanotox, which involves human cell culture techniques. In summary, human-relevant non-animal assays offer several advantages: using human cells or sub-cellular components they avoid species differences, and high-throughput systems allow the very rapid and cost-effective testing of multiple chemicals and multiple toxic endpoints, including novel ones. A moratorium should be introduced on all non-essential uses of ENM. This will ensure the protection both of human health and environmental safety, as well as fulfilling citizens' wishes to maintain high animal welfare standards and prohibit unnecessary laboratory animal use, especially with inhumane and misleading methods.
4.4. Toxicity of ENM	American Chemistry Council	The Committee recommended that if an ENM is ingested in the nanoform, then repeated dose toxicity studies are recommended together with the appropriate in vitro studies for genotoxicity. The Panel agrees that a proper hazard characterization should be conducted; however, application of in vitro testing should take into consideration the intricacies of testing particulates in these systems. This is also highlighted in the committee report (line 862-866) where it is noted that in vitro assays are available for endpoints such as mutagenicity and genotoxicity, but they have not been validated for ENM and as such are useful only for screening purposes. The Panel recommends reestablishing this thought in the Conclusion section

		where reference is made to applying the in vitro studies (line 925) The opinion also highlights that uncertainties exist for characterizing, detecting and measuring ENM in food, feed or the body. The Committee recognizes there is limited information on absorption, distribution, metabolism and excretion, as well as the toxicity of ENM. We too recognize that it is a difficult task in many cases to determine actual exposure potential, and the Panel supports ongoing OECD and NIOSH efforts aimed at developing strategies to address exposure assessment.
4.4. Toxicity of ENM	CIAA	This section raises more safety concerns by quoting different references and suggesting that there is a lot unknown about ENM. line 750 to 752: Microscale chemicals are tested according to existing legislation with their fine particulate fraction, which are an inherent part of these products. Eliminate 750-752!
4.4. Toxicity of ENM	Friends of the Earth (Europe, Australia, Germany), EEB	In chapter 4.4.1. please specify which of the discussed nano materials are of relevance for the food, food packaging and feed production and processing. The text as it is does not refer to the concrete context of food and feed and gives only a general overview of the toxicity of ENM. This does not provide sufficient information about toxicity of ENM in this specific application. The impact of SiO2 is still controversial even if SiO2 agglomerates. As mentioned above the behaviour of agglomerates on basis of ENM is unknown. See EFSA opinion page 10: “Agglomerates may preserve some of the ENM properties, the tendency to agglomerate can be hindered by the modification on the surface. (…) It can be assumed that ENM agglomerates break up under certain conditions that occur in food, feed, the gastro intestinal tract and biological tissues”. Chen and von Mikecz (2005) found negative impacts of SiO2: Chen M, von Mikecz A. 2005. Formation of nucleoplasmic protein aggregates impairs nuclear function in response to SiO2 nanoparticles. Experiment Cell Res 305:51-62. Also Di Pasqua et al. (2008) found that both mesoporous silica nanomaterials and 250nm spherical particles of silica dioxide were cytotoxic. Di Pasqua A. K Sharma, Y-L Shi, Toms B, Ouellette W, Dabrowiak J, Asefa T. (2008). Cytotoxicity of mesoporous silica nanomaterials. J Inorgan Biochem 102 pp.1416–1423. More data on SiO2 and its agglomerates are necessary to assess the impact on human health.
4.4.1. Acute, subacute and subchronic oral toxicity to ENM	Nanotechnology Industries Association	• 4.4. Toxicity in ENM • 4.4.1. Acute, subacute and subchronic oral toxicity to ENM o Line 649: Comment: The sentence that includes "the most important facts are summarised" should be removed as many of the reports included are actually speculative. o Line 689-690: Comment: Is there a reference for this study?
4.4.1. Acute, subacute and subchronic oral toxicity to ENM	CIAA	line 649 : The sentence that includes "the most important facts are summarised" should be removed as many of the reports included are actually speculative.
4.4.1. Acute, subacute and subchronic oral toxicity to ENM	RIVM (National Institute for Public Health and the Environment)	L 652. The general mechanisms of injury are described in this section. We miss the general mechanism of ROS formation that can lead to for instance lipid peroxidation, oxidative stress and DNA damage. L655-658: Please consider adding the following text to further explain the potential effects on the GI-tract: Given the fact that

		nanoparticles may interact with phagocytic and/or epithelial cells, and may induce inflammatory responses, oral uptake of nanomaterial may hypothetically have repercussions for conditions such as irritable bowel disease and celiac disease. An interaction of nanomaterial with epithelial cells might potentially also lead to easier access of proteins into the tissues, thereby facilitating development of food allergy or food allergic reactions.
