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SINTEF REPORT

TITLE

**Nanotechnology applications in Fisheries and Aquaculture
- Global and Norwegian Opportunities and Challenges -
- A Pre-project -**

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ABSTRACT

This pre-project was dealing with the study of the possibilities to apply Nanotechnology in the different aspects of Fisheries and Aquaculture at Norwegian and Global level taking also into account the positive and the potential negative aspects to introduce this new technology.

The study was carried out mainly based on literature studies, opinions of relevant scientists, participating in Nanotechnology conference and also by preparing an application of use of nanotechnology in fish feed. This report has reviewed also relevant literature regarding the use of Nanotechnology in food systems, where fish belong; however no information of direct application of Nanotechnology has been found. This work has included a section of health effects of Nanotechnology on humans, environment and animals especially in marine species like crustacean and fish.

KEYWORDS	ENGLISH	NORWEGIAN
GROUP 1	Nanotechnology	Nanoteknologi
GROUP 2		
SELECTED BY AUTHOR		

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1. INTRODUCTION

In the literature we find many definitions of Nanotechnology:

- “Nanoscience and Nanotechnology is the understanding and applying atomic or molecular level manipulation and assembly of structures”
- Nanotechnology focuses on the physical/biological structures smaller than 100 nm. These structures can be manipulated and converted into nanomachines able to achieve functions previously not possible
- Nanotechnology involves research and technology development at the 1 nm-to-100nm range. Also creates and uses structures that have novel properties because of their small size and builds on the ability to control or manipulate at the atomic scale

Nanotechnology is an interdisciplinary science: “ *Chemists, physicists and medical doctors are working alongside engineers, biologists, and computer scientists to determine the applications, direction and development of Nanotechnology*”

There are two approaches in Nanotechnology: the “bottom-up approach” which arrange smaller components into more complex assemblies and the “top down approach” which builds something by starting with a larger piece and carving away material (like a sculpture).

2. MATERIALS AND METHODS

The research in this project was based on interviewing scientists working with nanotechnology and literature data.

3. RESULTS

3.1 *Nanotechnology in Fisheries and Aquaculture*

This pre project aimed to identify some subjects where SINTEF Fisheries and Aquaculture could work in a near future.

The identified areas are:

- Fish feed, based on a previous concept developed by SINTEF : “Intel Feed”
- Improvement of packing, packing materials, quality and safety for Seafood
- A better utilization and Improvement of Protein and Lipid quality of fish products, including by-products
- Nano-Net is future netting for aquaculture, free from escapes, environmentally friendly and easy to deal with
- The Ghost Fishing problem
- Antibacterial surfaces in aquaculture

The fish feed subject was studied taking as a starting point the developments which has been done by SINTEF Fisheries and Aquaculture (SFH) and SINTEF Material and Chemistry (SMC), consisting on an “intelligent” fish feed where the behavior of the physical/chemical and

nutritional components can be manipulated according to the environment where the feed will act (e.g. water or fish itself).

With this we could prepare a feed with following characteristics:

- Leakage control of ingredients to the water environment
- Release control of ingredients at specific organs of the fish as intestine, stomach, etc.

INTELFEEED concept can be used for:

- Fish larvae feed
- On growing feed
- Medicinal feed

SFH and SMC decided to apply for a project to the Norwegian Research Council based on the application of Nanotechnology to produce a fish feed for salmon, where the release of the antinutrients compounds from the Soya meal are selectively controlled in the gut of the Salmon. This project was finally rejected by the Norwegian Research Council

The second subject we studied was the improvement of packing, packing materials, quality and safety for Seafood. This subject has already been studied by SINTEF Chemistry and Materials and an EU application was submitted already 2 years ago. The application was rejected, however I think this subject will continue to develop and it is especially important for Norway to invest in knowledge for packing nanotechnology for seafood.

The third subject which was to investigate a better utilization and improvement of protein and lipid quality of fish products, including by-products was not possible to be studied because of lack of funds for to make a proper studied.

The fourth subject: Nano-net is already being studied by SINTEF Material and Chemistry and SINTEF Fisheries and Aquaculture but the results still has not been fully published.

The fifth subject about on the biodegradable fishing nets, was taken by the Fisheries division of SINTEF Fisheries and Aquaculture to be followed up, resulting in an application that was sent to the fishery and aquaculture industry research fund with a negative outcome.

The last subject “antibacterial surfaces in aquaculture” was as the third subject was not studied due to the lack of funds.

In conclusion it was decided to use the time in this pre project to study the possibility of introducing a fish feed with special properties of controlled release of antinutrients contained in the soya meal.

The funds of this pre project were also used to study the status of Nanotechnology in food system.

3.2 Nanotechnology in Food Systems

Most of the nanotechnological research focuses on the development of applications in biosciences and engineering. In the food sciences, nanotechnology applications are still limited and the strategies to apply nanoscience to the food industry are quite different from these more traditional applications of nanotechnology. Food processing is a multi-technological, manufacturing industry involving a wide variety of raw materials, high bio-safety requirements, and well-regulated technological processes [1]

Nanotechnology has the potential to impact many aspects of food and agriculture systems. Food security, disease- treatment delivery method, new tools for molecular and cellular biology, new materials for pathogen detection and protection of the environment are examples of the important links of Nanotechnology to the science and engineering of agriculture and food systems. Examples of nanotechnology as a tool for achieving further advances in the food industry are as follows:

- Increased security of manufacturing, processing, and shipping of food products through sensors for pathogen and contaminant detection.
- Devices to maintain historical environmental records of a particular product and tracking of individual shipments.
- Systems that provide integration of sensing, localization reporting, and remote control of food products (smart/intelligent systems) and that can increase efficacy and security of food processing and transportation.
- Encapsulation and delivery systems that carry, protect, and deliver functional food ingredients to their specific site of action.

