HELLENIC REPUBLIC
National and Kapodistrian University of Athens
EST. 1837

Interdisciplinary M.Sc course in Nanomedicine

Academic year: 2018-2019

Thesis Title: *Current regulatory challenges in nanocosmetics, medical devices and food supplements*

Sofia-Paraskevi Makri, *Physicist*

Supervisor:
Dr. Konstantinos Gardikis, *Pharmacist, MSc, PhD, Scientific Affairs Director, APIVITA SA*

Co-Supervisors:
E. Efstathopoulos, *Professor, Medical Radiation Physics, Medical School, National & Kapodistrian University of Athens*
C. Demetzos, *Professor of Pharmaceutical Technology & Nano-Technology, Department of Pharmacy of the National & Kapodistrian University of Athens*

August 2019

Athens

Acknowledgements

I would like to express my gratitude to my supervisor Dr. Konstantinos Gardikis for the useful comments and remarks as well as for the opportunity to carry out this dissertation. Furthermore, I would like to thank the master's lecturers for the knowledge they offered us. Finally, I would like to thank my family and friends, who have supported me throughout the entire process.

Contents

Abbreviations

BCOP	Bovine Corneal Opacity and Permeability
CAS	Chemical Abstracts Service
CE	Conformité Européenne
CFDA	China Food and Drug Administration
CLP	Classification, Labelling and Packaging
CPNP	Cosmetic Products Notification Portal
CPSR	Cosmetic Product Safety Report
DSHEA	Dietary Supplement Health and Education Act
EBM	Engineered biomaterials
EC	European Commission
ECHA	European Chemical Agency
EFSA	European Food Safety Authority
EMA	European Medicines Agency
EU	European Union
FD&C Act	Federal Food, Drug and Cosmetic Act
FDA	Food and Drug Administration
GI	gastrointestinal
GMP	Good Manufacturing Practices
ICE	Isolated Chicken Eye
IECIC	Inventory of Existing Cosmetic Ingredients in China
ISO	International Organization for Standardization
IVD	in-vitro Diagnostic Medical Device
IVDR	in-vitro Diagnostic Medical Device Regulation
MD	Medical Device
MDR	Medical Device Regulation
NM	Nanomaterials
NMPA	National Medical Products Administration
NP	Nanoparticle
NSCNN	National Steering Committee for Nanoscience and Nanotechnology
PCPC	Personal Care Products Council
PIF	Product Information File
PLGA	poly(lactic-co-glycolic acid)
PMA	Premarket Approval Application
QD	Quantum Dots
REACH	Registration, Evaluation, Authorisation and Restriction of Chemicals
RP	Responsible Person

SCCS	Scientific Committee on Consumer Safety
SCENIHR	Scientific Committee on Emerging and Newly Identified Health Risks
SFDA	State Food and Drug Administration
US	United States
UV	Ultraviolent
VCRP	Voluntary Cosmetic Registration Program

Abstract

Nanotechnology is an emerging field capable of revolutionizing many different sectors, creating advanced products and systems that can be applied in a wide range of fields. Nanotechnology encompasses a wide range of sciences such as Physics, Chemistry, Biology, Engineering, Medicine and Informatics. It has been proved that nanoparticles (NPs) exhibit superior physical, chemical and biological properties as well as enhanced performance in comparison with their bulk counterparts. Nowadays, nano-enabled products are rapidly produced with variety of industrial applications and are applied in real life scenarios. However, alongside with the advantages and the excitement over the prospects of nanotechnology enabled materials, there have been increasing concerns regarding the risks and toxicity this field may present. On account of this, it is deemed crucial to establish a regulatory framework in order to protect human and environmental safety. However, to date, regulatory agencies have not yet defined a universal definition of nanomaterials while each region uses its own definitions and legislation to monitor nano-enabled products. Hence, the aim of this study is to present and discuss the existing regulatory framework for various products containing nanoparticles; in particular cosmetics, medical devices and food supplements in three regions, EU, US and China. This study is also outlining the type of NPs that are used in these three types of products as well as the benefits that they offer. Finally, this work is approaching the toxicity issues due to NPs.

1. Introduction

Over the past few decades, nanotechnology has increasingly been considered a promising technology that has revolutionized several sectors of industry.[1] Nanotechnology is a multidisciplinary scientific field that deals with the development and use of materials with dimensions in nanoscale that can be involved in a wide variety of systems (devices and products). In science and technology, the word ''nano'' comes from greek language and means very small. One nanometer equals one billionth of a meter.[2] A general definition of nanotechnology mentions that nanotechnology is the science, engineering, and technology conducted at the nanoscale, which is about 1 to 100 nanometers. In this scale, the properties of matter may differ significantly (i.e., quantum-scale effects play an important role) from that at larger scales.[3] Generally, nanomaterials are classified according to dimensionality in four classes: i) **0-D** when they have all external dimensions at the nanoscale, <100 nm (Nanoparticles, quantum dots, nanoshells), ii) **1-D** when nanomaterials have 2 external dimensions at the nanoscale, the third one is usually at the microscale (Nanofibers, nanotubes, nanowires, nanorods), iii) **2-D** when nanomaterials have only 1 external dimension at the nanoscale (platelets, thin films, layers, coatings) and iv) **3-D** when they have no external dimension at the nanoscale (Nanostructured material). Main types of nanoparticles that are usually found in a wide range of applications are: carbon nanotubes, magnetic NPs, quantum dots, dendrimers, polymers, liposomes etc.[4]

Due to their dimensions in nanoscale, nanomaterials have unique altered physical and chemical properties compared with their macroscale counterparts, such as the high surface to volume ratio and other novel physiochemical properties such as scattering, solubility, strength, diffusivity, toxicity, magnetic, optical, thermodynamic, etc. For this reason, nanotechnology is a rapidly advancing research field offering a wide range of opportunities for the development and application of structures, materials, or system with new properties in various areas like pharmaceutics, cosmetics, processed food, chemical engineering, high-performance materials, electronics, precision mechanics, optics, energy production, and environmental sciences.[1, 3]

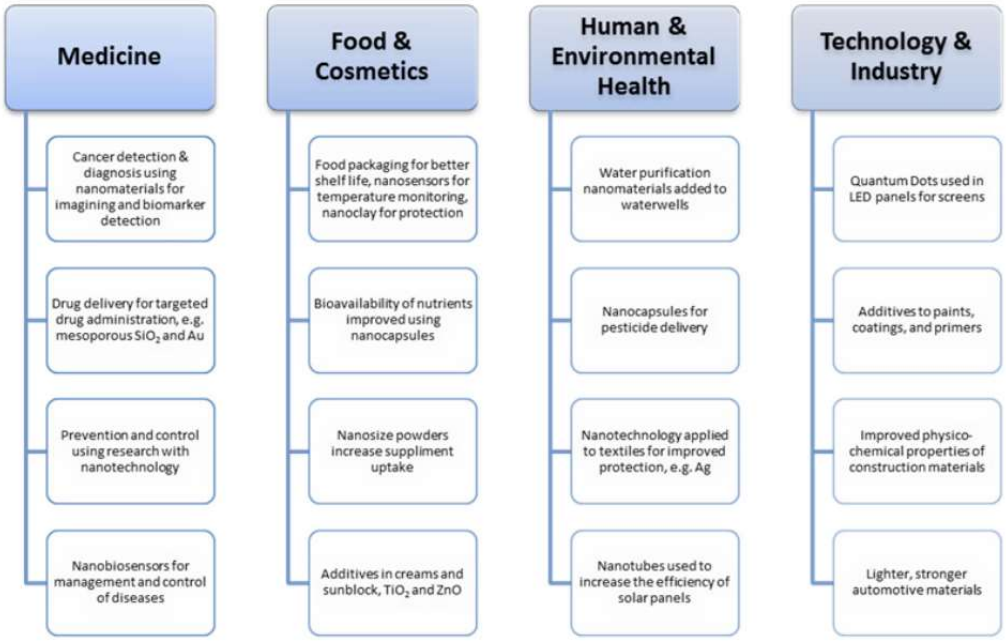

Figure 1. Applications and possibilities of NPs. [5]

According to StatNanoin 2018, more than 166,000 articles related to nanotechnology were published, indicating an increase of more than 7% compared to the corresponding period last year. In addition to research articles, over 9,000 review articles were also published in the past year, which comprise nearly 5% of all nanotechnology publications. Monitoring approximately 7000 commercial nano-enabled products available on global markets revealed that the properties of around 2330 products have been enabled or enhanced aided by nanoparticles.[6]

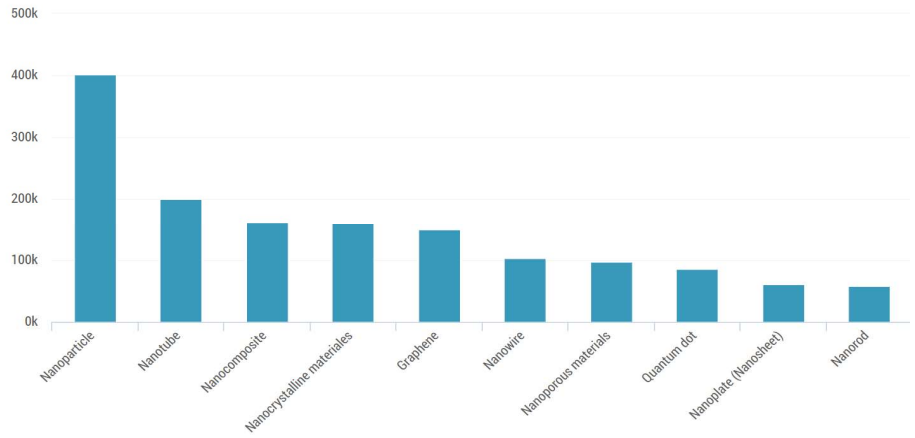

Figure 2. Nanomaterials with the highest growth rate in nanotechnology articles.[6]

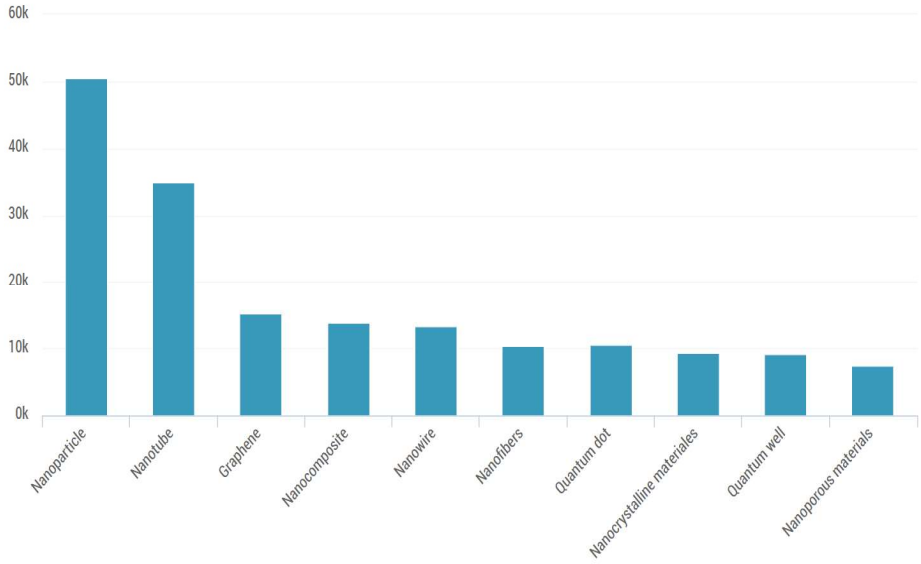

Figure 3. Nanomaterials with the highest growth rate in nanotechnology patents.[6]

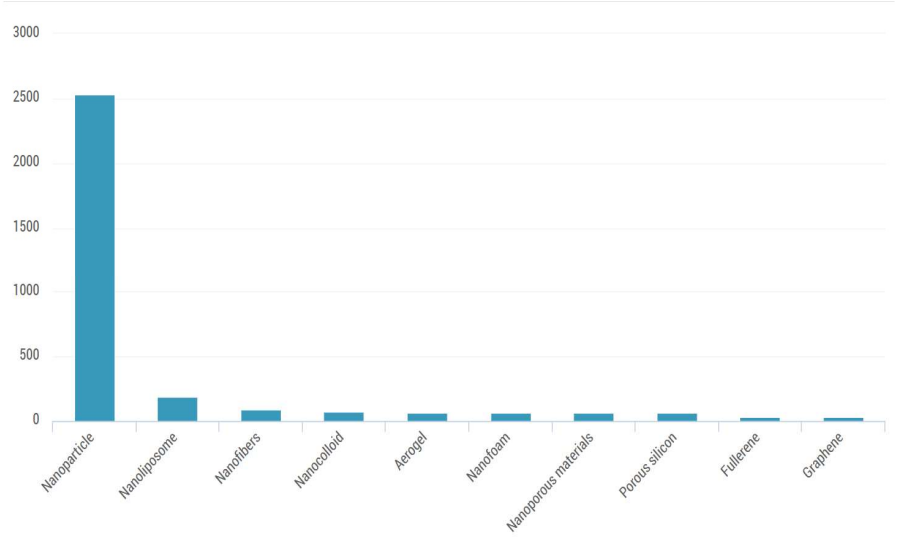

Figure 4. Nanomaterials with the highest growth rate in nanotechnology products.[6]

The number of nanotechnology products keeps increasing constantly and is becoming a part of our daily life. However, the novel properties that the nanomaterials exhibit, may have adverse effects and many of them are currently poorly understood.[7] The constant human and environmental exposure to nanomaterials has become a matter of great concern due to the potential nanotoxic effects.[8] Nanomaterials show many benefits in various fields, but there is not enough scientific evidence about how they impact on human

and ecosystem health in a holistic approach. There still existuncertainties about health, safety, and environmental effects as well as some definition and classification of nanomaterials are missing.[8] Thus, it is of paramount importance to identify the potential risk factors that are harmful to human health and the environment. For instance in the EU, the increasing number of specific provisions for nanomaterials inserted in EU legal instruments, as well as guidance and other technical documents, has alarmed the European Commission with regard to the need to ensure the use of harmonized terminology.[9] It is apparent that the toxicity properties of such nanomaterials are directly associated with the physical and chemical properties such as size, shape, chemical composition, surface covering, route and dose of administration of the concerned nanomaterials.[8] Today, there are several legislative measures such as regulations and documents with recommendations and guidelines in the European Union (EU), USA and other regions with specific references to NMs. (Figure 5) However, a single internationally accepted definition for NMs does not exist. Different organizations have a different approach in defining NMs. Recently scientists have challenged the arbitrarily selected size limit of 100 nm and propose that size definition is based on changes in material properties.[10] Nanotechnology is a novel and dynamic science that on one hand presents great potential, but on the other it lurks danger concerning the toxicity due to the lack of adequacy of information and the nature of nanoparticles. It is necessary that regulatory authorities from each region will attempt to address these risks and keep a balance with the benefits. In some ways, the regulatory regime that will emerge may be as innovative as the technology that it addresses.[11]

The present study provides a review of the current regulatory framework in nanocosmetics, medical devices and food supplements that contain NPs in three regions, EU, US and China.