4.4.1. Acute, subacute and subchronic oral toxicity to ENM	FOPH	line 720: The paper of Barnes et al (2008) concluded that with the comet assay no genotoxicity of amorphous silica nanoparticles was detectable. and in the paper of Jong and Borm (2008) I could not find any comment according to clastogenicity or genotxicity of ENM.
4.4.1. Acute, subacute and subchronic oral toxicity to ENM	FOPH	line 720: The paper of Barnes et al (2008) concluded that with the comet assay no genotoxicity of amorphous silica nanoparticles was detectable. and in the paper of Jong and Borm (2008) I could not find any comment according to clastogenicity or genotxicity of ENM.
4.4.1.1. Metals	CIAA	line 660 to 663:Selenium is not considered a metal, but a non-metal like sulphur, group 6 of the periodic system. Selenites are salts with a cation lines. 660- 663 must be listed in chapter “4.4.1.2. Other ENM”. Is it Mn or MnO like in line 971? 689-690: Is there a reference for this study?
4.4.1.1. Metals	Federal Institute for Risk Assessment	Line 685/686: Increased LDH and HBDH levels may be better termed “changes in serum biochemical parameter” than “blood effects”. In addition, dosing with 25 nm particles affected those parameters only slightly, while most pronounced effects were seen with 80 nm parti-cles (not 25 nm as in the draft), and no effects were observed with 155 nm particles. In addi-tion, it may be worth to note that the observation of effects considered adverse in kidney, liver and heart by Wang et al. (2007) contrasts not only with findings of Jani et al. (1994) (as written in line 688), but also by Fabian et al., 2008. These authors achieved Ti organ levels 2 to 3 magnitudes higher than those measured by Wang et al., but did not observe changes in biochemical parameters indicative for organ toxicity nor an increase in liver weights. Line 690: The following information may be added: “… with an NOAEL of 300 mg/kg bw/d (Kim et al., 2008).”
4.4.1.1. Metals	RIVM (National Institute for Public Health and the Environment)	L660-663: In the Selenium studies that are mentioned, also Zhang et al, 2005 should be referred. Zhang J, Wang H, Yan X, Zhang L. 2005. Comparison of short-term toxicity between Nano-Se and selenite in mice. Life Sci 76(10):1099-109. L672: Is it suggested that inflammation is mainly caused by the agglomerates or can this effect also be the consequence of Zn nanoparticles? L676: What is meant by “the sizes of ENM were checked in the gavage”?
4.4.1.1. Metals	ILSI Europe aisbl	Line 664. At last a reference to insoluble ENM, perhaps this is a better description than particulate, but this distinction needs to be drawn throughout the opinion, the same is true in line 651.
4.4.1.1. Metals	VdMi	line 660 to 663: Selenium is not considered a metal, but a non-metal like sulphur, group 6 of the periodic system. Selenites are salts with a cation lines. 660- 663 must be listed in chapter “4.4.1.2. Other ENM”. Is it Mn or MnO like in line 971?
4.4.1.2. Other ENM	RIVM (National Institute for Public Health and the Environment)	L691-700: Apart from the studies mentioned in this section, there is more information for instance on cationic PAMAM dendrimers (Duncan and Izzo, 2005), C60 polyalkyl sulfonate (Chen et al, 2006) and nano-magnetic ferrofluid (Xia 2005) that can be included in this section of the opinion. Chen Z, Meng H, Xing G, Chen C, Zhao Y, Jia G, Wang T, Yuan H, Ye C, Zhao F and others. 2006c. Acute toxicological effects of copper nanoparticles in vivo. Toxicol Lett 163(2):109-20.

		Xia Z, Wang, G, Tao, K, Li, J, Tian, Y. 2005. Preparation and acute toxicology of nano-magnetic ferrofluid. J-Huazhong-Univ-Sci-Technolog-Med-Sci 25 (1):59-61 (abstract). Duncan R, Izzo L. 2005. Dendrimer biocompatibility and toxicity. Adv Drug Deliv Rev 57(15):2215-37.
4.4.2. Toxicity from non-oral exposure to ENM and in vitro studies	FAO	line 723: Some publications have also described examples of interactions between nanoparticles and subcellular organelles that may lead to cell death by activation of apoptotic or necrotic pathways (Kagan et al., 2006; Hong et al., 2006; Xia et al., 2006)
4.4.2. Toxicity from non-oral exposure to ENM and in vitro studies	ILSI Europe aisbl	Line 720. This is a very general statement, is this implying that genotoxicity and clastogenicity are features of all insoluble nano particulates?