Most nanotechnological research focuses on the development of applications in biosciences and engineering. Strategies to apply nanoscience to the food industry are quite different from the more traditional applications of nanotechnology. Food processing is a multitechnological manufacturing industry involving a wide variety of raw materials, high bio safety requirements and well regulated technological processes. Four major areas in food production may benefit from nanotechnology; development of new functional materials, micro scale and nanoscale processing product development, methods and instrumentation design for improved food safety, and bio security. Figure 1 shows possible applications of Nanotechnology in the food industry.

Nanotechnology research in the food industry is getting focused on food packaging. With the increasing global customer base, food retailing is transforming. However, with the move toward globalization, food packaging requires longer shelf life, along with monitoring food safety and quality based upon international standards. With a different nanostructure, the gas and water vapour permeability of plastics can be engineered to preserve fruit, vegetables, beverages, etc. With the use of nanoparticles, bottles and packaging can be made lighter and stronger with better thermal performance and less gas absorption. These properties can extend the shelf life of products, as well as reduce transportation costs involved in shipping food.

Nanotechnology has the capability to transform the nature of food packaging materials in future. Certain nanoscale innovations could bring amazing improvements to food packaging in the forms of detection of pathogens, smart and active packaging, and barrier and mechanical properties. Packaging that incorporates nano materials can be “smart,” which means that it can respond to environmental conditions or repair itself or alert a consumer to contamination and/or the presence of pathogens. It offers several extraordinary benefits to improve food packages like advancements in fundamental characteristics of food packaging materials such as antimicrobial properties, barrier properties, strength, and stability to heat and cold.

In the United States, Japan, and Australia, active packaging is already being successfully applied to extend shelf-life while maintaining nutritional quality and ensuring microbiological safety. Examples of commercial applications include the use of oxygen scavengers for sliced processed meat, ready-to-eat meals and beer, the use of moisture absorbers for fresh meat, poultry and fresh fish, and ethylene-scavenging bags for packaging of fruit and vegetables. In Europe, however,

only a few of these systems have been developed and are being applied now. The main reasons for this are legislative restrictions and a lack of knowledge about acceptability to European consumers, as well as the efficacy of such systems and the economic and environmental impact such systems may have. The European "Actipak" (put reference) project will address these issues in the near future.

Nanoclays have shown the broadest commercial viability due to their lower cost and their utility in common thermoplastics like polypropylene (PP), thermoplastic polyolefin (TPO), PET, polyethylene (PE), polystyrene (PS), and nylon.

The rapid use of nano-based packaging in a wide range of consumer products has also raised a number of safety, environmental, ethical, policy and regulatory issues. The main concerns stem from the lack of knowledge with regard to the interactions of nano-sized materials at the molecular or physiological levels and their potential effects and impacts on consumers health and the environment. Research and development in the field of active and intelligent packaging materials is very dynamic and develops in step with the search for environmentally friendly packaging solutions. In this context, the design of tailor-made packaging is a real challenge, and it implies the use of reverse engineering approaches based on food requirements and not just on the availability of packaging materials any longer. Nanotechnologies are expected to play a major role, taking into account all additional safety considerations and filling present packaging needs.

Nanotechnology is also used as a deliver technology for functional ingredients as drugs, vitamins, antimicrobials, antioxidant, flavorings, etc. These functional ingredients come in a variety of different molecular and physical forms such as polarities, molecular weights and physical states.

A delivery system must perform a number of different roles:

1. As a vehicle for carrying the functional ingredient to the desired site of action.
2. As protection of the functional ingredient from chemical or biological degradation during processing, storage, and utilization;
3. It may have to be capable of controlling the release of the functional ingredient;
4. It has to be compatible with the other components in the system, as well as being compatible with the physicochemical and qualitative attributes (that is, appearance, texture, taste, and shelf-life) of the final product.

The characteristics of delivery systems are one of the most important factors influencing the efficacy of functional ingredients in many industrial products. A wide variety of delivery systems has been developed to encapsulate functional ingredients, including simple solutions, association colloids, emulsions, biopolymer matrices, etc. Each type of delivery system has its own specific advantages and disadvantages for encapsulation, protection, and delivery of functional ingredients, as well as cost, regulatory status, ease of use, biodegradability, and biocompatibility.

