### RESEARCH

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# Environmental impacts of nanomaterials: providing comprehensive information on exposure, transport and ecotoxicity - the project DaNa2.0

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#### Abstract

**Background:** Assessing the impact of new technologies or newly developed substances on our environment is a challenge, even more so if the applied test methods - both toxicological and analytical - are often found to be inadequate and need amendments or even new developments as it is in the case of nanotechnology. This is illustrated by numerous publications in the field of nano-ecotoxicology which although they have been investigating the impact of a number of nanomaterials on several organisms almost never allow for explicit statements on potential hazards of these nanomaterials. This fact not only hampers the knowledge communication to all non-scientists (e.g. consumers) but it also complicates the transfer of the obtained results for other scientists.

**Results:** Risk communication is an essential and thus integral part of risk management. For this purpose, the project DaNa2.0 (Data and knowledge on nanomaterials - processing of socially relevant scientific facts) provides processed and hence easy accessible information on the potential safety issues of nanomaterials, mainly via the project website www.nanoobjects.info. This will allow various stakeholder groups to get impartial information on potential effects of nanomaterials and help consumers to make informed decisions. DaNa2.0 is funded by the German Federal Ministry of Education and Research (BMBF) and supported by the Swiss government as well as by the European InterReg IVb programme. The DaNa2.0 team is an interdisciplinary group of scientists from different areas such as materials science, chemistry, biology and human and environmental toxicology. Extending the project team in DaNa2.0 with European experts allows for broadening of the existing knowledge portfolio by adding further cross-cutting topics and increasing our expertise, e.g. in the field of environmental exposure and fate.

**Conclusions:** On the project website www.nanoobjects.info, a unique link between nanomaterials in practical applications (e.g. environmental remediation) and their potential impacts is provided. The focus of this publication will be on all issues with environmental relevance, which are addressed in the 'Knowledge Base Nanomaterials' on the project website. These issues include environmental exposure and behaviour of nanomaterials and nano-ecotoxicology.

**Keywords:** Nanomaterials; Nano-objects; Nano-ecotoxicity; Knowledge base; Knowledge dissemination; Nanotechnology; Environmental impact

#### Background

Since the start of the DaNa project with its corresponding website www.nanoobjects.info in 2009 and its continuation in DaNa2.0 since 2013, the annual number of publications dealing with toxicological aspects of nanomaterials has been increasing exponentially. Numerous research papers on various nanomaterials and organisms

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have been published (as demonstrated by extensive reviews, e.g. [1,2]), resulting in a growing number of nanomaterials (NM) and their related information on the website. Besides the scientific output of these literature sources, there is an increasing demand for more information on this new technology, reflected in the growing number of visitors to the project website.

However, it is still difficult to provide simple and explicit statements on potential environmental hazards of nanomaterials. This is either due to a lack of information, e.g. regarding the physico-chemical properties of nanomaterials,

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or due to a high variation in test procedures and consequently to a high variability in test results. Beyond that, the transfer of nano-toxicity-related information to scientist from other fields and also to non-scientists is difficult. To bridge this gap, the project DaNa2.0 is working on improving these communication pathways from nanosafety-related science to various stakeholder groups. The major goal of the DaNa Knowledge Base Nanomaterials is to provide condensed, up-to-date and easy comprehensible information on the safety of nanomaterials for different recipient groups simultaneously [3-5]. The website provides a unique combination of material science-based information and nano-applications with toxicological information, helping, e.g. consumers in making informed decisions.

In order to assess the quality of the published studies and to consequently achieve an aggregation to the ones meeting the requirements of good scientific practice, the DaNa expert team developed the so-called Literature Criteria Checklist. The list of chosen criteria allows the side-by-side evaluation of background information on the applied nanomaterial together with the selected toxicological assays [4]. Ultimately, only those scientific findings complying with the checklist will be integrated in the knowledge base. This evaluation process along with the writing of the respective articles is achieved by a multidisciplinary core team that is supported since the start of the follow-up project DaNa2.0 (2013) by several external experts bringing in their expertise in specific fields.