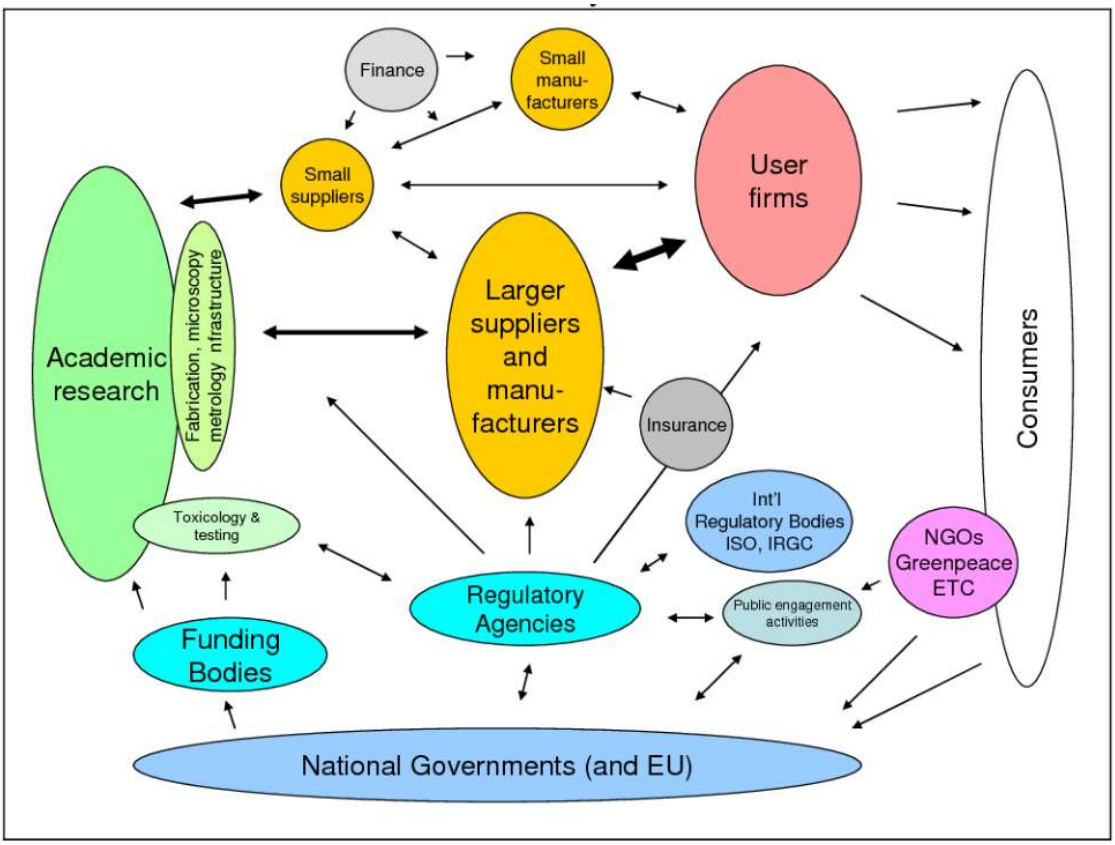

Figure 5. Schematic representation of the generic actors and main linkages in nanomaterials innovation systems. [12]

2. Nanocosmetics

2.1 What are nanocosmetics?

Cosmetics have been demonstrating rapid growth with the global cosmetic products market valued at USD 532.43 billion in 2017 while it is expected to reach a market value of USD 805.61 billion by 2023.[13] Nanotechnology displays the advance in the field of cosmetics by increasing the efficacy of the products in the context of their application, aesthetics and performance.[14] There are many applications of nanocosmetics, including anti-aging care, make-up, nails, deodorants, oral care, sunscreens and hair care.

Wide range of nanomaterials have been used in nanocosmetics such as: a) **liposomes** that are easy to prepare, present enhanced absorption of active ingredients by the skin and continuous supply of agents into the cells over a sustained period of time, b)**nanoemulsions** that provide higher stability and increase the shelf life of the product., c)**solid lipid NPs** with UV-resistant properties, d) **hydrogels** with long-lasting effect, e) **cubosomes** that have high heat stability and are capable of carrying hydrophilic and hydrophobic molecules, f) **nanocrystals** that allow safe and effective passage through skin, g) **dendrimers** are suitable for multifunctionalization, h) **nanocapsules** that decrease the penetration of UV filter, i) **silver and gold NPs** with enhanced antibacterial properties, ia) **fullerens or buckyballs** that have potential antioxidant ability (Figure 6).[15]

Nanomaterials in cosmetics can be divided in two categories: **i)** labile nanoparticles which disintegrate upon application to skin into their molecular components (e.g. liposomes, microemulsions, nanoemulsions), and **ii)** insoluble particles (e.g. TiO_2, fullerenes, quantum dots).The nano-enabled products seem to have superior performance in comparison to conventional like improved stability, enhanced skin penetration, controlled and targeted release and better encapsulation of drug.[16] Also, some of their advantages include improved UV protection, stronger structure, improved electrical conductivity, improved texture, and longer shelf life of the product which contain them.[17]

However, there are doubts about the healthy and the environmental impact of nanomaterials due to their size and their altered properties in nanoscale. There is a lack of knowledge about the toxicological profile and the potential health hazards of such materials. Hence, the need for the conduction of many more studies as well as the law-

making of more comprehensive legal framework for ensuring the human and environmental health is increased. Up to now there are not exist a uniform regulatory framework about nanocosmetics but every country approaches the cosmetics with different way.[18] The adoption of the Cosmetics Regulation is important because it provides a legislative instrument through which the challenge of distributing regulatory responsibilities for unknown risks may be explored.

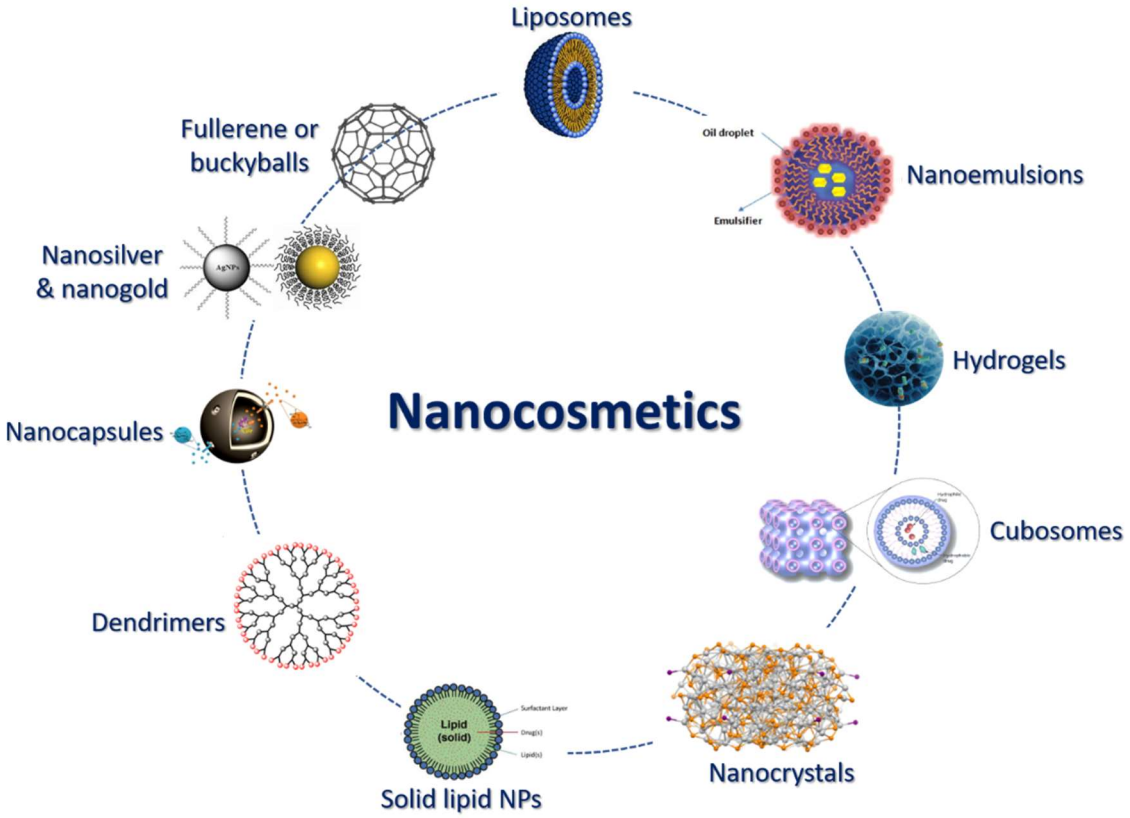

Figure 6. Type of NPs that are used in nanocosmetics. [14]

2.2 Regulation in Europe

2.2.1 EU Regulation 1223/2009

The first legal instrument in the EU containing a specific regulatory framework on cosmetic products is the revised Regulation on Cosmetic Products (1223/2009) that was entered into force on 11 January 2013. The Regulation covers all substances that are contained in

cosmetics including the nanomaterials. Specifically, it defines the term "nanomaterial" as "*an insoluble or biopersistant and intentionally manufactured material with one or more external dimensions, or an internal structure, on the scale from 1 to 100 nm* "[Article 10].[19] The revised regulation is robust about the pre-market notification of information on nanomaterials [Article 13] as well as the labelling of nanomaterials in the list of ingredients [Article 19]. The regulation also focuses on the risk assessment of cosmetics using nanomaterials as ingredients that require special attention [Article 16]. The Scientific Committee SCCS is in charge for these evaluations in order to increase as much as possible the safety of cosmetic consumers.[20, 21]

2.2.2 REACH and CLP

There are certain jurisdictive provisions that cover nanocosmetics implicitly. REACH (Registration, Evaluation, Authorisation and Restriction of Chemicals) and CLP (Classification, Labelling and Packaging) offered the best possible framework for the risk management of substances including nanomaterials. REACH is the most inclusive legislative provision for chemical substances in the EU and applies to chemicals in whatever size, shape, or physical state in order to be ensured the human and environmental safety. In REACH, challenges regarding the safety of nanomaterials are dealt with by the ECHA (European Chemical Agency) that is responsible for implementation of REACH. Moreover, the ECHA provides recommendations on scientific and technical issues concerning the implementation of REACH and CLP legislation in regard to nanomaterials. According to ECHA, nanomaterials that fulfil the criteria for classification as hazardous under Regulation 1272/2008 on classification, labelling and packaging (CLP) of substances and mixtures must be classified and labelled. This applies to nanomaterials as substances, or nanomaterials as special forms of the substance. Today, there are data sheets concerning the classification and labelling of many type of chemical substances.[22, 23, 24]

2.2.3 Placing a new cosmetic in EU market

In order to import a cosmetic product into the EU market, the product should meet some requirements according to EU Regulation 1223/2009.[20] All cosmetic products should provide a product information file (PIF) including a description of cosmetics product and a

product safety report (CPSR). To this end, a Responsible Person (RP) is appointed for the development of this file. The responsible person (RP) is a natural or legal person established in the European Community such as a manufacturer, an importer, or a distributor, acting on behalf of all the European countries. The role of the Responsible Person is to ensure that "*the product is safe for human health under normal and reasonably foreseeable conditions before the European authorities*" [Article 3]. Furthermore, the RP assures that the safety evaluation of product, established under annex 1, takes into account the intended use of the cosmetic product and the expected systemic exposure to individual ingredients in a final formation.

The collection of requirement data under the regulation is mandatory for importing of cosmetics in market. The cosmetic product safety report (CPSR) includes all this information concerning the cosmetic product and test data in order to ensure the human safety and identify the potential risks. Specifically, the safety assessment involves data about the quantitative and qualitative composition, the physical and chemical characteristics, the stability of product, the impurities, traces in the product and information on the packaging material as well as information regarding to the normal and foreseeable use, exposure to the product, exposure to the ingredients, toxicological profile of the ingredients and the potential undesirable effects of the product. Also, it is crucial that the product provides labelled warnings and instructions of use according to the relevant regulations. All these processes have to be complied with Good Manufacturing Practices (GMPs) set by the ISO 22716 standard according to the Article 8 of the EU Regulation. The GMPs are very important for the cosmetic product placing in EU market ensuring the quality of the manufacturing process of cosmetic products.