Challenges of Nanotechnology in Food Sciences a member of NSTDA

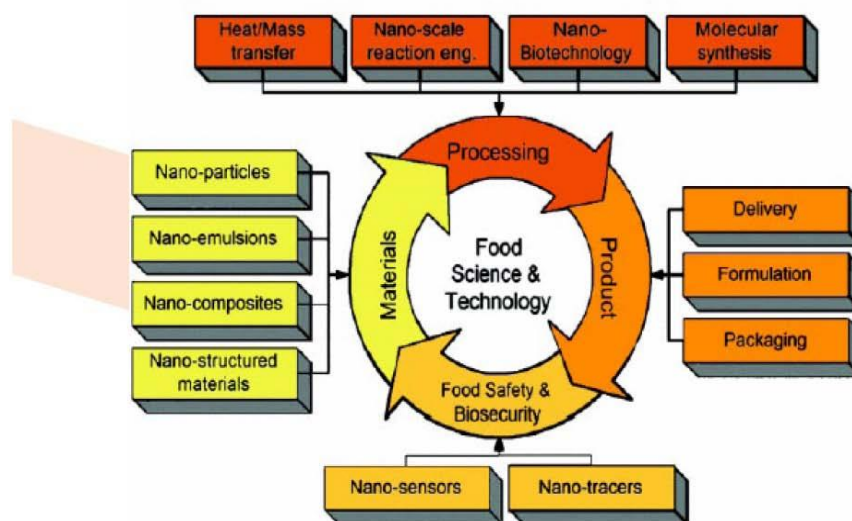


Fig. 1—Application matrix of nanotechnology in food science and technology.

A Driving Force for National Science and Technology Capability

Source : Wiwut Tanthapanichakoon, Ph.D National Nanotechnology Center (NANOTEC)

Some delivery systems are:

- Association colloids (surfactant, micelles, bilayers, reverse micelles etc)
- Nanoemulsions (droplets less than 100 to 500 nm)
- Nanostructured multiple emulsions (O/W/O, W/O/W)
- Biopolymeric Nanoparticles (use of food grade biopolymers)
- Nanolaminates (2 or more layers of material with nanometer dimensions that are physically or chemically bonded to each other)
- Solid lipid nanoparticles (Liquid lipid in emulsion liquid; emulsion lipid (oil) is replaced by high melting point lipid ; size 50 – 500 nm)
- Nanostructured multilayer emulsions

3.2.1 Report from the IFT's fourth International Food Nanoscience Conference (June 2009)

The conference was held on June 6 in Anaheim, USA and had as title: “Nanoscale Science and Technology of Food” and was organized in conjunction with the IFT’s fourth International Food Nanoscience Conference and the Annual Meeting & Food Expo.

One conference session focused on delivery systems for nutrients and functional compounds in foods. The systems discussed were those adopted mainly from the pharmaceutical industry. They were reported to effectively deliver compounds that are susceptible to degradation processes, such as chemical, biochemical and microbiological.

Professor Jochen Weiss from Germany discussed edible solid lipid nanoparticles (SLNs) for delivery bioactives such as lycopene and carotenoids. SLNs are formed by controlled crystallization of food nanoemulsions and range in size between 50 and 500 nm. Size is influenced by nucleation and crystal growth processes, which are determined by the initial drop size and the composition. Of the interfacial membrane that stabilizes the droplet. The major advantages of SLNs include large scale production without the use of organic solvents, high concentration of functional compounds in the system, long term stability, and the ability to be spray dried into powder form.

Challenges to production include dispersion of the functional compound within the lipid and polymorphic transformation due to loss of fluidity.

In another session Assistant Professor Cristina Sabliov from USA focused on polymeric nanoparticles for controlled release and targeted delivery of functional compounds. They are made using polymers and surfactants.

Edible polymer types, including alginic acid poly(lactic co-glycolic acid) and chitosan are used in her laboratory. Sabliov presented data on polymeric nanoparticles, including vitamin E, itraconazole (antimicrobial) and β -carotene as colorant. Synthesis methodologies composition and particles properties such as size and morphology were described for each nanoparticle. Further research is needed to establish optimum release conditions and body uptake in the case of vitamin E.

In a different approach to Nanotechnology on food Professor Jose Miguel Aguilera, outlined the natural and unintentional sources of nanotechnology in food. He cited examples of natural events that may result in formation of nanoscale materials or employ nanoscience principles. Examples included digestion of food and food structure building processes such as arrangement of amylose and amylopectin in a starch granule.

Concluding, Aguilera reiterated the importance of the current knowledge and tools derived from nanoscience in understanding the role of nanotechnology in food.

Three technologies were highlighted in a session dedicated to current commercial and near commercial applications. They included the *NutraLease* technology (reference) for delivery of additives and bioactive compounds for doff fortification, and antimicrobial coatings for food packing applications.

NutraLease consists of nanosized, self assembled liquid vehicles (NSSL) for delivery of nutrients and bioactives. NSSLs are novel, nano-based emulsions that are highly soluble and fully dilutable in aqueous phase. Vehicles for several compounds such as omega 3 fatty acids, carotenoids (lycopene) lutein, astaxanthin, Co Q10, phytosterols, and isoflavones have been developed and patented. They have been used successfully in clear beverages without phase separation or cloudiness.

Florentine Hilty from Switzerland indicated that nanonizing iron compounds help alleviate their bioavailability as observed through in vitro and animal studies. Additionally, the nanostructured compounds minimize undesirable quality changes in the final product. Size reduction is achieved using flame pyrolysis.

Bill Norwood, President of the nanoAgriSystems Inc., (website maybe) discussed the company's process that uses nanotechnology and chemistry to develop a system for production of antimicrobial coatings for packaging. The technology is based on an open-air flame technique for coatings packages and is targeted to the fruit and vegetable industry. It consists of a machine coupled to a specialized device known as nanomiser, which is used to convert antimicrobial solutions into fine (nanosize) droplets. The droplets are combusted by the flame into a vapour that coats the package. Several bacteria species has showed delayed growth when this technology has been tested. Commercialization of this technology is pending approval by regulatory agencies. In this conference was no mentioned any work dealing with developing of fish/marine technology. The Nanotechnology regulation was discussed by Annette Mc Carthy from the FDA. She assured the audience of the FDA's sufficient authority in the assessing the safety of nanomaterials (see more below at the following section).