In order to cover the full scope of potential interactions of nano-related applications with the various organisms, the DaNa2.0 expert team evaluates all scientific research papers with regard to human and ecotoxicological aspects thereby continuously updating the articles on the 25 market-relevant nanomaterials listed on the website [3].

This paper focuses on the DaNa2.0 activities with regard to environmental aspects, namely exposure and behaviour in the environment, as well as potential hazards towards environmental organisms. Furthermore, the website provides an overview of the German Nanosafety Research Landscape. On the DaNa2.0 internet platform, research projects funded by the German Federal Ministry of Education and Research (BMBF) are provided with the opportunity to present their project results related to environmental issues of nanotechnology in a recipient-friendly way.

#### **Results and discussion**

## DaNa2.0 - general structure of the platform (www.nanoobjects.info)

The content of the website is structured into different areas providing different types of information on the opportunities and potential risks of nanotechnology:

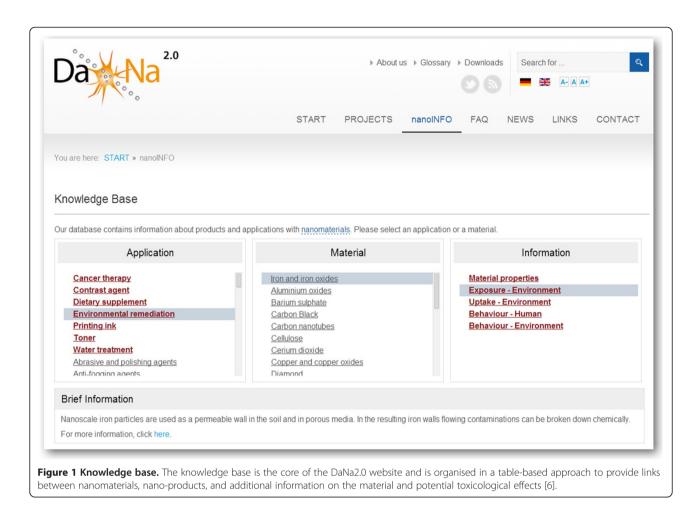
- 1. Knowledge base provides comprehensive articles on application, material properties, exposure, behaviour and toxicity of currently 25 engineered nanomaterials (ENMs)
- 2. Basics provides short information on basic issues from release to uptake behaviour, risk and fate of ENM
- 3. Cross-cutting section provides comprehensive articles on over-arching issues in nanotechnology, significant to more than one ENM
- 4. Methodology provides a collection of nano-relevant standard operating procedures (SOPs) (template and finalised documents) together with documents used for quality management of cited literature (Literature Criteria Checklist)
- 5. Projects section provides an overview on relevant funded national projects
- 6. Frequently asked questions (FAQ) section provides a collection of questions and answers to relevant nano-related issues and opportunity to directly ask questions to the DaNa2.0 experts
- 7. Glossary provides short definitions of related terms used in the articles
- News section provides announcements of new developments in nanotechnology, important publications, conferences and workshops, and more
- 9. Links section provides links to websites, networks, information platforms as well as national and international projects on nanotechnology and safety issues

The content of the articles within the knowledge base and also in the cross-cutting section is presented in three levels of complexity according to needs and interest of different recipient groups:

- 1. Short and basic interested citizens/consumers (public oriented)
- 2. Detailed journalists, stakeholders, scientists from other fields
- 3. Complex scientists from related fields, regulators

#### DaNa2.0 Knowledge Base Nanomaterials

The core section of the DaNa2.0 web platform is the knowledge base which provides a wealth of facts and data for currently 25 market-relevant nanomaterials. The actual state of knowledge is derived from evaluation of the scientific literature, from project reports and official reports from governmental agencies or organisations. An application-based database was established, as most visitors rather seek information related to a specific product than to a specific type of nanomaterial (Figure 1). The table-based approach allows the selection either of an application, e.g. 'environmental remediation' and