The final step of the process is the announcement of the cosmetic product on the CPNP (Cosmetic Products Notification Portal) in order to import and establish the product in the European market. The rules of this procedure are described thoroughly in Article 13 of EU Regulation.[10, 20]

2.2.4 Toxicity of NPs in cosmetics

There are concerns about the NPs used in cosmetic products and the potential toxicity that they can cause. Toxicity may depend upon size, shape, surface charge, age, etc of the nanomaterials, so their complexity is rising doubts about their human health and environmental impact. The toxicity is also affected from the degree of exposure and the route inside the human body. Nanomaterials can enter the human body by three possible ways: dermal absorption, inhalation and ingestion.[22]

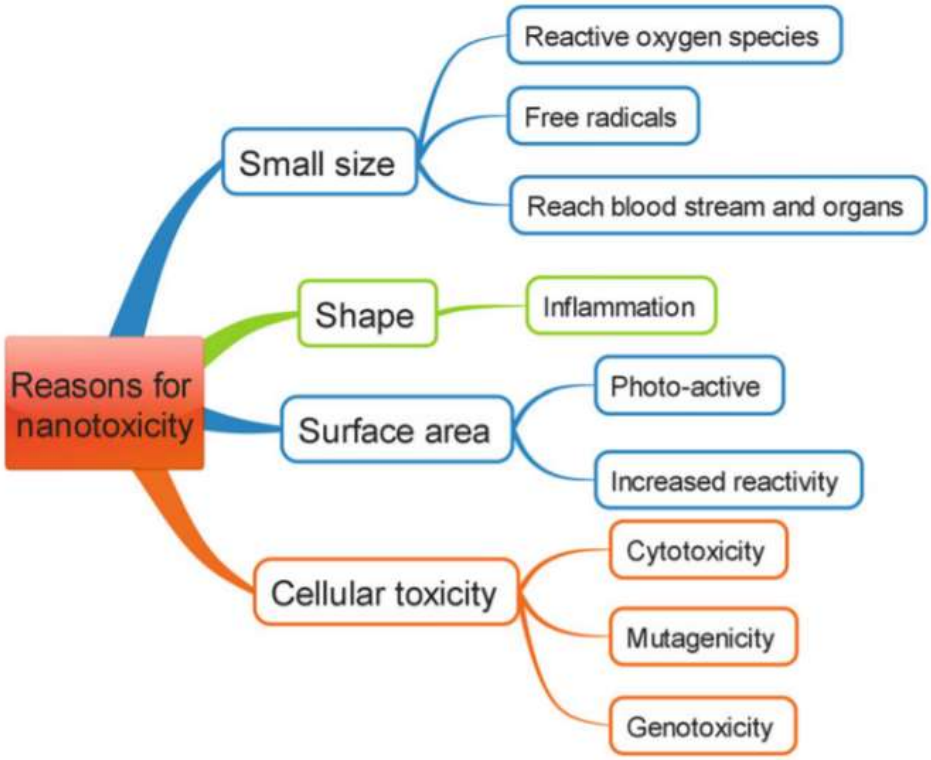

Figure 7. Some causes of nanotoxicity and some of the consequences.[25]

Dermal absorption: The cosmetics are applied directly to the skin. The skin is the largest organ of the body that comprises of three layers: the epidermis that is the exterior layer of skin, the dermis, and the hypodermis. The epidermis is split into several layers and its exterior layer, the stratum corneum, is responsible for the barrier function of the skin.[26] Nanomaterials sometimes have the capability to transcend into the deeper skin layers and even to pass into the bloodstream.[27, 28] Studies that were carried out in pig skin showed

that the penetration of nanomaterials in barrier layers occurred within 24 hours of exposure.[16]

Inhalation: In the case of inhalation of NPs, many studies show that the larger number of nanoparticles inhaled enter the pulmonary tract and some can travel via nasal nerves to the brain and enter into other organs via blood.[29] Likewise, nanomaterials pass quickly out of the body after ingestion but can sometimes be taken up to relocate to the organs. For instance, silver nanoparticles that are widely used in cosmetic products due to their antimicrobial properties, were administered in rats and the results have shown size decrease and irregularities in shape as well as drastically reduction in the function of mitochondria and cell viability. [30]

Ingestion: Similarly, NPs were located in spleen, heart, liver, bones, and pancreas of mice when they were orally ingested with zinc oxide nanoparticles. Copper nanoparticles, also demonstrated undesirable effects and heavy injuries to internal organs of the mice. [31]

Despite the fact that several studies have been carried out in order to evaluate the identification toxicity impact of nanocosmetics, it is still unclear what NPs can cause in health. Various conditions and specific properties of the NPs like their small size, their penetration into the body and their migration and mobility to organs should be evaluated.[18] This complexity leads to the need of testing for all possible parameters, but this requires both time and money investment in the near future.

2.2.5 Toxicity studies

The issue of the identification of the toxicity that a cosmetic ingredient can cause is of paramount importance for the overall safety assessment of the cosmetic product. The toxicity tests have been focused on ingredients that are raising concerns for human health and safety. For instance, colouring agents, preservatives and UV-filters (Annexes IV, V and VI to Regulation (EC) No 1223/2009) and banned and restricted substances, respectively (Annexes II and III to Regulation (EC) No 1223/2009).The EU guidance provides a wide range of toxicity studies in order to determine the undesirable effects of substances that are used in cosmetic products. In order to give a more comprehensive approach concerning the toxicity testing and safety assessment, the European Commission's (EC) Scientific

Committee on Consumer Safety (SCCS) released the tenth revision of The SCCS Notes of Guidance for the Testing of Cosmetic Ingredients and their Safety Evaluation (November 2018). It is an advisory guidance that provides recommendations with the aim to improve harmonized compliance with the cosmetic European Union (EU) regulation.

According to SCCS, the existed data relating to the toxicological profiles of the ingredients were delivered from experimental animals, using whenever possible the same exposure route as in humans. However, cosmetics are excepted because animal testing has been prohibited since 11 March 2013 according to EU Cosmetic legislation (Regulation 1223/2009). Thus, the marketing of cosmetic products that their ingredients have been tested in experimental animals is not allowed. The EU regulatory has been trying to reduce the using of experimental animals. In particular, it has been proposed a key strategy, the "Three Rs" (Replacement, Reduction and Refinement) aimed at achieving the goal of humane experimental techniques and the use of fewer animals. In this context, non-animal methods have been developed with the aim to evaluate the cosmetics safety and efficacy. For instance, tests about skin corrosion (EPISkin and EPIDerm), skin irritation (Episkin, which is a reconstructed human skin model), mutagenicity/genotoxicity, phototoxicity (3T3 NRPT-3T3 fibroblasts neutral red uptake phototoxicity testing, applicable to UV absorbing substances), dermal absorption (studies human/pig skin) and serious eye damage (Bovine Corneal Opacity and Permeability (BCOP) and the Isolated Chicken Eye (ICE) that are non-validated in vitro tests. Nevertheless, due to the complexity of the mammalian in vivo systems, there are presently no validated (animal-free) replacement methods for acute and repeated dose toxicity, including reproductive and developmental toxicity, and carcinogenicity.[26, 32]

It is worth noting that these toxicity tests have been used only for conventional cosmetic substances and they have not been validated for nanomaterials yet. Hence, it is raising doubts if these methods can be implemented for nanomaterials in order to evaluate the potential toxicity performance.[26, 33]

2.3 Regulatory in USA

2.3.1 Guidance for Industry: Safety of Nanomaterials in Cosmetic Products

Cosmetic products in US are regulated by the Federal Food, Drug and Cosmetic Act (FD&C Act). In particular, the FDA provides a guidance for "Safety of Nanomaterials in Cosmetic Products". An FDA Task Force (2017) was allocated to discuss issues relating to the safety and efficacy of nanotechnology products in the FDA as well as to provide a guidance (Ref. 2). According to the guidance, the FD&C Act doesn't require cosmetics and substances, except colour additives, that should be approved before their import in market. However, the manufactures ought to consider making certain that the nanocosmetics are safe and not adulterated, thus it is necessary to collect the relevant data that ensure that their product is safe for human health. The Task Force underline that FDA have to request the submission of data and other information addressing the effects of using nanomaterials in products that are not put through to premarket approval, such as cosmetic products.[34]

Initially, the FDA guidance provides a definition of term nanotechnology that manufacturers should utilize to determine whether their products engage nanotechnology. In particular:

1." whether a material or end product is engineered to have at least one external dimension, or an internal or surface structure, in the nanoscale range (approximately 1–100 nm)" and

*2." whether a material or end product is engineered to exhibit properties or phenomena, including physical or chemical properties or biological effects, that are attributable to its dimensions, even if these dimensions fall outside the nanoscale range up to 1000 nm"(*FDA, 2014b). [33]

Typically, at the premarket review phase, the FDA take on these two considerations in order to inform manufacturers if their product includes nanotechnology from a regulatory point of view. On their side, manufacturers must take into consideration these points, carrying out safety assessments. Specifically, nanocosmetic manufactures use toxicological data that already exist for each ingredient but also conduct additional toxicological tests in order to completely ensure the safety of the end product.

2.3.2 Nanomaterials characterization

According to the regulation, all ingredients of the cosmetics, including nanomaterials should be described thoroughly. This description comprises: the nanomaterial name, the Chemical Abstracts Service (CAS) number, the structural formula, the elemental and molecular composition (including: the degree of purity and unknown impurities). As mentioned before materials in nanoscale exhibit altered physicochemical properties and biological interactions, raising doubts about the performance, the quality, the safety and the effectiveness of the product that embedded them. So, physical and chemical characterisation of the nanoparticles as well as the identification of potential impurities are necessary for the adequacy and safety of the final nano-product. For instance, the size, shape, charge, agglomeration as well as the stability and solubility of the nanoparticle are some of the properties to be evaluated.[33]

2.3.3 Toxicology evaluation

The issue of the toxicity to identify the potential toxicological performance of nanoparticles depends on its route of exposure as well as the uptake and their absorption. Nanomaterials used in cosmetics can be soluble and/or biodegradable or insoluble, sufficiently stable and/or biopersistent nanoparticles. The last can cross biological membrane barriers and they are able to cause toxicity to organs. As mentioned in previous section the health hazard that nanomaterials can create depends on the degree of exposure and the route that can enter in the human body, dermal absorption, inhalation and ingestion. Manufacturers should therefore consider systemic exposure to nanomaterials and take into consideration the absorption, distribution, metabolism and excretion (ADME).

In respect to the toxicity tests, FDA recommend the same in vitro methods as were described in section 2.2.5 for EU. (Figure 8) In contrast to EU regulation FDA consider in vivo testing necessary in order to obtain several data regarding to translocation, biodistribution, accumulation, and clearance of substances. There are several issues that have to be taken into consideration concerning in vivo toxicity testing for nanomaterials such as the issue of dose metrics (mass, volume or number of particles). The manufacturer

should consider the surface area and number of particles, as well as mass concentration in the study design of in vivo toxicity testing. [FDA regulation] [33]

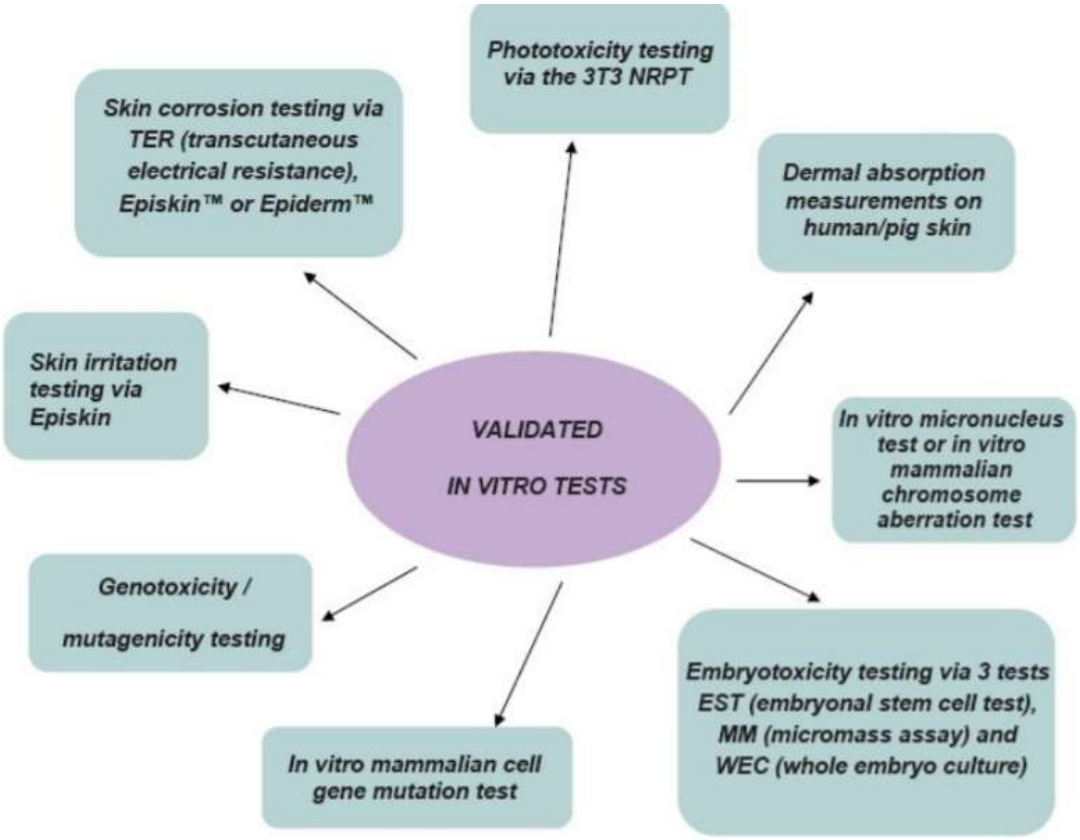

Figure 8. Validated in vitro Methods employed.[16]

2.3.4 Other regulations

For FDA is substantial to evaluate the potential risks of end products using nanomaterials. Thus, data collection seems to be the main objective for the FDA regulating more effectively the safety of cosmetic nano-enabled products.[19] Likewise, the manufacturers have to comply to the data collection requirements such as hazard identification, exposure assessment, dose-response assessment, risk management and characterization Furthermore, FDA recommend other regulations for cosmetic products that manufacturers can follow. For instance, Personal Care Products Council (PCPC) provides regulations on voluntary facility registration (Voluntary Cosmetic Registration Program (VCRP) and

voluntarily collection information of substances and the potential hazard effects of cosmetics.[18]