3.2.2 Regulations: position of FDA and EFSA about Nanotechnology regarding food safety

The position of the food authorities on use of Nanotechnology in food systems can differ according to the region. For example, in the United States of America it is the Food and Drug Administration (FDA) (website reference) the institution which is responsible to dictate the rules for the use of Nanotechnology in food systems. In Europe is the European Safety Food Authority (EFSA) (website reference)?.

At the present the FDA does not currently have a specific definition for Nanotechnology and regulatory assessments are based not only on material size but also on their impact on identity and toxicity.

The Industry has requested from FDA to prepare a guide that clarifies when nanoscale versions of approved food additives are covered by the existing regulations. FDA is in the process of drafting a guidance document that will address all manufacturing changes including nanotechnology. The FDA hopes to have a draft ready for publication by late 2009 (but it may not be ready until during the year 2010).

The position of European Food Safety Authority (EFSA) is based in a scientific opinion adopted on 10 February 2009 (Scientific Opinion of the Scientific Committee, question No EFSA-Q-2007-124a), which deals with the potential risks arising from nanoscience and nanotechnologies on food and feed safety. The Scientific Committee makes a series of recommendations; in particular, actions should be taken to develop methods to detect and measure ENMs in food/feed and biological tissues, to survey the use of engineered nanomaterials (ENMs) in the food/feed area, to assess the exposure in consumers and livestock, and to generate information on the toxicity of different ENMs

The EFSA has made a report (issued on March 9, 2009) based on consultation to the public. At the deadline EFSA had received 208 submissions, from about 30 interested parties (individuals, non-governmental organisations, industry organisations, academia and national assessment bodies).

All comments received were scrutinized and subsequently 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

At the present the legislation has not yet been completed.

3.3 Nanotechnology and health

The increasing interest in applying Nanotechnology in different areas of industrial development has lead to a parallel interest in study the environmental impacts of nanotechnology.

Until recently the potential negative impacts of nanomaterials on human health and the environment have been rather speculative and unsubstantiated [2].

However, within the past number of years several studies have indicated that exposure to specific nanomaterials, e.g. nanoparticles, can lead to several adverse effects in humans and animals [3].

This has made some people very concerned, drawing specific parallels to past negative experiences with small particles [5].

Some types of nanoparticles are expected to be benign and are FDA approved and used for making paints and sunscreen lotion etc. However, there are also dangerous nanosized particles and chemicals that are known to accumulate in the food chain and have been known for many years like asbestos, lead, ultra fine particles, and others.

The extrapolation of experiences with bulk materials to nanoparticles is very difficult because their chemical properties can be quite different. For instance, anti-bacterial silver nanoparticles dissolve in acids that would not dissolve bulk silver, which indicates their increased reactivity [6].

At present the global production of nanomaterials is hard to estimate for many reasons. Not even the definition of when something is nanotechnology is applied. Likewise, nanomaterials are used in a great diversity of products and industries and also there is a general lack of information about what and how much is being produced and by whom.

In 2001, the future global annual production of carbon-based nanomaterials was estimated to be several hundred tons, but already in 2003 the global production of nanotubes alone was estimated to be around 900 tons distributed between 16 manufacturers [7].

The Japanese company, Frontier Carbon Corp, plan to start an annual production of 40 tons of C₆₀ (fullerene)[8].

It is estimated that the global annual production of nanotubes and fiber was 65 tons equal to €144 million worth and it is expected to surpass €3 billion by 2010 representing an annual growth rate of well over 60% [9].

Even though the information about the production of carbon-based nanomaterials is scarce, the annual production volumes of for instance quantum dots, nano-metals, and materials with nanostructured surfaces are completely unknown.

The development of nanotechnology is still in its infancy, and the current production and use of nanomaterials is most likely not representative for the future use and production. Some estimates for the future manufacturing of nanomaterials have been made. For instance the Royal Society and the Royal Academy of Engineering [10].

3.3.1 Exposure of Nanomaterials to environment and to humans

Exposure of nanomaterials to workers, consumers, and the environment seems inevitable with the increasing production volumes and the increasing number of commercially available products containing nanomaterials or based on nanotechnology [11].

Exposure is a key element in risk assessment of nanomaterials since it is a precondition for the potential toxicological and ecotoxicological effects to take place. If there is no exposure – there is no risk. Nanoparticles are already being used in various products and the exposure can happen through multiple routes.

Human routes of exposure are:

- dermal (for instance through the use of cosmetics containing nanoparticles);
- inhalation (of nanoparticles for instance in the workplace);
- ingestion (of for instance food products containing nanoparticles);
- and injection (of for instance medicine based on nanotechnology).

Although there are many different kinds of nanomaterials, concerns have mainly been raised about free nanoparticles

Free nanoparticles could either get into the environment through direct outlet to the environment or through the degradation of nanomaterials (such as surface bound nanoparticles or nanosized coatings).

Environmental routes of exposure are multiple. One route is via the wastewater system. At the moment research laboratories and manufacturing companies must be assumed to be the main contributor of carbon-based nanoparticles to the wastewater outlet.

For other kinds of nanoparticles for instance titanium dioxide and silver, consumer products such as cosmetics, crèmes and detergents, is a key source already and discharges must be assumed to increase with the development of nanotechnology.