'textiles' (Figure 1, column 1), or of an ENM of interest, e.g. 'iron oxide' (Figure 1, column 2). In turn, the nanomaterial used in the respective type of application or the type of application belonging to an ENM is highlighted. Several applications with an environmental background are already included and the total list of applications which is updated on a regular basis. Having selected the desired application together with a material, the availability of further information on exposure, uptake, behaviour and the material is indicated (Figure 1, column 3). By selecting an issue of interest, the brief information box below the table not only provides a comprehensive statement on the state of knowledge but also allows accessing more complex information. The interested visitor is then navigated to the more in-depth content of the main knowledge base. Likewise, all 25 ENMs included in the knowledge base can also be accessed directly via the material menu.

The in-depth content of the knowledge base provides besides general material information (production, usage, properties) sections on exposure, uptake and behaviour with regard to human and environmental toxicology and potential risks for environmental organisms (e.g. fish, plants, mussels; see Table 1). Figure 2 shows an exemplary excerpt from the website for the nanomaterials 'iron and iron oxides' that deals with 'uptake and potential risks for environmental organisms'. The introduction of organism icons is currently introduced into the articles to facilitate the navigation and information access.

An overview on the huge variety of organisms, primary cells and cell lines used to study potential hazardous effects of nanomaterials is given in Table 1, summarising the 25 market-relevant ENMs currently listed in the knowledge base together with the corresponding environmental test systems. For 23 out of the 25 listed ENMs in the knowledge base, there is ecotoxicological information available. As for the human toxicology, the body of literature on environmental effects of nanomaterials is growing rapidly resulting in the aforementioned problems with comparability. In most cases, different types of the same nanomaterials were used or experimental conditions varied from study to study making it increasingly difficult to interpret and compare the results [9]. In order to deal with these issues, the DaNa expert team developed the 'Literature Criteria Checklist' to manage and monitor the quality

Nanomaterial	Environmentally relevant application or product with likely release to the environment	Anticipated most relevant release path/way	Studied organisms/cells/cell lines considered in the knowledge base at www.nanoobjects.info		
Aluminium oxide	Abrasive and polishing agents	Aerosols	Mud tube worm, shrimp, earthworm, basket shell, nematode, bacteria, daphnids, zebrafish, thale cress, rye, lettuce, corn, carrots, soy, cabbage, cucumber, radish, rapeseed, ryegrass		
Barium sulphate	Contrast agent	Waste water	No ecotoxicity studies available		
Carbon black	Printing ink, toner	Aerosol	Common mussel, brown algae (toothed wrack), fruit fly, amphipods		
Carbon nanotubes (CNT)	All applications anticipated so far involve CNT embedded in a matrix		Rainbow trout, earthworm, lugworm, tomato, cabbage, carrot, onion, radish, rape, lettuce, ryegrass, corn and cucumber		
Cellulose	Wound dressing	Solid waste	Bacteria, fungi, daphnids, fish, algae, rainbow trout liver cells		
Cerium dioxide	Diesel additive	Car exhaust	Bacteria, algae, zebrafish (embryo, adult), daphnids, rainbow trout liver cells, soybean		
Copper and copper oxide	Wood preservatives	Waste	Bacteria, protozoa, worms, snail, frog embryo, zebrafish, green algae, radish, ryegrass, duckweed, corn		
Diamond	Abrasive and polishing agents	Aerosol	Nematodes, frog embryos, daphnids		
Fullerenes	Cosmetics, sports equipment, lubricant	Direct release to surface or waste water	Bacteria, algae, daphnids, common mussel blood cells, oyster (larvae, adult, liver cells), zebrafish embryo		
Gold	Diagnostics and therapy	Waste water	Bacteria, daphnids, blue mussel, basket shell, zebrafish embryo, rainbow trout liver cell, cucumber, lettuce		
Graphene	No applications on the market, yet		Bacteria, nematodes, zebrafish embryo, tomato, spinach, cabbage		
Indium tin oxide (ITO)	All applications anticipated so far involve ITO embedded in a matrix		Various aquatic organisms (vertebrates, algae)		
Iron and iron oxides	Ground and waste water remediation	Intended release to ground water	Bacteria, medaka embryos, rainbow trout gill cells, pumpkin, lima bean, ryegrass		
Platinum	Catalytic exhaust converter	Aerosol formation	Zebrafish embryo, rainbow trout gill cells		
Quantum dots (QD)	All applications anticipated so far involve quantum dots embedded in a matrix		Bacteria, algae, daphnids, zebrafish embryo, rainbow trout		
Silicon dioxide	Skin care, textiles, therapeutics	Surface and waste water	Bacteria, green algae, mussel blood cells		
Silver	Textiles, antimicrobial applications	Wash-off, release of ions from silver-coated surfaces	Bacteria, fungi, green algae, daphnids, zebrafish embryo, medaka cell line		
Strontium carbonate	Pyrotechniques	Aerosol	No ecotoxicity studies available		
Titanium dioxide	Sunscreen, surface coating	Direct release to surface waters, run-off from coated surfaces	Bacteria, daphnids, nematodes, lugworm, mussel, snail, woodlice, rainbow trout, zebrafish (embryo and adult), green algae, onion, willow, tobacco		
Titanium nitride	All applications anticipated so far involve TiN embedded in a matrix		Rainbow trout gill and intestinal cells		