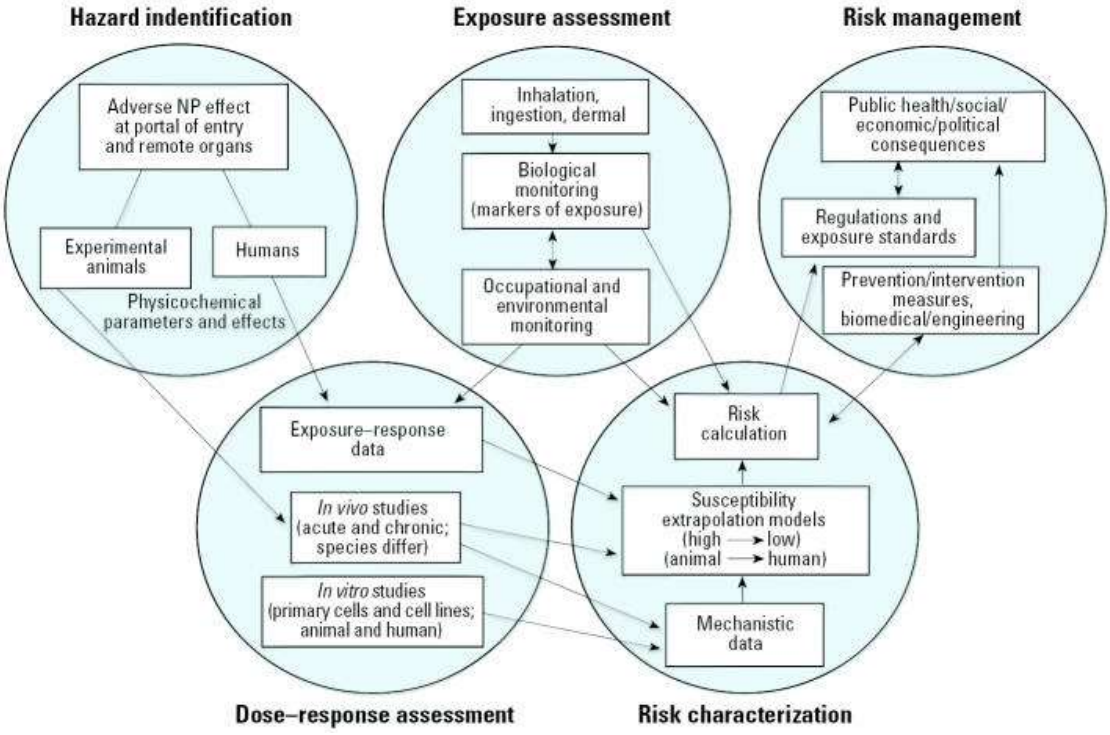

Figure 9. Risk assessment and risk management paradigm for NPs.[35]

2.4 Regulatory in China

The main regulatory agency for cosmetics in China is the National Medical Products Administration (NMPA), previously called China Food and Drug Administration. In addition, the Cosmetics Hygiene Supervision Regulations provides regulations for cosmetic products while the Safety and Technical Standards for Cosmetics 2015 includes prohibited, restricted or authorized substances in cosmetic products. [13, 36]

According to CFDA, the cosmetics can be divided in two types: non-special use cosmetics (for hair care, skin care and nail products, and fragrances) and special use cosmetics (hair regeneration products, deodorants and sunscreens). Each category of cosmetics requires a different type of permission from the State Food and Drug Administration (SFDA) and different pre-market applications depending on the type of cosmetic product and the origin

of the manufacturer.[33] All overseas cosmetics manufacturers must complete a safety assessment in order to obtain license before selling the product on the Chinese market.[13]

In respect of new cosmetic ingredients not included in the Inventory of Existing Cosmetic Ingredients in China (IECIC) it is necessary to register in the NMPA before being first used in cosmetic products. NMPA is not approving cosmetic products containing a new ingredient or a prohibited ingredient specified in Cosmetics Safety and Technical Standards 2015.

Regarding the testing, the new cosmetic products must be tested in NMPA-designated testing Institutions in China that have been authorized to perform the assessments for safety and quality of the end product. Cosmetics must undergo tests such as microbiological, toxicological, chronic toxicity, carcinogenic test, and conducting safe-for-human-use trials. China is trying to mitigate the requirements on animal testing for cosmetics and is working on setting up alternative methods and verification institutions. Animal testing are mainly required for imported cosmetics products while for national non-special use cosmetics they have stopped since 2014. [37]

In addition, the labelling of products is paramount of making products available on the market. Specifically, the Regulation on Cosmetic Label Management (former AQSIQ Decree No. 100) provides guidance and requirements on the contents and elements of labels, prohibited contents as well as marking and labelling methods. In parallel, the Regulations Concerning the Hygiene Supervision over Cosmetic gives general specifications on labelling (Article 12).

In conclusion, China has not established a specific regulatory framework for cosmetics that contains nanoparticles yet, but is considered that the nanoparticle are a new ingredient of cosmetic product and are assed in the context described above. China is monitoring legislation developed in EU and USA as a benchmark for their development. Especially they keep their eyes on the debate on REACH and nanomaterials.[14]

2.5 Future perspectives

The last decades, nanotechnology in cosmetic products is an attractive and promising field that presents rapid spread and commercialization. Growth of nanocosmetic market is

increasing constantly because it can offer excellent opportunities for both research and business.[25] Nevertheless, the impact in the human health and the environment is ambiguous. In this context, assessment and regulation of cosmetic products that consist of nanomaterials are crucial. It was realised that definitions and positioning concerning nanomaterials vary for every country and its administrative unit. The "lack" of a uniform and international regulation framework can cause undesirable results both for end users, who are unaware about the products that they use, andfor manufacturers/suppliers who find themselves without legal guidance on how to use and sell nanocosmetics. [38] It is crucial that the nanomaterials terms are defined and regulations to be more practical, thus specific thresholds should be included. Furthermore, the inadequacy of effective toxicity testing regarding to long-term exposure to NPs and bioaccumulation is a crucial issue that needs to bead dressed in order to prove the safety and the efficacy of these products. Each agency from each country is responsible for the implementation of the regulatory specifications by manufactures for the purpose of offering safe products.

As was described above, nanotechnology safety issues are mentioned in FDA comprehensive guidance, although, documented procedures are voluntarily applied by manufacturers creating a legal breach in this market. The FDA regulatory framework for products that are categorised only as cosmetics is minimal, especially compared to the regulation of products in the EU and other countries. The table below contrasts between the regulation of cosmetics in the EU and the US. (Table 1) In EU, the EC regulation is stricter in order to ensure the safety of users in contrast to China that has not established a specific regulatory framework for nanocosmetics. Hence, in comparison to the USA and the China, the EU market has a robust cosmetics regulation framework. It is clear that there is a need for more comprehensive and stricter regulatory framework for nanocosmetics in order to ensure human and environmental safety. The scientific community already recognized the need to develop innovative industrial analytical techniques, more accurate and detailed, ensuring reliable physicochemical and toxicological data.[39] The development of integrated risk assessment and decision frameworks is also crucial in order to enable forecasting the potential impacts of nanomaterials on human health and the environment and adequate risk management. This proceeding may require the development of new, reliable, reasonable but quicker risk assessment strategies to replace the current ones. [40]

In conclusion, producing a universal approach adopted by the US, EU and China is imperative for managing the entry of nanocosmetic products in the market. The inferences are significant for the jurisdictions themselves, as well as other jurisdictions that are looking to the US and the EU for guidance on how they could and should regulate nanocosmetics.[17]

Table 1. Differences of cosmetic regulation between EU and US.

EU	USA
Submit certain information prior to placing products on the market	Manufacturers choose whether to share their safety information with FDA
Report product non-conformities that may pose risks to human health	Cosmetics cannot be adulterated or misbranded but are not subject to affirmative reporting requirements for non-conformities
Comply with Good Manufacturing Practices (GMP)	No GMP regulations (only nonbinding recommendations) for cosmetics
Conduct safety assessments that contain specified data	Manufacturers generally define how they test their products and ingredients for safety
Comply with list of more than 1300 prohibited substances and hundreds of restricted substances	Except for colour additives and a few restricted ingredients, cosmetic manufacturers can use almost any substance in a cosmetic without FDA approval (though products that are harmful to health would be considered adulterated)
Follow safety and adverse event reporting requirements	No mandatory adverse event reporting
Register all cosmetic products in a centralised databased	Registration with FDA is voluntary (though some US states require registration of cosmetic manufacturers and distributors

3 Medical devices

3.1 Medical devices and nanomaterials

In the past few decades, medical devices had life-changing impact for patients, while at the same time lowering healthcare costs. Medical devices are products or equipment intended for medical use, covering a wide range of products, including common ones, e.g. sticking plasters, glasses and wheelchairs and even more high-tech equipment such as implantable devices, X-ray machines, MRI scanners and artificial limbs.[41] Also, devices that are used to examine human samples (blood, urine, tissue, etc.) outside of the body are termed in-vitro diagnostic medical devices (or IVD). These are widely used in healthcare and can be used for the detection of many types of infections/diseases as well as monitoring the progress of drug therapies. The development of these devices is significant for the healthcare industry and for the improvement of the quality of patients' life.

Nanotechnology is widely used in many aspects of the development of medical devices. Nano-enabled engineered biomaterials (nanobiomaterials) have shown unique properties when they are incorporated into medical devices. Engineered nanobiomaterials that are commonly used in MDs can be divided in the following groups: (1) surface nanostructures (e.g. nanosize hydroxyapatite scaffolds, CNT composites); (2) nano-objects bound or incorporated with in a medical device, not intended to be released (e.g. nanosilver–catheters, wound dressings, nanotitania–endosseous implants); (3) nano-objects/nanostructures on the surface of or within a medical device, intended or expected to be released from the device; (4) nano-scale medical devices (e.g. organic core EBMs with antiviral activity, drug nanocrystals, QDs, Chitosan, PLGA, liposomes, polymers); (5) nano-objects released from a MD as product of degradation, wear, or from mechanical treatment processes (e.g. process of polishing in dentistry, local generation of nanoparticles as a result of wearing of knee or hip implants). [42]

The safety of medical devices that contain nanomaterials is of increased significance, particularly in the case of direct contact with the surface of the human body. It is unclear if the absorption and distribution of nanomaterials in the human body could cause undesirable effects.[43]

The effect of a MD is not related to chemical, pharmacological, immunological, or metabolic processes. MDs consist of biomaterials, so the biocompatibility is the most important parameter that requires special attention. In order to ensure the biocompatibility of MD a series of assessments are conducted such as biocompatibility testing of the biomaterials by in vivo implantation or other studies, safety testing (local and systemic toxicology of the finished device or clinical product), and efficacy testing of the functioning finished device or clinical product. For MDs, all biocompatibility and adequacy testing happen preceding any clinical testing. Monitoring and applying the correct testing standards to improve and test devices can boost their market penetration in national and global scales.[44] There are ongoing discussions between medical device manufacturers and regulatory authorities regarding the standards and methods required for the evaluation process. The regulatory control of nano medical devices should consider the product's safety, efficacy and quality. The efforts for worldwide regulation and validation of medical devices are a topic of great interest and importance in order to protect human health.[40] However the wide range of the categories of devices lead to a difficult and intricate MDs regulatory environment.

3.2 Regulations in Europe

3.2.1 Regulation (EU) 2017/745and Regulation (EU) 2017/746

Medical devices are regulated by European Medicines Agency (EMA) and in the case of specific categories of medical device by European Union (EU) legislation. Two new regulations on MD were enacted on April 2017, in order to replace the existing directives. These are **Regulation (EU) 2017/745** on medical devices (MDR) and **Regulation (EU) 2017/746** on in vitro diagnostic medical devices (IVDR) and they came into force on May 2017. Both new rules have a transitional period, i.e. 3 years after publication for medical devices and 5 years after publication for in vitro diagnostic (IVD) medical devices, in order to become effective. The new regulations seem to have a stricter approach for medical devices in comparison to previous regulations (Directive 93/42 and 98/79) for in vitro diagnostics. The new regulations include recommendations and rules about the design and the manufacturing of the devices as well as the required clinical tests, the authorization and post-market monitoring. Regarding the using of nanomaterials in medical devices the

Regulation (EU) 2017/745 provides recommendations in Article 15 about the issues that manufacturers should take into account in order to ensure the safety of their product as well as in Articles 2 and 3 that include certain definitions with nanotechnology.[45]
Medical devices in EU should have a valid (Conformité Européenne) marking before their placing into the EU market. Medical devices are undergoing a conformity assessment with the aim to show that meet legal requirements and ensure their safety for human health as well as perform as intended. This assessment contains the surveillance of the manufacturer's quality system and a technical review from the manufacturer concerning the safety and performance of the device. The manufacturers must develop risk and quality management methods based on existing devices. Notified body is in charge of conducting the conformity assessment. A notified body is an organisation elected by an EU country to evaluate the conformity of certain products before their import on the market.[44]

Furthermore, the regulations highlighted the responsibility of importers regarding the reliability of the product: *"Importers shall keep a register of complaints, of non-conforming products and of product recalls and withdrawals, and provide the manufacturer, authorised representative and distributors with any information requested by them, in order to allow them to investigate complaints*".[40]

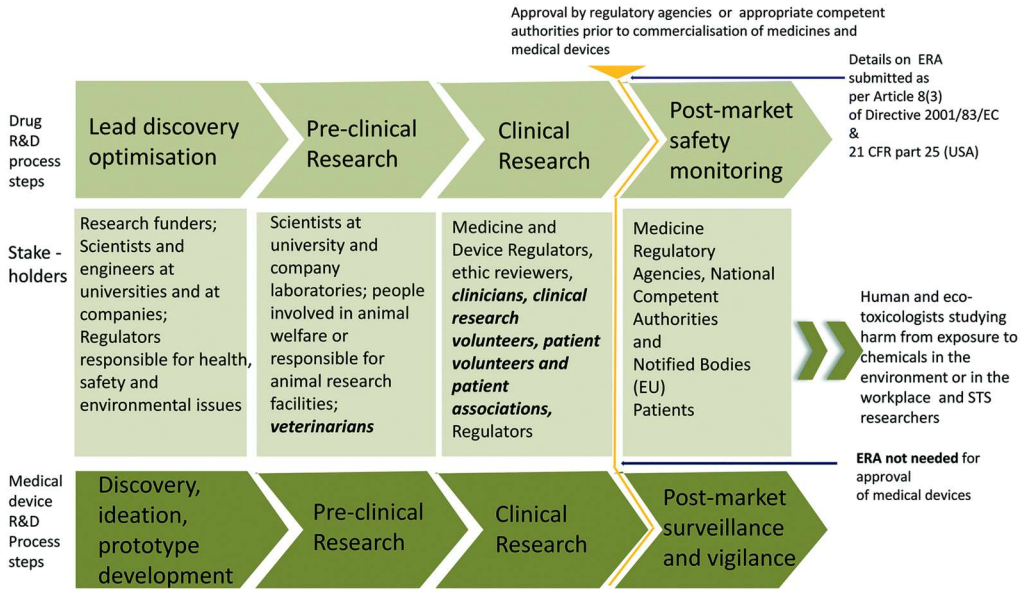

Figure 10. General stages of (nano) medicine and medical device research, development and commercialisation onto which key stakeholders involved in these stages are mapped. ERA: environment risk assessment. [46]

3.2.2 EU Scientific Committee on Emerging and Newly Identified Health Risks

The EU Scientific Committee on Emerging and Newly Identified Health Risks (SCENIHR) provides a "Guidance on the determination of potential health effects of nanomaterials used in medical devices". This Guidance provides information concerning the risk appraisers that need to be considered in the safety assessment of nanomaterials. In particular, guidance provides recommendations about physico-chemical characterisation of nanomaterials, the determination of hazards, and risk assessment of nanomaterials used in medical devices. A phased approach is recommended for evaluating the risk of the use of nanomaterials in medical devices based on potential release and characteristics of the nanomaterials to avoid unnecessary testing. (Figure 11). In the first phase is examined the probability of nanoparticles to be released in order to estimate the potential exposure. The objectives of this phase are to identify the kinetics of the particles to address the toxicity testing needed in Phase 2 based on potential exposure scenarios. Moreover, in this phase are identified the number and the duration of particle presence in a specific tissue because they affect the probability of adverse effects occurring. The phase 3 includes the hazard assessment (toxicological evaluations) and the 4 includes the risk characterisation/risk assessment based on the possibility for exposure. [47, 48]

Figure 11. The phases for risk evaluation of the use of NPs in MD.