However, as development and applications of these materials increases this exposure pattern must be assumed to change dramatically. Traces of drugs and medicine based on nanoparticles can also be disposed of through the wastewater system into the environment.

Drugs are often coated, and studies have show that these coatings can be degraded through either metabolism inside the human body or transformation in environment due to UV-light [12]. Which only emphasizes the need to studying the many possible processes that will alter the properties of nanoparticles once they are released in nature?

Another route of exposure into the environment is from wastewater overflow or if there is an outlet from the wastewater treatment plant where nanoparticles are not effectively held back or degraded.

Additional routes of environmental exposure are spills from production, transport, and disposal of nanomaterials or products [13].

While many of the potential routes of exposure are uncertain scenarios, which need confirmation, the direct application of nanoparticles, such as for instance nano zero valent iron for remediation of polluted areas or groundwater is one route of exposure that will certainly lead to environmental exposure. Although, remediation with the help of free nanoparticles is one of the most promising environmental nanotechnologies, it might also be one the one raising the most concerns. The Royal Society and The Royal Academy of Engineering are of the opinion that it is only a question of time before we will find nanomaterials such as nanoparticles in the environment – if we have the means to detect them.

The size of nanoparticles and our current lack of metrological methods to detect them is a huge potential problem in relation to identification and remediation both in relation to their fate in the human body and in the environment

Once there is a widespread environmental exposure human exposure through the environment seems almost inevitable since water- and sediment living organisms can take up nanoparticles

from water or by ingestion of nanoparticles sorbed to the vegetation or sediment and thereby making transport of nanoparticles up through the food chain possible

The term “nano (eco-) toxicology” has been developed on the request of a number of scientists and is now seen as a separate scientific discipline with the purpose of generating data and knowledge about nanomaterials effects on humans and the environment [14].

Toxicological information and data on nanomaterials is limited and ecotoxicological data is even more limited. Some toxicological studies have been done on biological systems with nanoparticles in the form of metals, metal oxides, selenium and carbon; however the majority of toxicological studies have been done with carbon fullerenes [15].

Only a very limited number of ecotoxicological studies have been performed on the effects of nanoparticles on environmentally relevant species, and, as for the toxicological studies, most of the studies have been done on fullerenes. However, according to the European Scientific Committee on Emerging and Newly Identified Health Risks [16], results from human toxicological studies on the cellular level can be assumed to be applicable for organisms in the environment, even though this of course needs further verification. In the following, a summary of the early findings from studies done on bacteria, crustaceans, fish, and plants will be given and discussed.

3.3.2 Effects of nanoparticles on bacteria

The effect of nanoparticles on bacteria is very important since bacteria constitute the lowest level and hence the entrance to the food chain in many ecosystems,

The effects of C₆₀ aggregates on two common soil bacteria *E. coli* (gram negative) and *B. subtilis* (gram positive) was investigated by Fortner et al. [17] on rich and minimal media, respectively, under aerobic and anaerobic conditions. At concentrations above 0.4 mg/L growth was completely inhibited in both cultures exposed with and without oxygen and light. No inhibition was observed on rich media in concentration up to 2.5 mg/L, which could be due to that C₆₀ precipitates or gets coated by proteins in the media. The importance of surface chemistry is highlighted by the observation that hydroxylated C₆₀ did not give any response, which is in agreement with the results obtained by Sayes et al. [18] who investigated the toxicity on human dermal- and liver cells. The antibacterial effects of C₆₀ has furthermore been observed by Oberdorster [19], who observed remarkably clearer water during experiments with fish in the aquarium with 0.5 mg/L compared to control.

Silver nanoparticles are increasingly used as antibacterial agent.

3.3.3 Effects of nanoparticles on Crustacean

A number of studies have been performed with the freshwater crustacean *Daphnia magna*, which is an important ecological important species that furthermore is the most commonly used organisms in regulatory testing of chemicals.

The organism can filter up to 16 ml an hour, which entails contact with large amounts of water in its surroundings. Nanoparticles can be taken up via the filtration and hence could lead to potential toxic effects [20].

Lovern and Klaper [21] observed some mortality after 48 hours of exposure to 35 mg/L C₆₀ (produced by stirring and also known as “nanoC₆₀” or “nC₆₀”), however 50% mortality was not achieved, and hence an LC50 could not be determined [22].

A considerable higher toxicity of $LC_{50} = 0.8 \text{ mg/L}$ is obtained when using nC_{60} put into solution via the solvent tetrahydrofuran (THF) – which might indicate that residues of THF is bound to or within the C_{60} -aggregates, however whether this is the case is unclear at the moment. The solubility of C_{60} using sonication has also been found to increase toxicity[23], whereas unfiltered C_{60} dissolved by sonication has been found to cause less toxicity ($LC_{50} = 8 \text{ mg/L}$). This is attributed to the formation of aggregates, which causes a variation of the bioavailability to the different concentrations. Besides mortality, deviating behavior was observed in the exposed *Daphnia magna* in the form of repeated collisions with the glass beakers and swimming in circles at the surface of the water[24]. Changes in the number of hops, heart rate, and appendage movement after subtoxic levels of exposure to C_{60} and other C_{60} -derivatives[25]. However, Titanium dioxide (TiO_2) dissolved via THF has been observed to cause increased mortality in *Daphnia magna* within 48 hours ($LC_{50} = 5.5 \text{ mg/L}$), but to a lesser extent than fullerenes, while unfiltered TiO_2 dissolved by sonication did not result in an increasing dose-response relationship, but rather in a variation response. Lovorn and Klaper have furthermore investigated whether THF contributed to the toxicity by comparing TiO_2 manufactured with and without THF and found no difference in toxicity and hence concluded that THF did not contribute to neither the toxicity of TiO_2 or fullerenes.