#### Table 1 Overview of all 25 market-relevant nanomaterials listed currently in the DaNa knowledge base

#### Table 1 Overview of all 25 market-relevant nanomaterials listed currently in the DaNa knowledge base (Continued)

Tungsten carbide (WC)	WC nanoparticles are used for tool production and are tightly bound in the tools		Rainbow trout gill cells
Tungsten carbide cobalt (WC-Co)	WC-Co nanoparticles are used for tool production and are tightly bound in the tools		Rainbow trout gill cells
Zeolite/clays	Fertiliser	Soil	No ecotoxicity studies available
Zinc oxide	Sunscreen, textiles	Direct release to surface waters, wash-off	Bacteria, protozoa, woodlice, zebrafish, rainbow trout, daphnids, green algae, ryegrass, corn, soybean, zucchini, thale cress
Zirconium dioxide	All applications anticipated so far involve $ZrO_2$ embedded in a matrix		Bacteria, algae, zebrafish embryo

Relevant environmental applications, the anticipated most relevant release path into the environment and the test organisms used in the studies were considered in this summary. As the whole ENM life cycle was not considered, disposal as relevant release at end of life was left out.

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both in adult medaka and in embryos, high concentrations of iron nanopar leginning of exposure and subsiding subsequently <sup>[5]</sup> . Moreover, modifications rere found in tissues of the gills and intestines of adult fishes. The viability iffected by the iron oxide particles <sup>[6]</sup> . Although gills of the common mussel tal inction after exposure proved to be minimal. Interestingly, the nanoparticles ompared with soluble iron salt <sup>[7]</sup> . Pumpkin plants growing in water containing iron oxide particles were found to "here was no uptake when the plants were grown in soils containing particles. In inchanged and appear normal compared with the untreated specimens. Interest of capable of taking up particles <sup>[6]</sup> . Moreover, another type of pumpkin and the omparable particle concentrations in hydroponic water, did not take up coated in released less ions compared to cop	caused by contact of of isolated cells of g ke up iron oxide par s did not exhibit inc take up particles in n spite of uptake, thk tingly, another plant he rye grass, which	of the particles with tissues jills of rainbow trout is not ticles, the changes in their reased or different effects leaves, roots, and sprouts. e plants were found to grow species, the lima bean, was both had been treated with	Cell with a cl nanoparticle in the cell to to orientate i magnetic fiel	hain of magnetile that are intercalated enable the bacterium steff on the earth's d @ Scheffel et al. he nano-form is assum	bine studies report a higher toxicity s one of the substances known to to d snails. noparticles are toxic to bacteria <sup>[1,2,</sup> e bacteria and induce <u>oxidative stree</u> he toxic effects differed depending of few ions and were not toxic.
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of all used literature prior to inclusion and publication in the knowledge base (see respective chapter).