3.2.3 Toxicological evaluation

The toxicity evaluation process for medical devices that contain nanomaterials is determined by its potential of external and internal exposure. The most common exposure routes to nanomaterials that released from medical devices are the following:

- inhalation exposure (intubation, dental procedures)
- dermal exposure,
- mucosal exposure (via various mucosal tissues, e.g. in the mouth, vagina/penis),
- oral exposure,
- parenteral exposure (introduced into the body by a means other than through the gastro-intestinal tract, e.g., intravenous or intramuscular
- ocular exposure

Medical devices can be divided in two types: Non-invasive and Invasive medical devices. The first type contains devices that come in contact with skin and the released nanstructures have a low potential to penetrate through the skin. Unlike, the released nanparticles of invasive devices are enabled to come into the body directly depending on the placement of the device used. So, they have high potential for systemic exposure
There are several validated test methods (in vitro, in vivo, in silico) that can assess MDs containing nanomaterials. The degree of nanomaterial toxicity depends upon the particle size and additional specific characteristics. Therefore, it is essential the tests to be conducted using the same nanomaterial with the same chemical composition, size and size distribution, surface properties and purity/impurity profile as the substance present in the medical device and should be fully characterized before testing. Thus, the information on the nature and stability of the test substance under experimental conditions is of prime importance for the interpretation of any test results. The guidance recommends if a similar (nano)material is used, this should be justified and documented.

According to guidance, attention should be given to some parameters that can influence the toxicological profile of materials that are used in MDs. For instance, the agglomeration/aggregation behavior, the insoluble/ partially-soluble nature of nanomaterials and surrounding media that can alter the performance of nanomaterials. Special care is therefore needed in regard to the applied doses, which can be affected by the above-mentioned parameters.

The validated toxicity testing methods that are applied in order to identify the toxicity risk of medical devices containing nanomaterials are: cytotoxicity, acute toxicity, irritation activity, delayed-type hypersensitivity, genotoxicity, in vitro genotoxicity testing, in vivo

genotoxicity testing, haemocompatibility, repeated-dose toxicity, implantation, chronic toxicity/carcinogenicity, reproductive and developmental toxicity.

According to ISO 10993-1: "Biological evaluation of medical devices-Part 1: Evaluation and testing within a risk management process", the safety assessments that are required with the aim to be identified the toxicological profile of nano medical devices relate on the type of medical device, type of contact and duration of exposure as is described in Table 2.[49]

Table 2. Framework for specific nanomaterial toxicity testing based on potential release (exposure) of nanomaterials from medical devices.[48]

Testing proposed	Non-invasive short term use	Non-invasive long term use	Invasive short term use	Invasive long term use
Low exposure	Phys: chem data	Phys: chem data	Phys: chem data	Phys: chem data
	Cytotoxicity in vitro	Cytotoxicity in vitro	Cytotoxicity in vitro	Cytotoxicity in vitro
	Irritancy in vitro	Irritancy in vitro	Irritancy in vitro	Irritancy in vitro
	Hypersensitivity	Hypersensitivity	Hypersensitivity	Hypersensitivity
		Genotoxicity in vitro		Genotoxicity in vitro
				General Immuno toxicity testing
Medium exposure Additional tests		Genotoxicity in vivo	Other in vitro plus in silico testing*	28/90 day in vivo toxicity test
		Immuno toxicity at location site	Genotoxicity in vitro and in vivo	In vitro and in vivo (repeated dose) genotoxicity testing
		Persistence /accumulation studies at location site only		ADME including persistence /accumulation studies
High exposure Additional tests	Selected in vivo acute toxicity tests focussed on location site(s)	Selected in vivo chronic toxicity tests focussed on location site(s)	In vivo acute toxicity tests	In vivo chronic toxicity tests may include reprotox depending on patient group.

3.2.4 Classification of medical devices in EU

It is crucial for manufacturers to know the class that the MD is matched because this can affect the regulatory specifications, the procedure of approval of the MD and its associated costs. The EU regulation framework have defined a MD classification scheme. Essentially, the devices are grouped in 4 types: Non-invasive devices, Invasive medical devices, Active medical devices, Special Rules (including contraceptive, disinfectant, and radiological diagnostic medical devices). The MDR and IMDR provide different classification scheme.

The **medical devices** have the following classes:

Class I: Provided non-sterile or do not have a measuring function (low risk)

Class I: Provided sterile and/or have a measuring function (low/medium risk)

Class IIa: (medium risk)

Class IIb: (medium/high risk)

Class III: (high risk) [43]

The **in-vitro medical devices** can be divided in these classes:

I. All Other Devices

II. Devices for Self-Testing Not Listed in Annex II

III. Device of List B, Annex II

IV. Device of List A, Annex II [50]

In the case of the devices incorporate or consist of nanomaterials are classified in the highest risk class, class III, if they present a high or medium potential for internal exposure and should be subjected to stricter evaluation procedures or in class II if they present a low/negligible potential for internal exposure. [49]

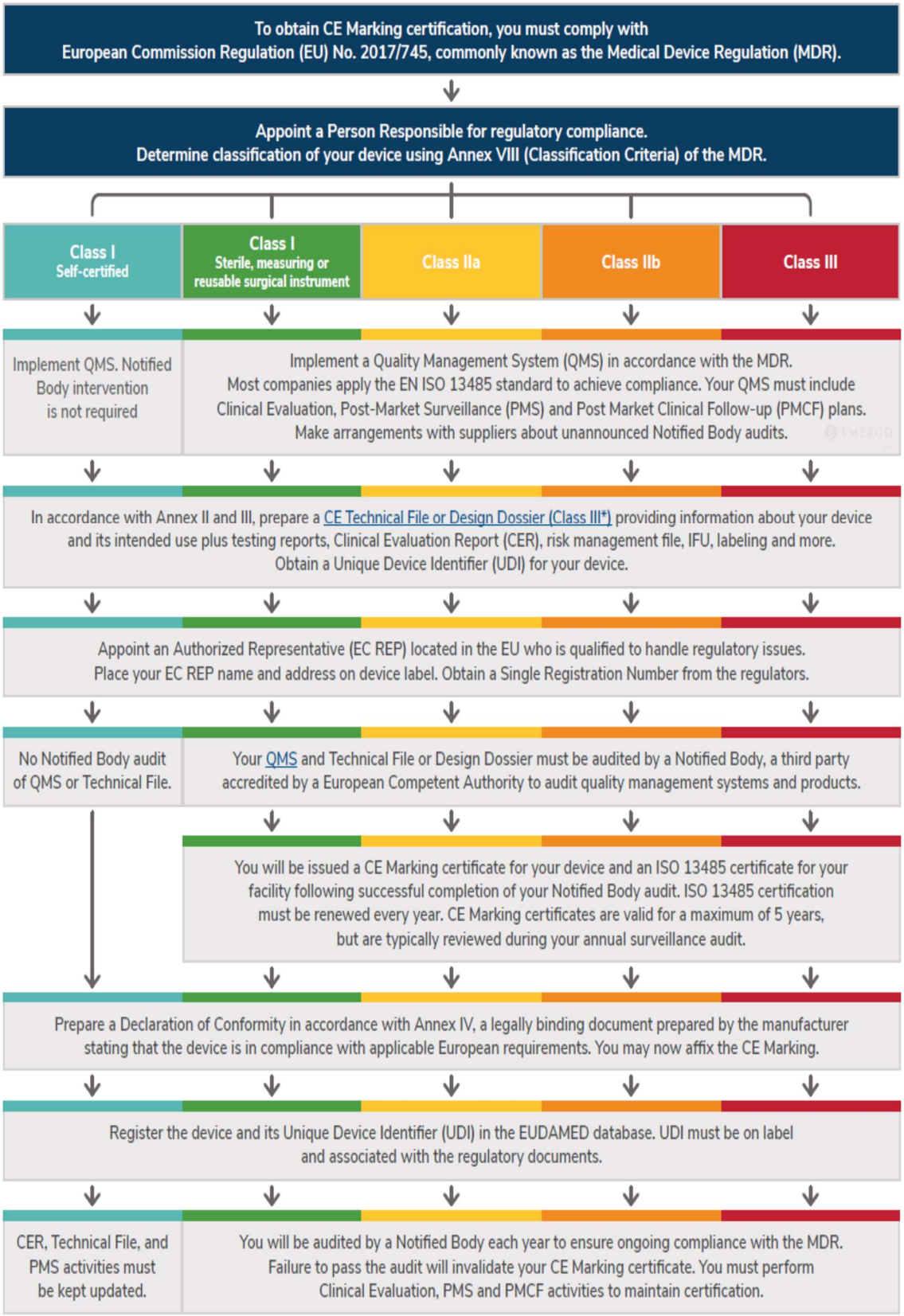

Figure 12. CE approval process in Europe.[51]

Device classification in Europe →	Class I Self-certified	Class I Sterile, measuring or reusable surgical instrument	Class IIa	Class IIb	Class III
How long you should expect to wait after submission until approval is granted.[1]	See note 1*	See note 1	See note 1	See note 1	See note 1
Validity period for CE Marking certificate.[2]	Not Applicable	5 years	5 years	5 years	5 years
Registration renewal should be started this far in advance.[3]	Not Applicable	6 months	6 months	6 months	6 months
Complexity of the registration process for this classification.[4]	Simple Complex 1	Simple Complex 2	Simple Complex 3	Simple Complex 4	Simple Complex 5
Estimated cost (USD) of gaining regulatory approval.[5]	Low High 2	Low High 3	Low High 4	Low High 5	Low High 5

Figure 13. EU classification of Medical Devices.[50]

3.3 Regulations in US

3.3.1 Current regulatory framework

The FDA Nanotechnology Task Force (2006) produced a guidance for FDA-regulated products such as new drugs, medical devices, and foods containing nanomaterials. The purpose of this guidance is to provide recommendations with the aim of the continued development of nano-enabled products that they will be safe and effective. The FDA Nanotechnology Task Force provides guidelines concerning the existed tests and the necessity to conduct additional ones when is needed. Also, highlight some points that must be taken into consideration when nanomaterials are engaged in regulated product such as routes of exposure of nanomaterials, properties related to absorption, distribution, metabolism, and excretion (ADME) and particle size, size distribution, surface charge, aggregation etc. However, FDA does not categorically judge the nanotechnology product intrinsically benign or harmful. FDA regulates nanotechnology products under existing statutory authorities and try to enhance its scientific knowledge with the aim of evaluating the safety and the efficacy of the products by developing new regulatory pathways

grounded in the best available science. Moreover, the FDA conduct post-market monitoring in MDs but the manufacturer is fully responsible for ensuring that the product meet all applicable legal requirements, including safety standards.[52]

3.3.2 ISO 10993-1: "Biological evaluation of medical devices

FDA has developed a guidance, ISO 10993-1 in order to provide recommendations concerning medical devices that come into direct contact or indirect contact with the human body and to determine the potential for an unacceptable adverse biological response resulting from contact of the component materials of the device with the body. This guidance document also incorporates considerations, including the test considerations such as cytotoxicity, implantation, genotoxicity, carcinogenicity etc), the use of risk-based approaches to determine if biocompatibility testing is needed, chemical assessment recommendations, and recommendations for biocompatibility test article preparation for devices with submicron or nanotechnology components and for devices made from in situ polymerizing and/or absorbable materials. [53, 54]

3.3.3 Classification of medical devices by FDA

The FDA suggests 3 classes that medical devices can be categorized, Class I, Class II and Class III. The classification of MD depends on the "intended use" of the device and the upon "indications for use". The MD in classes II and III must undergo a pre-market approval by the FDA before they can be imported in US market but not for MD in class I. Specifically the three classes are:

- Class I: low risk (e.g. dental floss, tongue depressors, arm slings, and hand-held surgical instruments)
- Class II: higher-risk devices (e.g. condoms, X-ray systems, gas analyzers, pumps, and surgical drapes)
- Class III: highest-risk devices (e.g. implantable devices (devices made to replace/support or enhance part of your body) such as defibrillators, pacemakers, artificial hips, knees, and replacement heart valves)

In addition to the classification, the MD has its own product code and regulation number. It is reasonable for class I and II MD are usually not required clinical studies in contrast to class III MD.[43, 55, 56] A Class III requires a device manufacturer to submit a Premarket Approval Application (PMA) to the FDA. The requirements that should be submitted are data from preclinical or clinical testing that can ensure the safety and the effectiveness of the product. In the case of the modification to an FDA-approved Class III device that could affect safety or efficacy, such as the introduction of nanomaterials, would require FDA approval of a PMA supplement. On the contrary, manufacturers of Class I or Class II medical devices can make modifications to FDA-approved products without being required to submit.[57]