4.4.3. Metrics for dose-response relations of ENM	UK Food Standards Agency	• The opinion makes reference to dosimetrics but does not provide any further detail or explanation to the reader
4.4.3. Metrics for dose-response relations of ENM	TNO Quality of Life, BU Quality & Safety, Zeist	A general issue concerning ENMs is dose metrics. Not only use of ENMs in food/feed, but also other applications of ENMs that require a toxicological review, involve serious reconsideration of dose metrics. For instance, most toxicological data uses mass to describe certain effects of ENMs. Materials with sizes in the nano-range may have altered characteristics that correlate better with their relative surface area, than with their mass. At this point, dose metrics for nanomaterials is a point of discussion. Still, it seems unlikely that one specific dose-metric will be sufficient to describe dose-response effects for all types of nanomaterials reliably. In order to obtain data that is as interpretable as possible, it is advised that a number of characteristics of the test compound should be recorded (such as mass, number and surface area).
4.4.3. Metrics for dose-response relations of ENM	Federal Institute for Risk Assessment	Line 729-730: The study of Poland may also interpreted in such a way, that morphology is rather a determinant of effect quality than effect quantity (dose-response). In analogy to as-bestos fibres, absolute particle numbers (for the relevant particle/fiber class) may be an ap-propriate metric.
4.4.3. Metrics for dose-response relations of ENM	ILSI Europe aisbl	This section appears to imply that there could be some kind of generic toxicity of nanoparticles. Why should there be? Would size per se be the determinant of toxicity rather than physicochemical properties?
4.4.3. Metrics for dose-response relations of ENM	Food Safety Authority of Ireland	Line 729: "...a recent intraperitoneal study indicate that..." should read: "...a recent intraperitoneal study indicates that..."
4.4.4. Additional considerations	Nanotechnology Industries Association	• 4.4.4. Additional considerations o Line 750-752: Comment: Microscale materials are tested according to existing legislation with their fine particulate fraction, which are an inherent part of these products. Lines 750-752 are not appropriate.
4.4.4. Additional considerations	Scientific Committee of the Belgian Federal Agency for the Safety of the Food Chain	Line 736 - .. : an additional consideration of the “Trojan horse” effect with respect to FCM could be included: since ENM can be reactive towards proteins, lipids, …, they can also be reactive -when present in food- towards FCM (reaction of ENM with certain FCM chemicals and as such carrier of FCM into the food – cfr. comparable with food simulants to control specific migration limits)
4.4.4. Additional considerations	VdMi	line 750 to 752: Microscale chemicals are tested according to existing legislation with their fine particulate fraction, which are an inherent part of these products. Eliminate 750-752!

4.4.5. Conclusion on Toxicity of ENM	RIVM (National Institute for Public Health and the Environment)	L753-774: General remark on the conclusion on toxicity It is of extreme importance for interpretation of toxicity studies, that the form in which the metal reaches the internal organs is analyzed. Are it the nanoparticles itself that enter the body or are the ions, originating from the metal, or both entering the body? Although this point is already mentioned in the conclusions on toxicokinetics, this should also be incorporated here.
5. Environmental impact of nanotechnologies in food and feed area	Nanotechnology Industries Association	• 5. Environmental impact of nanotechnologies in food and feed area o Line 776-786: Comment: this paragraph postulates a risk on uses not present and thus information not available. The paragraph furthermore discusses the hazard, not the risk, as outlined in the title of this Opinion. This paragraph is therefore neither relevant nor appropriate.
5. Environmental impact of nanotechnologies in food and feed area	FAO	line 781: Other nanomaterials will be released into the environment intentionally, for example as pesticides or plant growth treatments. It would be therefore also important to consider the impact of such nano-agrochemical applications throughout their life cycle and their possible consequences in the food chain.
5. Environmental impact of nanotechnologies in food and feed area	CIAA	This section is sparse, therefore it might have been better not to include the section at all at this stage. line 776 to 786:This chapter postulates a risk on uses not present and thus information not available. Could please the risk and not only the hazard be looked at?. We thought that the aim of this paper is to look at risks!