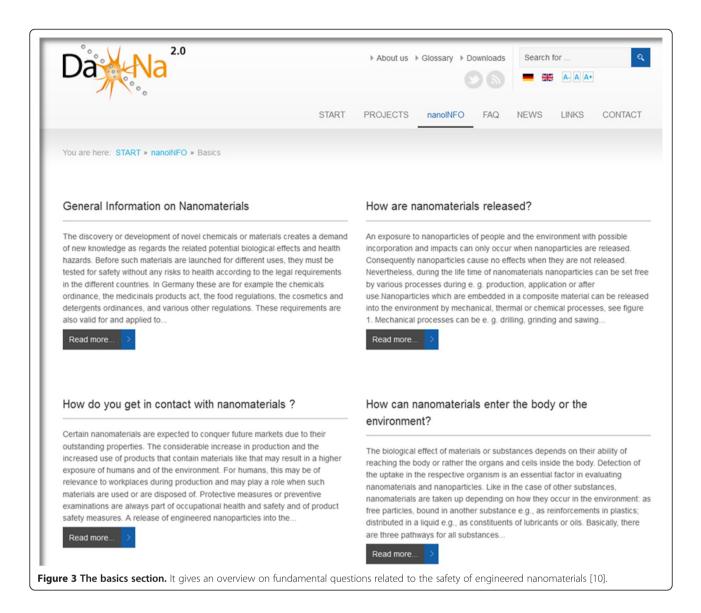
In general, consumer products with a high likelihood of the applied ENM to get in close contact to the user such as sunscreen or textiles are of biggest concern for humans whilst at the same time these ENMs are also the most prone to enter our environment. Therefore, these ENMs have also a great potential to be hazardous for the environment and its inhabitants, and ultimately, a polluted environment will always act back on humans and their conditions of living. Keeping this in mind, the DaNa2.0 team supported by the external experts is working continuously on constantly updating the state of knowledge in these fields. Specifically for environmental issues, the external experts provide background in (environmental) risk assessment, ecotoxicity, NM release and transport, and exposure assessment.

#### Other sections with environmental relevance Basics section

The basics section provides fundamental introductions into nanotechnology and safety issues, such as general information on release of nanomaterials, uptake and behaviour in organisms and environmental media, as well as a section on how to assess the potential risk NM pose to humans and the environment (Figure 3).

#### Cross-cutting section

This section deals with overarching issues with significance to ENMs in general. The two articles currently published on the website deal with coatings and with different crystal structures of ENM, respectively. Both issues are not only of relevance to materials scientists but also need consideration in (eco)toxicological testing. This section will be further expanded during the term of