Experiments with the marine species *Acartia tonsa* exposed to 22.5 mg/L stirred nC_{60} have been found to cause up to 23% mortality after 96 hours, however mortality was not significantly different from control[25]. And exposure of *Hyella azteca* by 7 mg/L stirred nC_{60} in 96 hours did not lead to any visible toxic effects – not even by administration of C_{60} through the feed[26].

Only a limited number of studies have investigated long-term exposure of nanoparticles to crustaceans. Chronic exposure of *Daphnia magna* with 2.5 mg/L stirred nC_{60} was observed to cause 40% mortality besides causing sub-lethal effects in the form of reduced reproducibility (fewer offspring) and delayed shift of shield [27].

Templeton et al.[28] observed an average cumulative life-cycle mortality of $13 \pm 4\%$ in an Estuarine Meiobenthic Copepod *Amphiascus tenuiremis* after being exposed to single walled carbon nanotubes (SWCNT), while mean life-cycle mortalities of 12 ± 3 , 19 ± 2 , 21 ± 3 , and $36 \pm 11\%$ were observed for 0.58 , 0.97 , 1.6 , and 10 mg/L .

Exposure to 10 mg/L showed:

1. significantly increased mortalities for the naupliar stage and cumulative life-cycle;
2. a dramatically reduced development success to 51% for the nauplius to copepodite window, 89% for the copepodite to adult window, and 34% overall for the nauplius to adult period;
3. a significantly depressed fertilization rate averaging only $64 \pm 13\%$.

Templeton also observed that exposure to 1.6 mg/L caused a significant increase in development rate of 1 day faster, whereas a 6 day significant delay was seen for 10 mg/L .

3.3.4 Effects of nanoparticles on Fish

A limited number of studies have been done with fish as test species. In a highly cited study, Oberdorster [29] found that 0.5 mg/L C_{60} dissolved in THF(?) caused increased lipid peroxidation in the brain of largemouth bass (*Mikropterus salmoides*). Lipid peroxidation was found to be decreased in the gills and the liver, which was attributed to repair enzymes. No protein oxidation was observed in any of the mentioned tissues, however a discharge of the antioxidant

glutathione occurred in the liver possibility due to large amount of reactive oxygen molecules stemming from oxidative stress caused by C₆₀ [30].

For *Pimephales promelas* exposed to 1 mg/L THF-dissolved C₆₀, 100 % mortality was obtained within 18 hours, whereas 1 mg/L C₆₀ stirred in water did not lead to any mortality within 96 hours. However, at this concentration inhibition of a gene which regulates fat metabolism was observed. No effect was observed in the species *Oryzia latipes* at 1 mg/L stirred C₆₀, which indicates different inter-species sensitivity toward C₆₀ [31]

Smith et al. observed a dose-dependent rise in ventilation rate, gill pathologies (oedema, altered mucocytes, hyperplasia), and mucus secretion with SWCNT precipitation on the gill mucus in juvenile rainbow trout.

Smith et al. also observed:

- dose-dependent changes in brain and gill Zn or Cu, partly attributed to the solvent;
- significant increases in Na+K+ATPase activity in the gills and intestine;
- significant dose-dependent decreases in TBARS especially in the gill, brain and liver;
- and significant increases in the total glutathione levels in the gills (28 %) and livers (18 %), compared to the solvent control (15 mg/l SDS).
- Finally, they observed increasing aggressive behavior; possible aneurisms or swellings on the ventral surface of the cerebellum in the brain and apoptotic bodies and cells in abnormal nuclear division in liver cells.

Recently Kashiwada [32] reported observing 35.6% lethal effect in embryos of the medaka *Oryzias latipes* (ST II strain) exposed to 39.4 nm polystyrene nanoparticles at 30 mg/L, but no mortality was observed during the exposure and postexposure to hatch periods at exposure to 1 mg/L. The lethal effect was observed to increase proportionally with the salinity, and 100% complete lethality occurred at 5 time higher concentrated embryo rearing medium. Kashwada also found that 474 nm particles showed the highest bioavailability to eggs, and 39.4 nm particles were confirmed to shift into the yolk and gallbladder along with embryonic development. High levels of particles were found in the gills and intestine for adult medaka exposed to 39.4 nm nanoparticles at 10 mg/L, and it is hypothesized that particles pass through the membranes of the gills and/or intestine and enter the circulation.

3.3.5 Effects of Nanoparticles on Plants

To our knowledge only one study has been performed on phytotoxicity, and it indicates that aluminum nanoparticles become less toxic when coated with phenatrene, which again underlines the importance to surface treatments in relation to the toxicity of nanoparticles[33].

4. DISCUSSION

Nanotechnology is an emerging area of science that has the potential to generate new products and processes and it is developing at fast speed. However, some insecurity still exist special when hazard effects on the environment, animals and humans are still not fully understood mainly because of lack of methodology to detect potential toxicity of the nanomaterials /nanoparticles which are used in this technology.

It is still a debate between those who claim that nanotechnology will have a positive impact on society and those who consider it dangerous. And this is more obvious when Nanotechnology is being applied to food systems.