5. Environmental impact of nanotechnologies in food and feed area	RIVM (National Institute for Public Health and the Environment)	General comment related to environmental issues: Although environment is captured in the name of the committee, and it is mentioned in the introductory text of the public consultation, chapter 5 on the environmental impact of nanotechnologies in food and feed area is very concise. To our opinion, this section needs to be extended or at least reference should be made to other documents where a more extended view is given on the fate and behavior of nanomaterials in different environmental compartments and their potential effects to ecosystems. L777 – 778: The definition of impact is not clear; we presume that the result of exposure and effect is meant. Both exposure and effect do not only depend on the characteristics and properties of the ENM itself, but also on the type product in which they are incorporated. L780 – 781: It is unclear how this will result in re-entry into the food chain. L776 – 781: An overview of relevant emissions to the different environmental compartments is lacking, e.g. - emissions during the production phase (surface water, air), - waste disposal by consumers (surface water via STP, groundwater via leakage from landfills, soil via landfills, air via waste incineration), - runoff of to surface water and leaching to groundwater of nano-agrochemicals and nano-pesticides, - manure of farmed animals fed with nano-containing feed (soil, leaching to groundwater). L782 – 783: It is unclear if migration from recycled material is seen as a problem for the food or for the environment. L783 – 784: It is unclear why the release of antimicrobial ENM is seen as a secondary effect on the environment.
5. Environmental	EFFAT	line 775

impact of nanotechnologies in food and feed area		We suggest a change in the title as follows: 5. Environmental impact and occupational hazard of nanotechnology in food and feed area after line 786, We suggest the addition of the following paragraph: Workers engaged in research, development, manufacture, packaging, handling, transport, use and elimination of nanomaterials and nanotechnology products will be most exposed, and therefore most at risk of any harmful effects. Health and safety at work must have priority in any nanomaterials surveillance system. Training, education and research is necessary in order to allow health and safety specialists (e.g. labour inspectors, preventive services, occupational hygienists, company physicians) preventing known and potential exposures to nanomaterials.
5. Environmental impact of nanotechnologies in food and feed area	Food Safety Authority of Ireland	It would be helpful if some of the "limited information" available on this topic was discussed more in this section.
5. Environmental impact of nanotechnologies in food and feed area	Friends of the Earth (Europe, Australia, Germany), EEB	The report barely addresses the relevant ecotoxicological studies and does not discuss the broader environmental impacts of nanomaterials use in food and agriculture. No data on toxicology are mentioned although there are studies available: Titanium dioxide: - Federici, G. Shaw. B, Handy, R. (2007). Toxicity of titanium dioxide nanoparticles to rainbow trout (*Oncorhynchus mykiss*): Gill injury, oxidative stress, and other physiological effects. *Aquatic Toxicol 84(4) pp.415-430.* - Hund-Rinke K, Simon M. 2006. Ecotoxic effect of photocatalytic active nanoparticles (TiO2) on algae and daphnids. Environ Sci Poll Res 13(4):225-232. - Lovern B, Klaper R. 2006. Daphnia magna mortality when exposed to titanium dioxide and fullerene (c60) nanoparticles. Environ Toxicol Chem 25(4):1132-1137. - Lovern, S. Strickler, J. Klaper, R. (2007). Behavioral and Physiological Changes in *Daphnia magna* when exposed to nanoparticle suspensions (Titanium Dioxide, Nano-C60, and C60HxC70Hx). *Environ Sci Technol 41, pp. 4465-4470.* Zinc oxide: - Luo J. 2007. Toxicity and bioaccumulation of nanomaterial in aquatic species. J U.S. Stockholm Junior Water Prize. doi: 10.2175/SJWP(2007)1:01 Antibacterial substances - Oberdörster G, Oberdörster E, Oberdörster J. 2005 . Nanotoxicology: an emerging discipline from studies of ultrafine particles. Environ Health Perspect 113(7):823-839. - Handy, R. Owen, R. Valsami-Jones, E. (2008). The ecotoxicology of nanoparticles and nanomaterials: current status, knowledge gaps, challenges, and future needs. *Ecotoxicol 17, pp. 315–325.* - Throback, I. Johansson, M,. Rosenquist, M. Pell, M. Hansson, M. Hallin, S. (2007). Silver (Ag(+)) reduces denitrification and induces enrichment of novel nirK genotypes in soil. *FEMS Microbiol Lett 270(2), pp.189–194.* CNT:

		- Templeton P, Ferguson P, Washburn K, Scrivens W, Chandler G. 2006. Life-Cycle Effects of Single-Walled Carbon Nanotubes (SWNTs) on an Estuarine Meiobenthic Copepod. Environ Sci Technol 40:7387-7393. - Cheng J, Flahaut E, Cheng S. 2007. Effect of carbon nanotubes on developing zebrafish (Danio rerio) embryos. Environ Toxicol Chem 26(4):708-716. Scott-Fordsmand, J. Krogh, P. Schaefer, M. Johansen, A. (2008). The toxicity testing of double-walled nanotubes-contaminated food to Eisenia veneta earthworms. *Ecotoxicol Environ Safety 71(3), pp.616– 619.* Broader environmental costs of nanomaterials production: In their evaluation of both top-down and bottom-up nano-manufacturing methods Şengül et al. (2008) found that the manufacture of nanoparticles has an unexpectedly high environmental footprint. This is related to the highly specialised production environments, high energy and water demands of processing, low yields, high waste generation, the production and use of greenhouse gases such as methane and the use of toxic chemicals and solvents such as benzene. The U.K. Royal Commission on Environment and Pollution was told that in one fullerene manufacturing process – which the Woodrow Wilson Center's Project on Emerging Nanotechnologies suggests can itself be highly energy intensive and polluting - only 10% of the finished product was usable, with the rest sent to landfill (U.K. RCEP 2008). In a life-cycle assessment of carbon nanofibres, Kanna et al. (2008) found that producing carbon nano-fibres may have the potential to contribute to global warming and ozone layer depletion, and cause environmental or human toxicity that is as much as 100 times greater per unit of weight than those of conventional materials like aluminium, steel and polypropylene. Khanna V, Bakshi B, Lee L. 2008. Carbon nanofiber production: Life cycle energy consumption and environmental impact. J Indust Ecol 12(3):394-410. Şengül H, Theis T, Ghosh S. 2008. Towards sustainable nanoproducts: An overview of nanomanufacturing methods. J Indust Ecol 12(3):329-359. More information on toxicity of ENM to the environment: www.foeeurope.org/activities/nanotechnology/Documents/Nano_food_report.pdf, page 33 ff It is very important to generate information on the amount of ENM disperse to the environment and to develop the understanding of environmental impact of ENM.
5. Environmental impact of nanotechnologies in food and feed area	VdMi	line 776 to 786: This chapter postulates a risk on uses not present and thus information not available. Could you describe the risk and not only the hazard. The aim of this paper is risk! As long as there is no proven use, there is no need for authorities to study hazards and risks of products in the responsibility of companies.
6. Proposed guidance for risk assessment (RA) of ENM in food and feed area	Nanotechnology Industries Association	• 6. Proposed guidance for risk assessment (RA) of ENM in food and feed area o Line 792-793: Comment: The NIA welcomes the fact that 'the Scientific Committee view is that the general [Risk Assessment] paradigm can also be applied to the RA of ENM in the food and feed area.' o Line 794-802: Comment: this paragraph implies that there is 'the possibility of additional endpoints [for ENM]', suggesting that there are entirely new toxicities that we can currently not. This conclusion needs to be clarified. It furthermore needs to be clarified if new guidance documents will be developed, since the EFSA Opinion concludes that

		'Current guidance documents in the food and feed area do not address ENM.' o The NIA welcomes the conclusion that 'RA of ENM needs to be carried out on a case-by-case basis'.
6. Proposed guidance for risk assessment (RA) of ENM in food and feed area	TNO Quality of Life, BU Quality & Safety, Zeist	In general we agree with the EFSA Scientific Committee view that the current used risk assessment paradigm is applicable to ENM, but needs to be expanded/ re-evaluated to consider whether the current used methods/ studies are also suitable for ENM. Although it is noticed that, due to a lack of sufficient data and information, it is difficult to give a detailed specific risk assessment guidance for food and feed applications, to our opinion some aspects could be further clarified. To our opinion at this stage there is a need for: - a sound definition of ENM; - recommendations how to test if (and how fast) nano-sized materials dissolve in a couple of key physiological fluids - recommendations on how detect/measure and how to analyse (metrics) ENM in food/feed. - a screening approach on the basis of which a differentiation can be made between safe use of nanoparticles/nanomaterials and situations which pose a clear risk to human health and the environment, without performing a full risk assessment.
6. Proposed guidance for risk assessment (RA) of ENM in food and feed area	The UK Government Chemist	799-802 I agree; routine analytical methods capable of measuring a wide range of ENM are unlikely to emerge in the near future.
6. Proposed guidance for risk assessment (RA) of ENM in food and feed area	FAO	line 848: This is an important point, especially in the case of nutrients where the gap between optimal and dangerous levels is narrow. This is the case for some lipid soluble vitamins which are potentially toxic if absorbed rapidly or completely.
6. Proposed guidance for risk assessment (RA) of ENM in food and feed area	CIAA	line 788:the properties between macroscale (beyond nanoscale) and dissolved chemical (smaller than nanoscale) as defined and those of nanoscale is not fully explained. 802 :Does this mean new guidance is going to be given? 807 :It is necessary to include here what is the proposed function of the ENM. What property of the ENM has been changed from bulk to make it necessary to use it in this form in the food/feed. How does it differ in this respect from the bulk material. It is this change in the property that is likely to have a significant impact on the risk assessment.