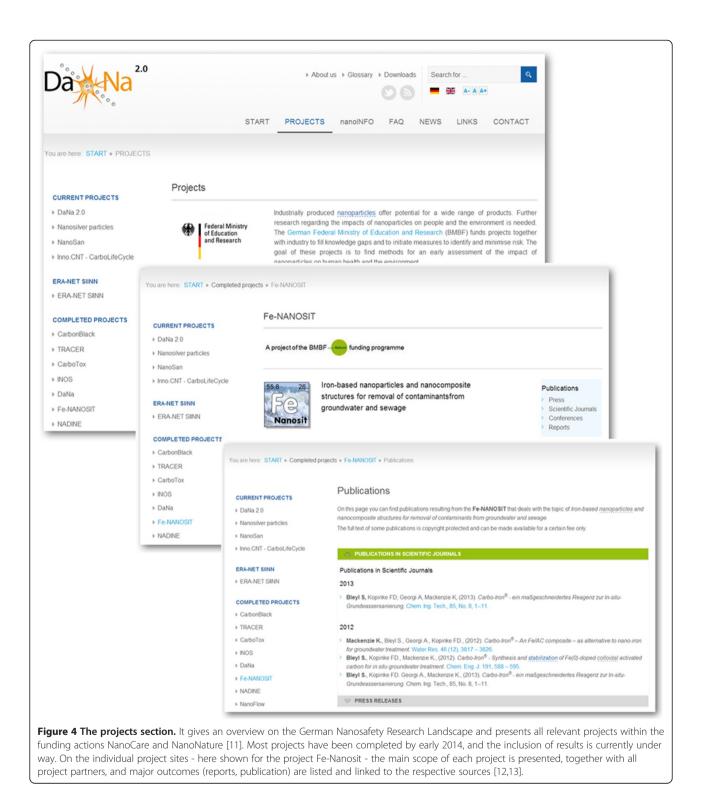
the project by, e.g. including articles on ENMs in paints or on ENM detection in the environment.

#### Projects and standard operating procedures

Besides the knowledge base, another main focus of DaNa2.0 is to give an overview of the German Nanosafety Research Landscape including previous, current and future nano-(eco)toxicology-related projects funded by the BMBF. Twelve projects were part of the funding action NanoNature (running from 2009 to 2014), referring mostly to environmental applications. The projects conducted research on, e.g. the improvement of catalytic processes or filtration techniques by applying nanotechnology. Some projects also covered the assessment of potential ecotoxicological hazards of the nanomaterials investigated (e.g. Fe-Nanosit). Three projects dealt with iron-based nanoparticles or nanocomposites applied

in environmental remediation, specifically the decontamination of ground water (e.g. Fe-Nanosit, see Figure 4). Most of the funded projects have been completed until early 2014. Hence, future tasks for the DaNa2.0 team are to first display these results (reports, publications, etc.) on the respective project pages and next to implement these new findings, if having met the necessary quality criteria to enter into the knowledge base (Figure 2). Furthermore, SOPs developed within the projects will be published via the website. New projects will start soon and will be likewise presented.

Additionally, the DaNa2.0 team supported the exchange and interaction between projects from the funding initiatives NanoCare and NanoNature with annual cluster meetings which are planned to continue once the new projects have started. One successful outcome of these cluster meetings was the creation of an interest group



of ecotoxicologists focussing on the specific peculiarities of iron-based nanomaterials that need to be considered in ecotoxicity testing. This interest group for example organised a workshop focusing on physicochemical characterisation of these nanomaterials before conducting ecotoxicological tests. Accounting for the need to harmonise experimental practices, the DaNa2.0 team in close cooperation with project members from the nano-funding initiatives developed a template for SOPs. The aim was to provide a common format to share, compare and describe methodologies specific to nanomaterials. But the template

may also be used by other projects or serve as a basis for future projects and publications. The pdf form is available for download via the website [14].

#### Frequently asked questions

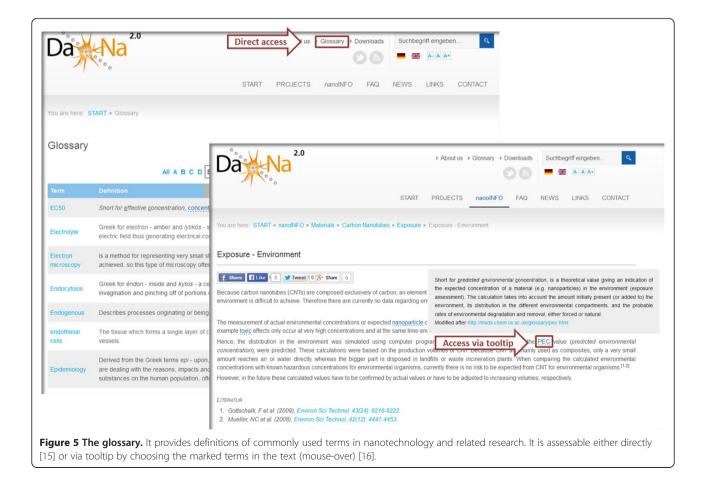
The *Frequently asked questions* (FAQ) section gives answers to the most common and important questions on nanomaterials and nanotechnology. These include questions specifically relating to environmental issues, e.g. *How are nanoparticles displayed in the context of recycling management? How can I recognize whether a product contains nanoparticles? How dangerous are nanoscale particles that are already present in the environment?* There is also the possibility to directly interact with the DaNa2.0 experts by submitting questions related to nanomaterials via email. The DaNa2.0 team will respond and publish the answer on the website if considered to be of public relevance.