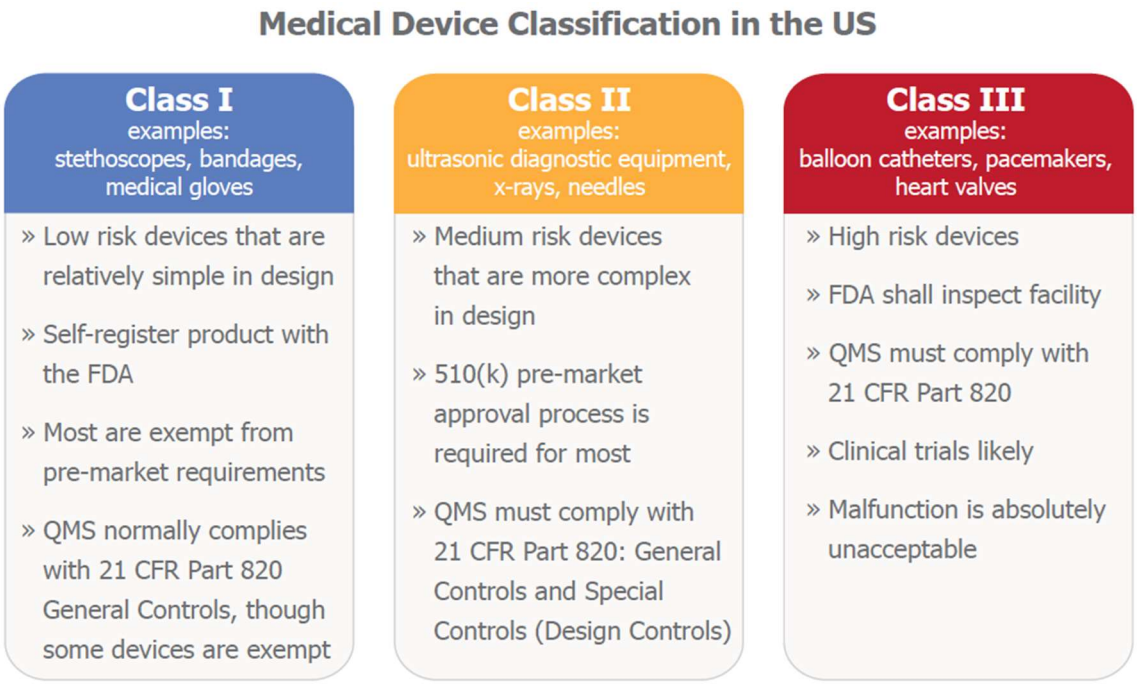

Figure 14. US Medical Device Classification.[58]

3.4 Regulations in China

The China Food and Drug Administration (CFDA) (now NMPA) is the administrative body responsible for the regulation of medical devices in China. NMPA classifies MD with a similar way with EU and US (Table 1). Specifically, China split the MD in Class I, Class II and Class III. Likewise, with EU and US, Class I devices are low risk devices that can be effectively

monitored through regular administration, while Class III devices are complex implants or life supporting devices with high risk.[59] The CFDA has announced a two-year plan to develop medical device standards covering risk management, quality control and clinical trials as well standards for active and non-active medical devices and IVD products.[60]

In respect of using nanomaterials in MD, China seems to keep up the nanotechnology challenges slowly. In comparison to EU and USA, China doesn't have a thorough regulation framework for MD that contain NPs. They have been made an effort on development of nanotechnology in China such as the National Steering Committee for Nanoscience and Nanotechnology (NSCNN) in 2000. Since then, the Chinese government has comprehended that should attend to the nanotechnology progress. Organizations and Universities in China are carrying out research studies concerning the toxicological and environmental effects of nanomaterials. In 2009, China established the 'China REACH' that is similar to the EU's REACH regulatory framework in order to manage the nanotechnology chemicals.[55]

In conclusion, China keeps it eyes in the EU and FDA regulatory approach concerning the nanotechnology in products. However, it is far apart from the enacting of effective regulatory framework able to manage nanotechnology-based-risks.[55]

Table 3 Comparative risk classification of medical devices. [43]

Country		Description	Examples	Risk
EU	**FDA/NMPA**			
Class I	Class I	1. Noninvasive, external 2. 2. Invasive, transient or used in oral and nasal cavities or ear canal	Bandages, tongue depressor, surgical retractor	**Low risk**
Class IIa	Class II	1. Noninvasive wound healing or store/channel fluids 2. Invasive in body orifice/stoma (not surgical), short-term use	Hypodermic needles, suction equipment	
Class IIb		1. Noninvasive, modify blood/body fluids/liquids for infusion 2. Invasive in body orifice/stoma (not surgical), long-term use 3. Invasive (surgical short term) administers medicinal products or radiation, undergoes chemical exchange 4. Reproductive devices	Lung ventilator, bone fixation plate	
Class III	Class III	Surgically invasive: 1. Any contact/activity cardiovascular or central nervous system 2. Breast implants 3. Short-term biological effect or absorbed 4. Long-term life supporting/ sustaining 5. Long-term administers medicinal products or products with ancillary action or undergoes chemical exchange 6. Use of human or animal cells, tissues, or derivatives	Heart valves, defibrillator	**High risk**

3.5 Future perspectives

The principle of MDs is that should provide improved treatment to the benefit of patients without causing undesirable effects. The most important issues concerning regulations for nano-enabled medical devices include their definition, classification, premarket approval procedures, good manufacturing practices, labeling, and post market surveillance. The main challenges are the use of nanobiomaterials, risk assessment, and risk management due to the novel properties of these substances. There are several issues concerning to safety testing of MDs such as lack of universally accepted assessments to establish the nature of the released materials as well as the methodology to define their distribution (toxicokinetic studies). Furthermore, the absence of appropriate in vitro models for identification of the hazard of EBMs considering the routes of exposure and duration of the contact (acute and chronic toxicity) are some challenges that need to be overcome. Issues related to the environmental risk assessment should be also taken into consideration. Thus, it is necessary the development of methodology for characterizing intrinsic physical and chemical properties of engineered EBMs as well as their system-dependent properties (i.e. interactions, biopersistence, biotransformation and behavior) in relation to human health and the environment as well as dosimetry. The using of in silico models and methodologies can contribute to the better prediction and assessment of the human and environmental risks.[41]

Due to the complexity of MDs, establishing a regulatory framework for their evaluation is a complicated task.[42] The new regulatory EU framework for medical devices targets to address existing gaps and seems to be more robust for monitoring the technical evolution of some devices. The most significant alteration in the new medical device regulation is represented by the new data management. Lack of data and the access in clinical data are the main issues for gaining intimate recommendations on the use of medical devices and their post market analyses. Furthermore, the regulation recommends the reduction and thorough evaluation of class III MDs. All these suggestions could probably delay market entrance and reduce innovation and investments because the manufacturers don't have experience, skills and resources to satisfy the new requirements. However, changes have

been introduced to address real problems, gaps, and critical issues mainly related to safety.[61]

The FDA approach on MDs regulation and other nanotechnology products has been challenged as lacking specificity. The primary concern of FDA is to collect more data and to establish valid testing criteria. Currently, there is no existing regulatory framework, but several draft guidance documents have been published.[56] China, has not established a legal framework for nano-medical devices and nano-products yet. So, it doesn't have an effective scheme to assess and manage the risks and the safety of these products. Chinese authorities are trying to track and harmonize with EU and FDA recommendations about nanoproducts regulation. In conclusion, the ideal outcome for MDs regulation is a balance between "underregulation," which could have inappropriate and possibly harmful impact, and "overregulation", which could limit innovation.[56]

4 Food supplements

4.1 Food supplements and nanotechnology

Food supplements are concentrated sources of nutrients (i.e. mineral and vitamins) or other substances with a nutritional or physiological effect that are marketed in "dose" form e.g. pills, tablets, capsules, liquids in measured doses. They may include a wide range of nutrients and other ingredients such as vitamins, minerals, amino acids, essential fatty acids, fibre and various plants and herbal extracts. Dietary supplements are not medicinal products, thus, they cannot exert a pharmacological, immunological or metabolic action. They are intended to correct nutritional deficiencies, maintain an adequate intake of certain nutrients, or to support specific physiological functions but not to treat or prevent diseases in humans or to modify physiological functions.[62]

Nanotechnology has introduced new properties in food supplements. Nanoformulations of supplements are being prepared especially with the purpose to improve bioavailability, protect active ingredients against degradation, or reduce side effects. A wide range of nanoscale carriers are used in order to improve the functionality of nutrients or other substances containing in food supplements (Figure 16). Nanocarriers are intended to improve the stability and bioavailability as well as to protect valuable nutraceuticals at food processing or digestion. Several studies focus on the formulation design for the enhancement of nutraceuticals' bioavailability with the purpose to ensure effective preservation or maximization of their bioactivity and safety.[63] The enhancement of bioavailability is achieved through various ways such as by the improved solubility of bioactive compounds under gastrointestinal (GI) conditions, their protection from the chemical conditions in the GI tract, and controlled release within the GI tract, or by an improved transfer through the intestinal wall. The particle size, surface properties, and physical state of the nanomaterials used in food supplements are crucial characteristics affecting their final nutritional value.[64]

For instance, nanoliposomes are widely used in dietary supplements because they can encapsulate simultaneously lipophilic and hydrophilic materials. As a result, sensitive bioactive compounds are protected, enhancing their bioavailability. Thus, nanoscale liposomes ensure the sustained-release and improved storage stability.[65] Likewise,

nanocapsules based on lipid formulations having larger surface area than micron-sized carriers can enhance solubility, bioavailability, and controlled release of the encapsulated compounds (e.g proteins) with the potential to successfully be applied in functional foods.[66] Also, inorganic porous materials, such as various silica- or aluminosilicate-based materials/composites, clays, etc., have been used for the delivery of drugs, providing advantages in formulation and engineering. They have suitable architecture, large surface area, and stability in biological fluids; thus, they are used for high loading capacity, controllable release, and improved targeting.

The aim of dietary supplements is to maintain the human health by delivering necessary compounds that cannot be received sufficiently through the regular diet. Food supplements are regulated, usually, by the European Food Safety Authority and the U.S. Food and Drug Administration, as well as by various national regulations issued most frequently by the Ministry of Health and/or the Ministry of Agriculture of particular countries around the world with the aim to protect consumers against potential health risks and to guarantee that they are not given misdirecting information.[67]

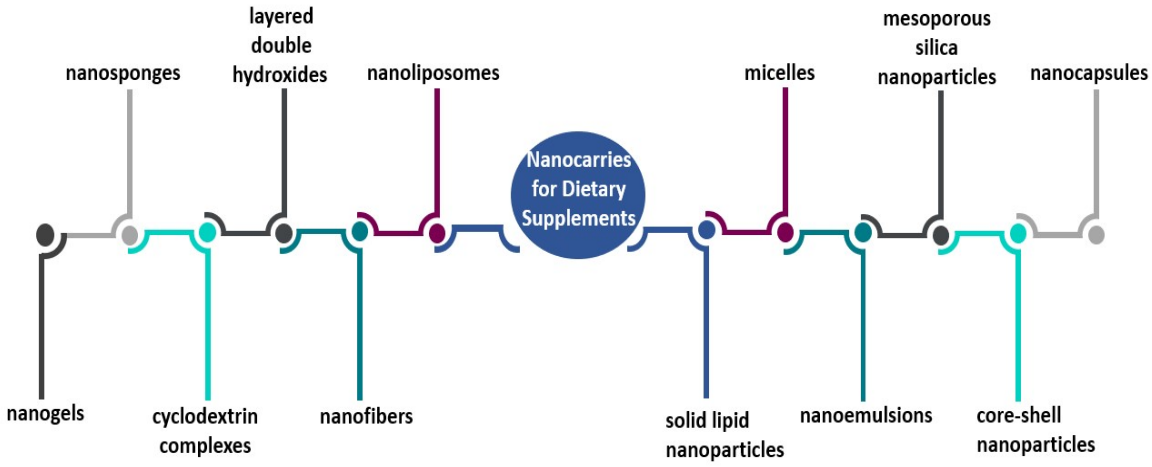

Figure 15. Most frequently used nanoformulation types of dietary supplements.[66]

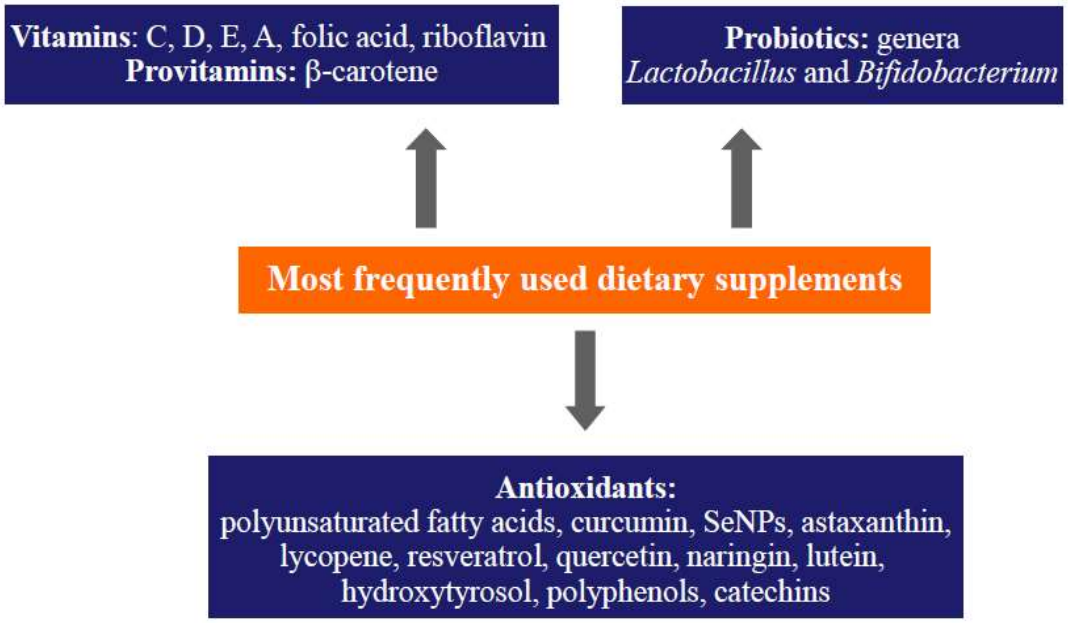

Figure 16. Most frequently used human dietary. [68]