6. Proposed guidance for risk assessment (RA) of ENM in food and feed area	Friends of the Earth (Europe, Australia, Germany), EEB	The currently used risk assessment paradigm (hazard identification, hazard characterization, exposure assessment and risk characterization) is not applicable for ENM. RA methods must be modified re. the special properties of ENM. It must be recognised the problems inherent in using mass-based metrics for nanomaterials dose. Many (although not all) manufactured nanoparticles are more toxic per unit of mass than larger particles of the same chemical composition. The Project on Emerging Nanotechnologies at the Woodrow Wilson International Center for Scholars has suggested that the toxicological impact of 58,000 tonnes of manufactured nanomaterials might be the equivalent of 5 million or even 50 billion tonnes of conventional materials. Maynard A. (2006). Nanotechnology: A research strategy for addressing risk. Woodrow Wilson International Center for Scholars Project on Emerging Nanotechnologies. Available at: http://www.nanotechproject.org/file_download/files/PEN3_Risk.pdf (accessed 24th November 2008).

		EFSA also fails to evaluate the health and environmental risks and challenges of more complex next generation nanoproducts and to examine whether or not existing risk assessment methodologies can be effectively modified to cope with these. This was a key concern of the recent UK Royal Commission on Environmental Pollution. U.K. RCEP. (2008). Novel materials in the environment : The case of nanotechnology. Royal Commission on Environmental Pollution. Available at: http://www.rcep.org.uk/novel%20materials/Novel%20Materials%20report.pdf (accessed 14 November 2008). We object to EFSA framing the risk assessment of nanofoods, nano-packaging and nano-agricultural inputs as necessary to achieve the public health benefits of such products that are claimed by industry proponents. These claims, which are communicated uncritically by EFSA at the outset of the draft opinion, are never subject to the careful assessment they warrant. Friends of the Earth suggests that it is unacceptable to use claims of public health or social benefits to counterbalance or justify new toxicity risks and the introduction of even more highly processed foods without subjecting claims of advantage to the same level of scrutiny as claims of risk. We agree on the important statements: ▪ "If there is no information of disappearance of the nanostructure, it shall be assumed that the nanoform is still present in the GI tract."" ▪ "As it is difficult to analyse food and feed for the presence of ENM, a conservative approach in the RA is to assumed that the entire amount of ENM added or migrating from FCM is present in its nanoform."
6. Proposed guidance for risk assessment (RA) of ENM in food and feed area	RIVM (National Institute for Public Health and the Environment)	L841-842: "In that case, it shall be assumed that it still is present in its nanoform". Please indicate that this is a conservative approach. L851: "These tests should be able to pick up toxic effects of ENM". This is probably true for most toxic effects, so maybe include the word "most" in this sentence. L859: "administration via gavage is a more well-defined mode". Please explain what is meant with this phrase. L865-866: "They are generally suited for screening purposes and studies on mechanisms of toxicity". Do they refer to in vitro assays for mutagenicity/genotoxicity and oxidative stress or for in vitro assays in general?
6. Proposed guidance for risk assessment (RA) of ENM in food and feed area	ILSI Europe aisbl	Line 801: risk assessment should always be carried out case by case; this does not only apply to ENM.
6. Proposed guidance for risk assessment (RA) of ENM in food and feed area	DSM Nutritional Products Ltd	4. Provide guidance on the data needed for risk assessment 4.1 Guidance We welcome the provision of guidance, given the caveats discussed above. 4.2 Recommendations We are fully in agreement with the Recommendations, but would be grateful if recommendation at 962 is modified: "expanded to explain that in respect to in vivo studies alterations to existing protocols should be suggested (rather than new methods developed)."
6. Proposed	Food Safety Authority	Line 788: "Properties of materials at nanoscale may be different from chemicals in the nanoscale or dissolved forms, and

guidance for risk assessment (RA) of ENM in food and feed area	of Ireland	existing toxicological knowledge on chemicals cannot be fully extrapolated to ENM". It is difficult to make comparisons between different forms of a substance if they are referred to differently. Here it is difficult to know what the sentence means as we do not know if the materials and chemicals mentioned are of the same substance?
Overall Conclusions and Recommendations	Scientific Committee of the Belgian Federal Agency for the Safety of the Food Chain	The overall conclusions and recommendations are clearly formulated. The possible risks are numerous and the indications or evidence for the risks are scarce to nonexistent. Anticipation and vigilance are necessary but without turning nanoscience and -technology into a bogey. Probably the technology itself will help answering the questions that are raised in this document e.g. through proper diagnosis etc.