#### Glossary

In some instances, the usage of terms very specific to nanotechnology and related fields is hard to avoid. In order to provide further explanation, the glossary not only gives quick and easy definitions for general terms like 'zeta potential', 'agglomerate' or 'surface charge' but also covers terms from environmental sciences, such as 'bioaccumulation' or 'predicted environmental concentration'. It is assessable either directly in alphabetic order or via tooltip by choosing the marked terms in the text (see Figure 5).

#### Literature criteria checklist

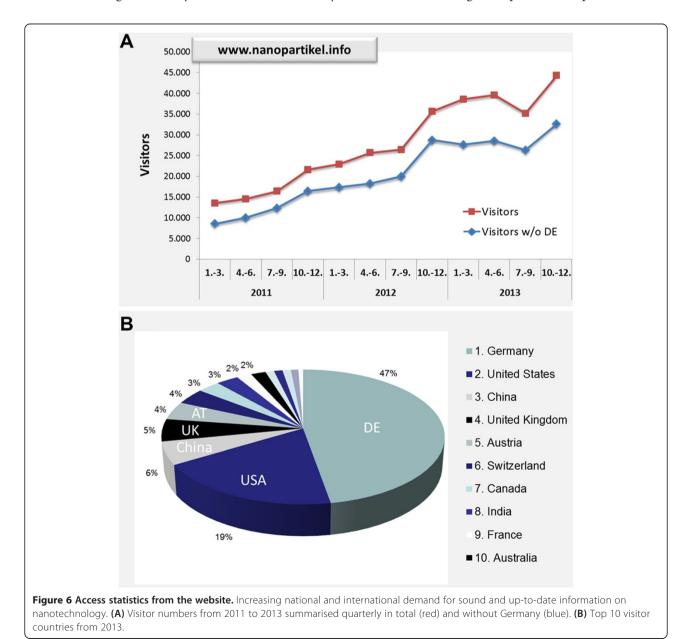
The need to develop the DaNa Literature Criteria Checklist arose from the necessity to manage the evaluation of all nano-toxicity data regarding its scientific value prior to including it in the knowledge base. In this way, all data with insufficient background information on the investigated ENMs or on the applied methodology will be excluded from the knowledge base, as these data will lead to false conclusions on potential toxic effects of nanomaterials. The assessment criteria cover (1) the extent of the physical-chemical characterisation of the ENMs, (2) the toxicity testing procedures and (3) general issues (e.g. data evaluation). In addition, mandatory assessment criteria are specified, in distinction to desirable criteria. The complete list of quality criteria is accessible to all interested scientists via the DaNa2.0 website [17]. Evaluating every scientific publication according to the criteria finally allows for selection of solely those papers that provide sound



background information on the nanomaterials and the toxicological tests applied [4]. Hence, only scientific facts complying with the checklist will be included in the knowledge base.