4.2 Regulation in EU

4.2 EU Framework

The EU General Food Law Regulation (EC) No 178/2002 provides laws and recommendations concerning food with the aim to ensure a high-level protection of human health and consumer interest, taking into account the variety of substances in food products while ensuring the effective functioning of the EU market.[69] The major legislation concerning food supplements is Directive 2002/46/EC that contain harmonized lists of the vitamins and mineral substances used in the manufacture of food supplements and the labelling requirements for these products. Specifically, the regulation in annex I provides a list of vitamins and minerals that may be applied for nutritional purposes in food supplements while annex II presents a list of permitted sources (vitamin and mineral substances) may also be the manufacturing of those vitamins and minerals.[70] Regarding labelling, food supplements should meet requirements (Article 6) such as: **(i)** the analytical reference names of the nutrients or substances that characterize the product **(ii)** the recommended dose for daily consumption **(iii)** a warning not to exceed the stated recommended daily dose; **(iv)** a declaration to the effect that the supplement is not a

substitute for a varied diet and **(v)** a statement to the effect that food supplements should not be used as a substitute for a varied diet. The use of substances other than vitamins or minerals in the manufacture of food supplements are subject to Regulation (EC) No 2015/2283 on novel food, including the case of nanomaterials. In the EU, there are several regulations on food supplements, for example, sources of vitamins, minerals and additional substances that may also be subject to Regulation (EC) No 1925/2006 on fortification of foods or Regulation (EC) No 609/2013 on foods for specific groups. The substances added to food supplements to perform certain technological functions, for example to add color, sweeten or preserve, that are assessed as food additives under Regulation (EC) No 1333/2008.[69] Finally, dietary supplements are regulated by the European Food Safety Authority (EFSA) whose role is described in the next section. In the EU market, there are nutrition claims which are statements about a relationship between food and health. A product should meet these claims in order to be marketed. The EFSA is responsible for evaluating the scientific evidence supporting health claims, the types of which are as following: **(i)** 'Function Health Claims' (relating to growth, development and functions of the body, or referring to psychological and behavioral functions, or on slimming or weight-control), **(ii)** 'Risk Reduction Claims' (on reducing a risk factor in the development of a disease), and **(iii)** Health Claims referring to children's development.[68]

4.2.1 EFSA (European Food Safety Authority)

EFSA is an independent source of scientific advice that contributes to the adoption of legislation by European Commission. EFSA has a crucial role in supporting policymakers at European and national levels in developing policies and setting diet-related public health targets. EFSA has been conducted a comprehensive assessment of permitted substances that are used as sources of vitamins and minerals in food supplements in the EU. The assessments include the evaluation of the safety of a nutrient source at the intake levels suggested by the applicant, and the bioavailability of the nutrient from the source i.e. the effectiveness with which the mineral or vitamin is released into the body. Concerning the products that contain a nutrient source not included in the permitted list, companies have to submit an application to the European Commission prior to market entrance. Under Directive 2002/46/EC, EFSA then prepares a scientific opinion to support the European

Commission's evaluation of the request. Based on EFSA's work, the European Commission reviews and updates the list of vitamin or mineral substances that may be used in food supplements.

If a substance is intended to be used in food without having a prior history of safe use in the EU before 1997, EFSA is requested to provide a scientific opinion on its safety according to Regulation (EC) No 2015/2283 on novel foods including newly developed, innovative food, and food produced using new technologies and production processes. According to this regulation, dossiers need to provide data to the EC concerning the compositional, nutritional, toxicological and allergenic properties of the novel food as well as information on respective production processes, and the intended uses and use levels. Under the Novel Foods Regulation, food consisting of engineered nanomaterials is novel food. Thus, supplements containing or consisting of nanomaterials are considered to be novel food and require pre authorization according to Novel Foods Regulation in order to ensure a high level of protection of human health. EFSA has released a guidance regarding risk assessment of the application of nanoscience and nanotechnologies in the food and feed chain. In particular, this guidance defines nanomaterials as the materials that have particle sizes in the nanoscale (1–100 nm) or materials that contain particles having a size above 100 nm which present properties that are characteristic of the nanoscale (for example large specific area and different toxicokinetic behavior), as well as materials that are not engineered as nanomaterial but contain a fraction of particles more than 50% in the number–size distribution (as per the recommended European Commission definition), with one or more external dimensions in the size range 1–100 nm. However, the definition for nanomaterial according to EC is still under review and the EFSA recommends taking future reviews into account during the safety evaluation of nanomaterials. [71]

4.2.2 Risk evaluation of nanomaterials

According to EFSA, the risk assessment of nanomaterials has four main pillars: the chemical analysis of NPs composition, their physicochemical properties, their interactions with tissues and potential exposure levels. The first step of nanomaterials safety assessment is the identification of nanomaterials. Afterwards, all dossiers related to nanomaterial

characterization have to be accompanied by thorough information on the particle size distribution and other parameters based on techniques that described in Appendix C of the guidance. The next step is the characterization of nanomaterials in relevant food/feed matrix as well as the test media used in in vitro and in vivo testing. Further testing may be required, and a tiered framework is described. The guidance outlines a scheme in order to identify the hazard effect of nanomaterials. The first phase is the investigation of the rate of degradation of the nanomaterial to no nanomaterial form under conditions representative of the gastrointestinal tract. In the second phase, all in vitro studies should be conducted for identification of hazards. In case further studies are needed, they are carried out for identification of the nanomaterials with potential to cause immunological, proliferative, neurotoxic, reproductive organ or endocrine-mediated effects as appropriate (90-day toxicity test). The results of this phase will determine if further comprehensive studies are needed, such as human kinetic data from volunteer studies, additional toxicokinetic study, reproductive and developmental toxicity, additional immunotoxicity, neurotoxicity, carcinogenicity/mutagenicity, endocrine effects, gut microbiome. It is crucial that applicants utilize a comprehensive approach to cover the risk assessment of the fraction in the nanoscale, including the application of recommendations and test strategies as were described in the EFSA Guidance (EFSA, 2018).The schematic general outline for risk assessment of nanomaterials can be summarized in Figure 18.[72, 73]

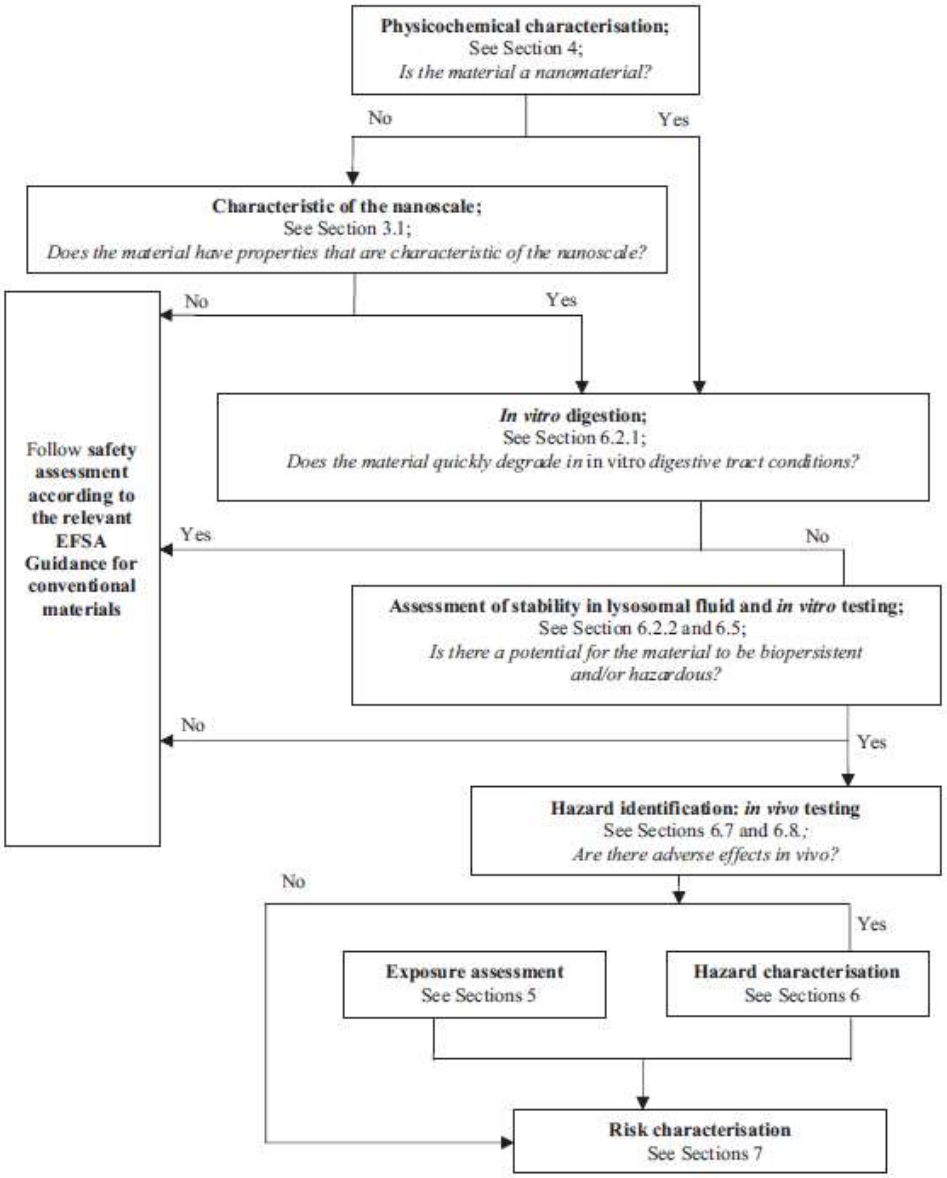

Figure 17. Schematic outline for risk assessment of ingested11 nanomaterials for human and animal health, focussing on hazard characterisation.[72]

4.3 Regulatory in US

4.3.1 US Framework

In the USA, food supplements are regulated by the US FDA under the Federal Food, Drug and Cosmetic Act (FD&C Act). They are regulated differently from other foods and drugs and their classification as a dietary supplement, conventional food, or drug depends on the

intended use. The classification as dietary supplements is determined by the information and data that the manufacturer provides on the product label. However, there are several food supplement product labels that do not include this information. In addition, the Dietary Supplement Health and Education Act of 1994 (DSHEA) together with the FD&C Act formed a new legislation framework for dietary supplements. DSHEA defined dietary supplements as products that contain one or more dietary ingredients (including vitamins; minerals; herbs or other botanicals; amino acids; and other substances) or their constituents and are intended to supplement the diet. Also, these products are intended to be taken by mouth as a pill, capsule, tablet, or liquid and are labeled in the front panel as dietary supplements[74].DSHEA provided recommendations concerning the addressing of safety issues, labelling and health claims upon dietary supplements. Furthermore, DSHEA contributed to the developing of a database of dietary supplements and the establishment dietary intake regulations, safety, claims, and scientific issues arising in connection with labelling and composition. DSHEA attend to the conditions under which dietary supplements are adulterated based on the existing food standards for adulteration to dietary supplements and requires the presence of instructions on product label concerning the conditions of use recommended or suggested by manufacturers. In addition, DSHEA also noted that the food supplements which don't follow good manufacturing practices during the production process are considered adulterated.[75] Specifically. FDA has provided Good Manufacturing Practices (GMPs) for dietary supplements concerning manufacturing, preparation and storage of products in order to ensure the best quality of product. For instance, GMPs aim to prevent the presence of the wrong ingredients, the addition of too much or too little of a dietary ingredient, the possibility of contamination (by pesticides, heavy metals, bacteria, etc.), and the improper packaging and labeling of a product. [61]

The label of a food supplement may include one of following types of claims or disclaimer: a health claim, nutrient content claim, or structure/function claim in order to describe the relationship between a food, food component, or dietary supplement ingredient, and reducing risk of a disease or health-related condition. A structure/function claim is a statement describing how a product may affect the organs or systems of the body and may not mention any specific disease. Structure/function claims do not require FDA approval, but the manufacturer must provide FDA with the text of the claim within 30 days of importing the product on the market disclaimer. Product labels containing such claims must

also include a disclaimer that reads. "This statement has not been evaluated by the FDA. This product is not intended to diagnose, treat, cure, or prevent any disease." Moreover, additional information may be placed in product label, such as general information of the product (name, net quantity of contents, name and place of manufacturer, packer, or distributor, directions for use, and supplement facts panel) as well as serving size, list of dietary ingredients, amount per serving size (by weight), percent of Daily Value (%DV). Also, the label should include the presence of nondietary ingredients such as fillers, artificial colors, sweeteners, flavors, or binders. [61]

In the case of using a new dietary substance in products, which was not sold as a dietary supplement before 1994, the manufacturer must inform FDA of their intent to market it, as a dietary supplement containing the new dietary ingredient and provide reasonable evidence for safe human use of the product. Supplement ingredients sold in the United States before October 15, 1994, are not required to be reviewed by FDA for their safety before they are marketed, because they are presumed to be safe based on their history of use by humans. In contrast to medicinal, there is no legal framework by FDA to for the approval of dietary supplements for safety or effectiveness before market entrance. When a product enters the market, FDA should prove that the product is not safe in order to restrict its use or remove it from the market. Before its market placing, manufacturers must obtain FDA approval by providing convincing evidence that it is both safe and effective.[61]

Safety and effectiveness assessment of food supplement ingredients are essential for ensuring of heath safety. Until now, dietary supplements are not required by federal law to be tested before they are marketed, so there is a lack of data concerning these substances. There is historical background for some ingredients but not for all. Scientists can use several approaches to evaluate dietary supplements for their potential health benefits and risks. In particular, they may investigate history of use, conduct laboratory studies using cell or tissue cultures, and experiment with animals. Studies on people (e.g., individual case reports, observational studies, and clinical trials) provide the most direct evidence of a dietary supplement's effects on health and patterns of use.

4.3.2 Nanotechnology in food supplements: US regulatory

As mentioned, in order to address safety and regulatory issues by the ever increasing use of nanotechnology in foods, supplements, and cosmetics, the US FDA developed a dedicated task force on nanotechnology (2007). In April 2012, the FDA released a draft guidance focusing on nanotechnology in foods. It is noteworthy that there is no document that addresses the use of nanotechnology in dietary supplements. Since they are considered to belong under the category of foods by FDA, the draft guidance on foods directly impacts the dietary supplements industry. The guidance attends to the modification in the physical and chemical properties of a food substance that can affect its bioavailability through altered absorption, distribution, metabolism and excretion. In addition, they acknowledge that such changes in the substance's biological interactions can affect the level at which toxic effects may occur. FDA considers that the altered chemical properties of the ingredients are essentially a new dietary ingredient and thus can require a notification to the FDA.[76]

Regarding the labeling of new ingredients, there are no specific requirements for labeling products containing nanomaterials and there is no requirement for nanomaterials to be labeled as such. These ingredients are addressed like any other new dietary ingredient and their safety evaluation is not required for pre-market approval. Hence, the lack of labeling guidelines means that consumers may be misleading and sometimes to use products without knowing the presence of nanoscale ingredients. Moreover, there are cases that some product labels imply that they include nano-sized materials that the products do not in fact contain. So, the doubts about the labelling of these products as well as the absence of a legal framework make it difficult for the consumer to determine the true nature of their ingredients.