Overall Conclusion and Recommendations	Friends of the Earth (Europe, Australia, Germany), EEB	There is a discord between EFSA's findings and its recommendations. EFSA recognises that: many nanomaterials pose serious new toxicity risks; that the toxicological properties of nanoparticles cannot be predicted from what we know of the particles' properties in bulk form; and that existing risk assessment methodologies require modification for nanomaterials. However its recommendations are largely confined to calling for further risk research and the better development of characterisation, detection and monitoring methodologies. We suggest that based on EFSA's own findings, as well as those of SCENIHR, the United Kingdom's Royal Society and other high level scientific investigations, there must be clear recommendations for nanomaterials to face mandatory risk assessment as new chemicals, using nanomaterial appropriate risk assessment methodologies and metrics, before they are permitted for use in commercial products. Once they have passed safety and social benefit tests, nano ingredients and nano-formulated foods should be labelled to give consumers the capacity to make an informed choice about eating such foods, and to enable any adverse public health effects to be traced to their source.
Conclusions	RIVM (National Institute for Public Health and the Environment)	L883: Which section is the guidance section? Section 6 (proposed guidance for risks assessment of ENM in food and feed area) or the recommendations?
Conclusions	ILSI Europe aisbl	Line 906: as argued also in our comments on other sections, we consider it misleading to note that "the adequacy of currently existing toxicological tests to detect all aspects of potential toxicity of ENM has yet to be established". Inability to detect toxicity is an aspect of the test, not of the material being tested, so ENM would not differ from macro materials in this respect.
Recommendations	UK Food Standards Agency	• It would be helpful to prioritise the long list of recommendations (pg 23-24) in order of importance and urgency • There is an urgent need for toxicological data on a representative selection of nanomaterials
Recommendations	CIAA	line 931:Nanotechnology needs to be specified 971:Is it MnO or Zn or Mn like in line 750
Recommendations	VdMi	line 931: Nanotechnology needs to be specified 971: Is it MnO or Zn or Mn like in line 750?
Recommendations	BioIndustry Park del Canavese	I was not able to find in the opinion any reference to the metallic nanoparticles used by craftsmen since very early times. In the old Mesopotamia, as well as in Middle Ages and Renaissance, silver and copper nanoparticles were created by the artisans by adding copper and silver salts, oxidizing them, and finally reducing the ions at 600° back to metals, so forming the nanoparticles that give a nice optical effects. Since these nanoparticles were used to decorate recipients for food and beverages, it is conceivable that the specific nanoparticles involved do not have any acute toxic effects when ingested.
Additional	Scientific Committee	Line 947/952: should the presence of impurities such as residual catalyst (referred to on line 347) not be addressed in this list of

recommendations	of the Belgian Federal Agency for the Safety of the Food Chain	recommendations (considerations)?
Additional recommendations	The UK Government Chemist	936-7 More complex ENM offer brighter prospects for the analytical chemist, as they may produce highly definitive chemical fingerprints which can be used to track the in vivo distribution of whole particles, or even of particular fragments as they are metabolised.
Additional recommendations	RIVM (National Institute for Public Health and the Environment)	L965-967: Special attention should also be given to the potential effects of ENM on the development of chronic digestive diseases and food allergy.
Additional recommendations	EFFAT	After line 937 We suggest the addition of the following bulletpoint: Generate information by the set-up of a mandatory declaration and registration system with EFSA before the marketing and distribution of any food/feed containing ENM or with FCM containing ENM on the European Community territory. The declaration by the producer shall include adequate information on the type, quantity, nature of processing of ENM and location of the factories. Line 976 We suggest the following addition: "In relation to impacts on the environment and occupational hazard" After line 978 We suggest the addition of the following bulletpoints: Investigate the occupational hazard related to the research, development, manufacture, packaging, handling, transport, use and elimination of nanomaterials and nanotechnology products, in cooperation with the services of the European Commission, with the European Agency for Safety and Health at Work, the European Centre of Disease Prevention and Control and the European Environment Agency and stakeholders.