#### Dissemination of nanotechnology-related information

The main aim of the teams' dissemination activities is to provide the general public with sound and up-to-date information related to nanotechnology. The most important instrument for this task is the website www.nanoobjects. info. Since the launch of the website in 2009, a constant increase in the visitor numbers was observed, demonstrating a high need for information (Figure 6). The statistical data shown in Figure 6 clearly demonstrate that not only the overall visitor numbers are increasing but also there is a growing international awareness level for a national project. Amongst the top 10 visitor countries, the USA and China come in second and third place, respectively, after Germany but ahead of other European countries such as the UK or Switzerland. Besides the DaNa2.0 website, the expert team uses additional channels to communicate the collected knowledge on nanotechnology-related issues to interested laymen. Members of the DaNa2.0 team participate regularly in group discussions, public dialogue events, conferences (e.g. SETAC) and fairs, and interact with journalists (interviews, commentaries, etc.). Beyond that, the annual cluster meetings, the place where partners of the



BMBF-funded projects are given the opportunity to exchange and interact, are also partly open to the public.

#### Conclusions

The DaNa2.0 website offers easy access to complex scientific issues related to the environmental impact of nanomaterials. The information provided is constantly extended, updated and adjusted to the latest developments in the field of nanotechnology. Taking into account the rapid development in the field of nanotechnology, keeping all the content of www.nanoobjects.info constantly updated is a challenge. Hence, the involvement of European environmental experts in the DaNa2.0 consortium will not only provide further input by, e.g. including further issues with environmental relevance, such as risk assessment and realistic environmental exposure, but will also facilitate the inclusion of latest research results. Likewise, the 'entry port' for scientific facts to be integrated into the knowledge base, the Literature Criteria Checklist, is currently being critically revised. With regard to ecotoxicity testing, specific criteria to, e.g. evaluate the application of nanomaterials to the test media (e.g. soil) and also concerning analytic methods have to be developed and included.

The DaNa2.0 consortium will further strengthen the cooperation and exchange with other dissemination organisations (databases, websites, projects) on the national and international level. Examples are the associations to EU projects such as eNanoMapper (www.enanomapper.net), NanoValid (www.nanovalid.eu) and the InterRegIVbfunded project NANORA (www.nanora.eu), the latter is initiating a French version of the knowledge base. International contributions include also the dialogue with the European NanoSafetyCluster and contributions to an international activity concerning curation of nanotechnology data coordinated by the US NCIP Nanotechnology Working Group [18]. Additionally, members of the DaNa2.0 team participate in several public dialogues or discussions on nanotechnology and nano-toxicology in order to inform the public and raise awareness for their activities. The team is also contributing to other international activities such as the OECD WPMN activities [19]. Citations of the DaNa2.0 website as single national database in the European Commission's 'Commission Staff Working Paper' [20], in 'The JRC Web Platform on Nanomaterials' [21] and in the commentary 'Focusing the research efforts' [22] clearly demonstrate the importance and quality of the provided data, respectively. Finally, the comprehensive collection of environmental relevant facts on nanomaterials will assist in defining knowledge gaps and further research needs.

#### Methods

This article describes how information of nanotechnology and its potential adverse effects are collected, evaluated and edited in order to provide generally understandable facts and information on a web-based platform. A more detailed description of the methodology applied can be retrieved from the paragraphs introducing the single sections of the website.

#### Abbreviations

BMBF: German Federal Ministry of Education and Research; DaNa2.0: Data and knowledge on nanomaterials - evaluation of socially relevant scientific facts; ENM: engineered nanomaterial; JRC: Joint Research Centre; NM: nanomaterials; OECD: Organisation for Economic Co-operation and Development; WPMN: Working Party on Manufactured Nanomaterials; SETAC: Society of Environmental Toxicology and Chemistry; SOP: standard operating procedures.

#### **Competing interests**

The authors of this article form the core team of DaNa2.0. The core team and the external experts are funded by the German Federal Ministry for Education and Research (BMBF) under grant no. 03X0131. They are also the authors of the content published on the project website www.nanoobjects.info.

#### Authors' contributions

DK and CM drafted and revised the manuscript. KN included information regarding the website and access statistics. BM and CS provided input regarding materials science and cooperation and synergisms to other projects. HK and CS included information on cooperation and synergisms to other projects. All authors read and approved the final manuscript.

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