In this report we have dealt with the application of nanotechnology in the fisheries and aquaculture field. At present the information of application in this field is scarce and only in the area of packaging some research has been done. SINTEF Fisheries and Aquaculture in cooperation with SINTEF Chemistry and Materials applied for a research project where nanotechnology is applied in fish feed technology but no funding was achieved. It seems that nanotechnology still has not reached the status of a real tool in fisheries and aquaculture.

This pre-project also performed a literature study on nanotechnology in food systems and found scarce or no information of nanotechnology and fish as a food system.

Our conclusion is that when the legislation of FDA from USA and EFSA from the EU is ready then the applications of nanotechnology on food systems will greatly increase. In general nanotechnology is developing around the world as we can see in figure 2 and the food “Nano-marked” will also increase dramatically in the coming years (figure 3).

Some projects have aimed to predict how will the world in 2040 and suggested that in: *“2040 will be the common use of nano produced food, which has the correct nutritional composition and the same taste and texture of organically produced food, means that the availability of food is no longer affected by limited resources, bad crop weather, water problems or others.”* (Figure 4)

Some other predictions about what will be the world with nanotechnology are:

- Nanotechnology will have an impact on the food industry, from how food is grown and produced, processed to how it is packaged, transported and consumed.
- Companies are developing nanomaterials that will make a difference not only in the taste of food, but also in food safety, and the health benefits food delivers.
- Nanotechnology will transform the entire food industry in the next 20 years.
- In agriculture, various applications will aim at reducing pesticide and water use, improving plant and animal breeding, and creating nano-bioindustrial products.