References	Federal Institute for Risk Assessment	The following BfR-publications are available (www.bfr.bund.de): Nanotechnology: Health and environmental risks of nanomaterials – Research Strategy –, ed. BAuA, BfR, UBA. 2007, 79 p. BfR Consumer Conference Nanotechnology Pilot project to identify consumer risk perception, ed. R. Zimmer, R. Hertel, G.-F. Böl. BfR-Wissenschaft 03/2008, 86 p. Public Perceptions about Nanotechnology Representative survey and basic morphological-psychological study, ed. R. Zimmer, R. Hertel, G.-F. Böl. BfR-Wissenschaft 05/2008, 117 p. Risikowahrnehmung beim Thema Nanotechnologie – Analyse der Medienberichterstattung, ed. R. Zimmer, R. Hertel, G.-F. Böl. BfR-Wissenschaft 7/2008, 214 p. In addition, the BfR as National Focal Point got a comment from

		Prof. Dr. Jörg Kreuter Institut für Pharmazeutische Technologie Biozentrum-Niederursel Johann Wolfgang Goethe-Universität Max-von-Laue-Strasse 9 D-60438 Frankfurt He added 5 publications showing uptake of nanoparticles from the gastrointestinal tract after oral administration (up to 19% of the administered dose). These papers will be send elec-tronically to EFSA. The references are: Araujo L, Sheppard M, Löbenberg R, Kreuter J. Uptake of PMMA nanoparticles from the gas-trointestinal tract after oral administration to rats: modification of the body distribution after suspension in surfactant solutions and in oil vehicles. International Journal of Pharmaceutics 176 (1999): 209-224 Landry F B, Bazile D V, Spenlehauer G, Veillard M, Kreuter J. Peroral Administration of 14 C-Poly(D,L-Lactic Acid) Nanoparticles Coated with Human Serum Albumin or Polyvinyl Alcohol to Guinea Pigs. Journal of Drug Targeting (1998) 6, No 4:293-307 Löbenberg R, Araujo L, Kreuter J. Body distribution of azidothymidine bound to nanoparticles after oral administration. European Journal of Pharmaceutics and Biopharmaceutics (1997) 44:127-132 Kreuter J. Peroral administration of nanoparticles. Advanced Drug Delivery Reviews (1991) 7:71-86 Nefzger M, Kreuter J, Voges R, Liehl E, Czok R. Distribution and Elimination of Polymethyl Methacrylate Nanoparticles After Peroral Administration to Rats. Journal of Pharmaceutical Sciences (1984) 73:1309-1311
Glossary / Abbreviations	CIAA	line 1351:It might be better not to use expressions not contained in the pre-standard DIN ISO CEN TS 27682.
Additional considerations	VCI	4.4.4. Additional considerations 734 Some other aspects increase the uncertainty in assessment of ENM. The presence of ENM in 735 food might affect normal food components or contaminants. Hence, food containing ENM with 736 actively charged surfaces can absorb proteins, lipids, nucleic acids and carbohydrates. It has 737 been speculated that absorption of ENM is accompanied by transport of food 738 components/molecules that are not normally absorbed and thus may create an (unwanted) port 739 of entry **("Trojan horse" effect)**, and that this might change their toxicity (Lomer et al., 2002; 740 Borm and Kreyling, 2004). If particles that pass through the epithelial cells via transcytosis by 741 M-cells this may lead to accumulation within the Peyers Patches and subsequently a possible 742 immune reaction. The surface properties (e.g. coatings) that increase the active uptake of 743 encapsulates might also be a reason for concern. Thus, lectins used for coatings of nano 744 encapsulates can be cytotoxic or induce inflammatory responses (Govers et al., 1994; Des 745 Rieux et al., 2006). 775 **5. Environmental impact of nanotechnologies in food and feed area**

776 During production, use and disposal of ENM in the food and feed area, dispersal of ENM to the
777 environment is likely. Possible environmental impacts are influenced by the characteristics and
778 properties of the ENM and may be more or less pronounced depending on the specific ENM. In
779 some instances, there is the possibility of re-entry of certain ENM as contaminants in the food
780 and feed chain. Such contamination may arise from the traditional processes of food and feed
781 waste disposal, e.g. via sewage, from waste incineration or leakage from landfills.
782 **Recycling processes of food packaging material** containing ENM should be considered, as the
783 process may affect the migration of the ENM in the recycled material. There may also be
784 **secondary environmental implications during disposal from possible release of antimicrobial**
785 ENM from FCM. However, there is presently only limited information available of these
786 processes related to ENM in food and feed.

787 **6. Proposed guidance for risk assessment (RA) of ENM in food and feed area**
788 Properties of materials at nanoscale may be different from chemicals in the macroscale or
789 dissolved forms, and existing toxicological knowledge on chemicals cannot be fully
790 extrapolated to ENM (e.g. SCENIHR, 2007a). A number of national and international advisory
791 committees have recommended strategies for the RA of ENM (e.g. SCENIHR, 2007a; SCCP,
792 2007). In agreement with these, the Scientific Committee view is that the general paradigm can