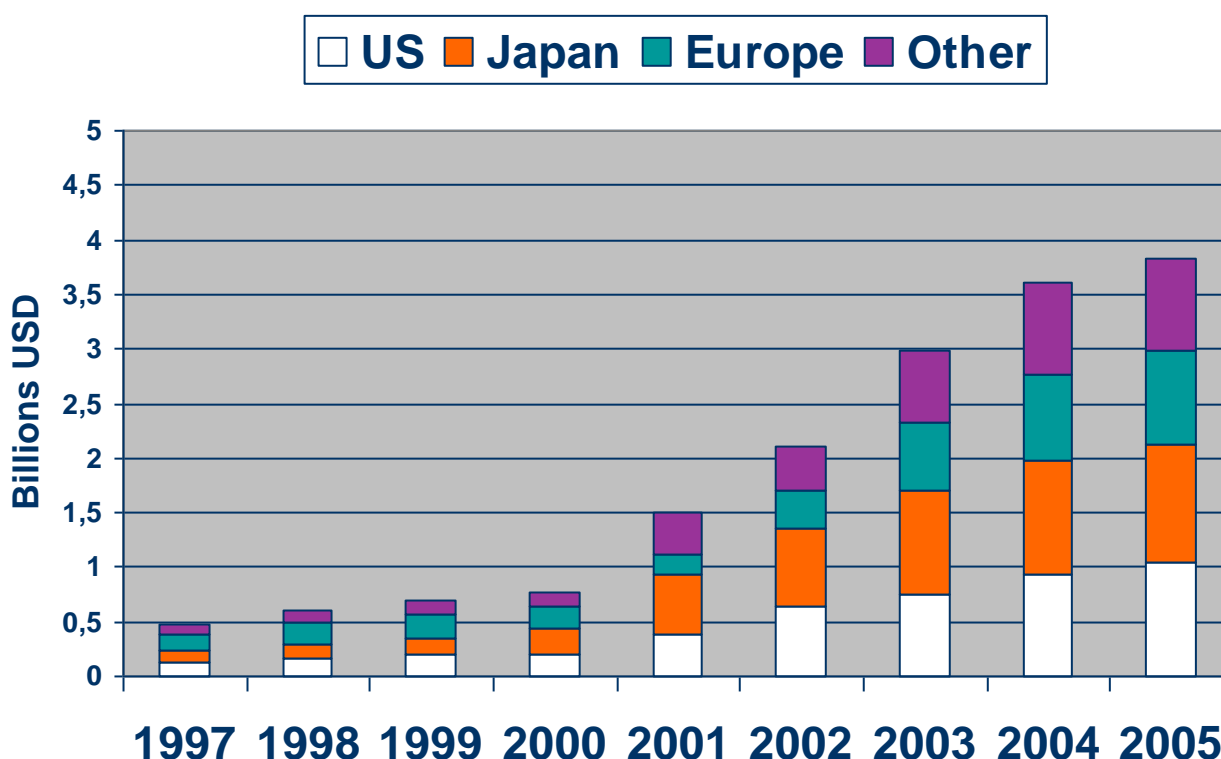


Figure 2. World spending in Nanotechnology

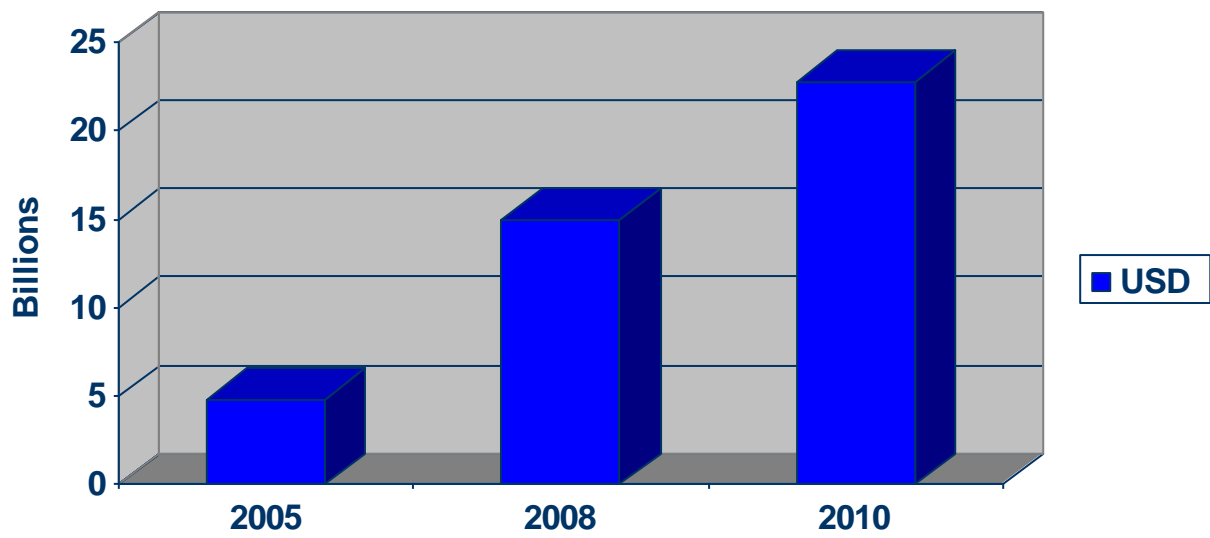


Figure 3. Food Nan marked Source: *Helmut Kaiser Consultancy, Germany*

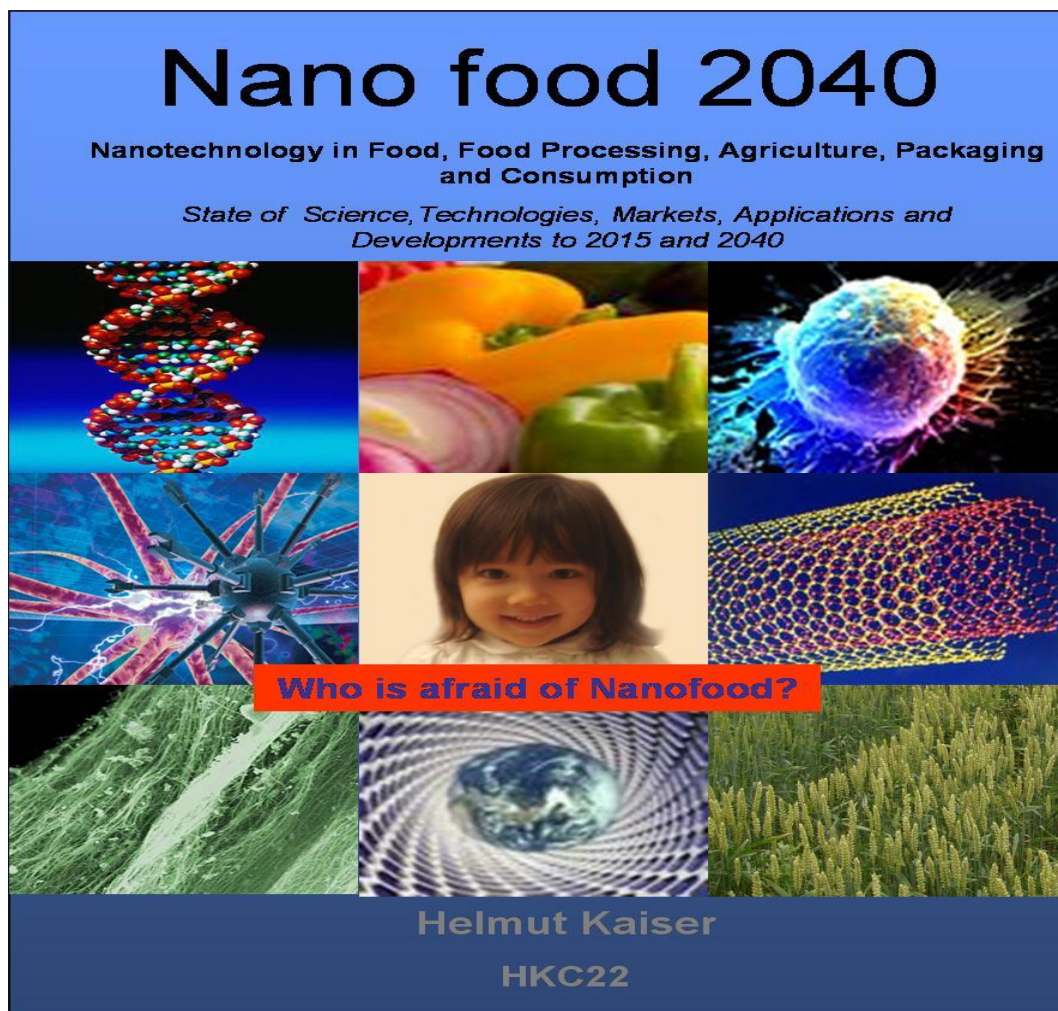


Figure 4. Nanotechnology in year 2040

As we mentioned above one of the greatest hinders for the developing of nanotechnology are the limitation of methods to detect the nanoparticles and to evaluate their toxicity to the environment and the all forms of life. At the present, however, research is aiming to find these methods which will give the security that is needed to develop nanotechnology at a higher speed.

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