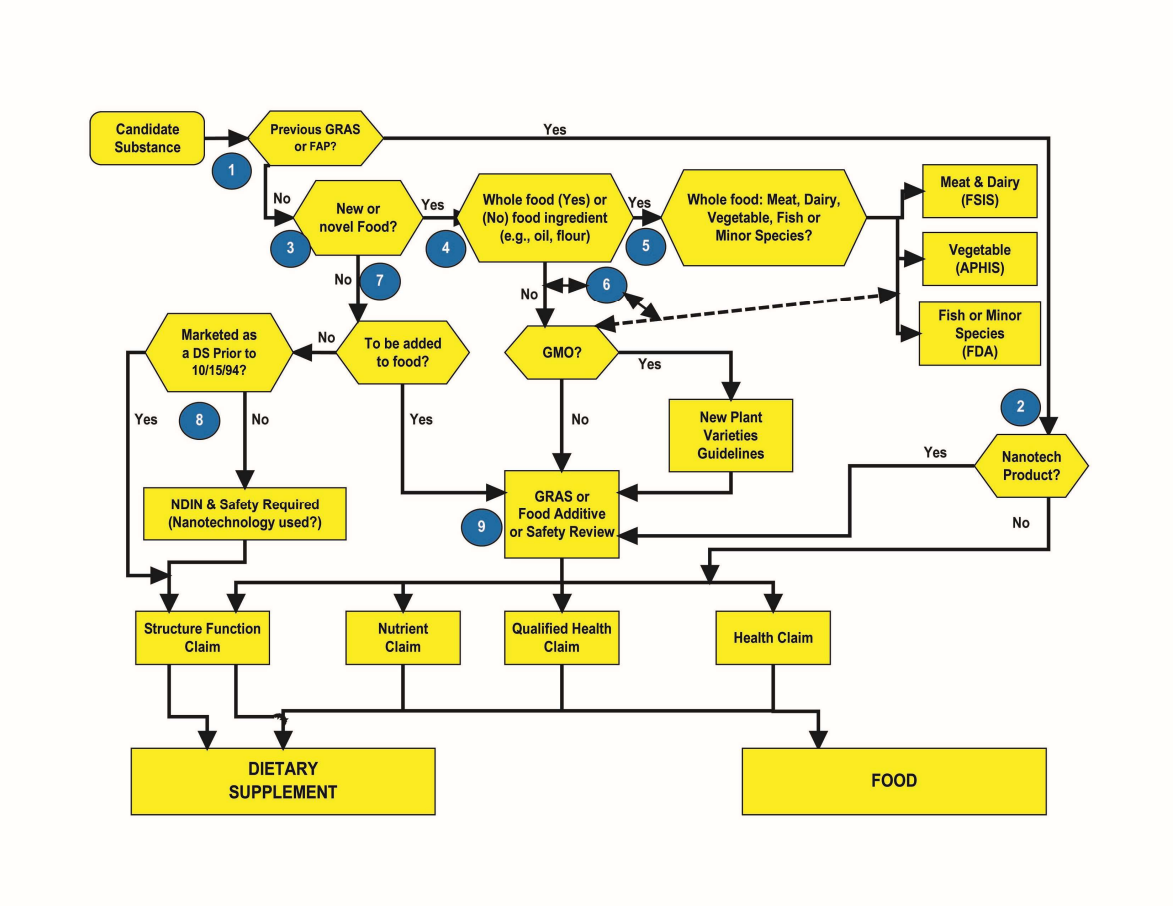

Figure 18. A multi-step process that includes a series of significant questions for innovators and marketers.[77]

4.4 Regulations in China

In China, food supplements are termed healthy food and are considered as food products that have specific health functions or supply vitamins and (or) minerals. However, they are not used for the purpose of curing diseases and cause no acute, sub-acute or chronic health effect to human body. The Food Safety Law of the People's Republic of China (2015 version) is regulating companies that wish to enter products in the marketplace. Specifically, the companies should apply and obtain the health food registration certificate or filing certificate. In the case of domestic health foods produced in China, the registration shall be conducted with CFDA, whereas, the filing shall be carried out with Provincial Food and Drug Administration (FDA). In contrast, the imported health foods produced in oversea factories should apply both the registration and filing with the CFDA. Companies overseas shall have a Chinese representative office or appoint a Chinese agent to deal with registration or filing

and obtain such certificates. Health food registration and filling shall be carried out according to Administrative Measure on Health Food Registration and Filing (herein named the Measure). According to the Measure, several documents are required for health food filing such as product formulation materials, information about production process and product technical requirements. Also, the safety and function assessment material are very important, which refers to reports of functional components/characteristic ingredients, stability, hygiene health and others if necessary, etc. Similar documents are required for health food registration. According to the Measure, tests must be conducted in order to ensure safety and the effectiveness. In particular, safety and toxicology tests, animal and (or) human function tests, functional components/characteristic ingredients tests, hygiene health test, stability test and others are required.[78]

While some major countries and regions including China have not yet formulated any specific legislation on the regulation of healthy food with nanoscale materials, in general it is subject to the same public health and food safety regulations that apply to other food types. According to the World Health Organization, the potential health and environmental risks of nanoscale materials need to be assessed before they are introduced into food. It is stipulated in the Public Health and Municipal Services Ordinance that all be fit for human consumption. This principle also applies to nanotechnology-derived food products and the food trade have the responsibility to ensure the safety of engineered nanomaterials in their food products if they are to supply such products. While consumers are likely to benefit from the technology, new data and measurement approaches are needed to ensure safety of products using nanotechnology. [79]

4.5 Future perspectives

Use of nanotechnology in food supplements is growing and novel products are being developed to exploit the advances in this technology.[80] Nanomaterials enhance the bioavailability and increase the stability of individual active ingredients of food supplements but all nanoscale materials applied in these products should be used with careful consideration and only after thorough investigation of cytotoxicity due to possible increased nanosize-based toxicity effects that could cause undesirable effects in human

health.[66] Regulation of using nanomaterials in food supplements is currently incomplete in all countries. As mentioned, the EU has issued some recommendations for nanomaterials in food. The challenges in regulating nanomaterials remain, because of knowledge gaps in traditional risk assessments protocols. Current definitions for nanomaterials might fail or be difficult to address all potential risks. The altered physicochemical characteristics of nanomaterials and their interactions with environmental factors affect both evaluations of exposure and hazard for nanomaterials. Registration and labelling of these products by manufacturers is crucial for the evaluation of NP exposure ,their composition, physiochemical characteristics and production volume.[81] It is apparent that there is a need for a solid regulatory framework that addresses and specifically manages the potential risks of nanotechnology in food supplements and other products that include nanomaterials. Hence, it is important for human health safety to establish a regulatory framework for managing nanomaterials and its applications, facilitating the safe application and commercialization of nanotechnology products. [79]

5 Conclusion

Nanomaterials have unique physicochemical and biological properties compared to their larger counterparts. Because of their small size, shape, surface, charge, and agglomeration behavior, the properties of nanomaterials may significantly impact their interactions with biomolecules. Thus, nano-enabled products are developed, such as nanocosmetics with enhanced stability, shelf-life and UV resistance, food supplements with better bioavailability and advanced medical devices. Despite the fact that advances in nanotechnology are offering new opportunities, there are still numerous challenges and chances for improvement. The validation product safety and environmental impact that arise from the utilization of nanotechnology is a major challenge moving forward. So, testing of nanosystems should be required before they enter the market.[82]

In general, thorough studies on the safety profile of the nanomaterials are required. The industries and manufactures should fabricate the nanosystems in such a way that their value and health of the customers are improved. Furthermore, they should provide data to prove that their product is safe and effective. There are substantial doubts concerning the toxicity and safety of the nanomaterials so researches from various sciences try to determine the possible health hazard and toxicity. Lastly, stringent regulatory framework should be imposed for products that are based on nanotechnology in order to ensure the safety of products.[13] However, the regulatory challenges presented by nanomaterials and nanotechnology are many: the diversity of nanomaterials, lack of characterization data, and lack of standardization in nomenclature and metrics. Manufacturers developing nano-enabled products should work with current data in the production process and monitor products once marketed. The industry can consult the regulation authorities which may afford an opportunity to clarify the methodologies and data that will be needed to meet obligations. Regions that don’t have clear recommendations and laws for nanoproducts will unavoidably follow the EU and US regulatory approaches on nanotechnology. Flexible, product and science-based approaches are being used to facilitate innovation and to fulfil the regulation 's mission to bring safe and effective medical products to the market, while safeguarding public health.[55]

6 References

[1]T. Singh, S. Shukla, P. Kumar, V. Wahla, V.K. Bajpai and I.A. Rather, "Application of Nanotechnology in Food Science: Perception and Overview" *frontiers in Microbiology*, no.8 (2017):1-7, doi: 10.3389/fmicb.2017.01501

[2]Demetzos Costas, *Pharmaceutical Nanotechnology: Fundamentals and Practical Applications* (Adis, 2016)

[3]S. Logothetidis, "Nanotechnology: Principles and Applications" Part of the NanoScience and Technology book (Springer, 2011): 1-22

[4]J. Behari , "Principles of nanoscience: an overview" *Indian Journal of Experimental Biology,* no.48 (2010): 1008-19

[5]https://gaeu.com/artiklar/this-is-nanotechnology-one-of-the-fastest-growing-markets-in-the-world/

[6]https://statnano.com/nanomaterials

[7]https://www.nowfoods.com/now/nowledge/nanotechnology-nutritional-supplements-scientific-and-regulatory-review-and-update

[8] V. Kumar, N. Dasgupta, S. Ranjan, *Nanotoxicology: Toxicity Evaluation, Risk Assessment and Management* (CRC Press, Taylor & Francis Group, 2018)

[9]Monica, J. C., Van Calster, G., & Patsa, A. "A Nanotechnology Legal Framework. Nanotechnology Environmental Health and Safety" (2014):265–311, doi:10.1016/b978-1-4557-3188-6.00012-8

[10]D.R. Boverhof, C.M. Bramante, J.H. Butala, S.F. Clancy, M. Lafranconi, J. West, S.C. Gordong, "Comparative assessment of nanomaterial definitions and safety evaluation considerations", Regulatory Toxicology and Pharmacology, No. 73, (2015):137-150, doi: 10.1016/j.yrtph.2015.06.001

[11] https://smartech.gatech.edu/handle/1853/59500

[12]https://www.semanticscholar.org/paper/How-can-regulation-shape-the-direction-of-Zwanenberg-R%C3%A0fols/bf166ad51e5eecf689b8c25059936e83dcd03a38/figure/0

[13]https://www.mordorintelligence.com/industry-reports/global-cosmetics-products-market-industry

[14] S. Kaul, N. Gulati, D. Verma, S. Mukherjee, and U. Nagaich, "Role of Nanotechnology in Cosmeceuticals: A Review of Recent Advances" *Journal of Pharmaceutics* (2018):1-20, doi: 10.1155/2018/3420204

[15] YoonMi Park, Young & Global Partners, *Presentation:* Nano-regulation in China, Japan, Korea, USA and EU

[16] S. Nafisi, H.I. Maibach, "Nanotechnology in Cosmetics" Part of the Cosmetic Science and Technology: Theoretical Principles and Applications (Elsevier Inc, 2017): 337-368, doi: 10.1016/B978-0-12-802005-0.00022-7

[17] S. Raj, J. Shoma, U.S Sumod., M. Sabitha, "Nanotechnology in cosmetics: Opportunities and challenges" *Journal of Pharmacy and Bioallied Sciences,* no.4 (2012): 186-193, doi: 10.4103/0975-7406.99016

[18] Clare Shelley-Egan and Diana Megan Bowman, "The Challenge of Distributing Regulatory Responsibilities for Unknown Risks: Nano-Cosmetics and the EU Cosmetics Regulation as a Case Study", Clinical Research & Bioethics, J Clinic Res Bioeth, No. 6, (2015): 1- 10, doi: 10.4172/2155-9627.1000212

[19] M. Ajazzuddin, G. Jeswani and A. Kumar Jha, "Nanocosmetics: Past, Present and Future Trends" *Recent Patents on Nanomedicine,* no.5(2015): 3-11

[20] D.M. Bowman, N.D. May, A.D. Maynard, "Nanomaterials in Cosmetics: Regulatory Aspects" Part of the Analysis of Cosmetic Products (Elsevier B.V, 2018): 289-302, doi: 10.1016/B978-0-444-63508-2.00012-6

[21] REGULATION (EC) No 1223/2009 OF THE EUROPEAN PARLIAMENT AND OF THE COUNCIL of 30 November 2009 on cosmetic products

[22] http://ec.europa.eu/environment/chemicals/nanotech/reach-clp/index_en.htm

[23] H. Rauscher, K. Rasmussen and B. Sokull-Kluetgen, "Regulatory Aspects of Nanomaterials in the EU" Chemie Ingenieur Technik, no.3 (2017): 224–231, doi: DOI: 10.1002/cite.201600076

[24] REGULATION (EC) No 1907/2006 OF THE EUROPEAN PARLIAMENT AND OF THE COUNCIL of 18 December 2006 concerning the Registration, Evaluation, Authorisation and Restriction of Chemicals (REACH), establishing a European Chemicals Agency

[25] Ngô, C., & Van de Voorde, M. H. "Nanomaterials and Cosmetics" Part of Nanotechnology in a Nutshell, (Atlantis Press, 2014): 311–319, doi:10.2991/978-94-6239-012-6_18

[26] A. Lohani, A. Verma, H. Joshi, N. Yadav, and N. Karki, "Nanotechnology-Based Cosmeceuticals,"*ISRN Dermatology*" (2014):1-14, doi: 10.1155/2014/843687

[27]S. Nanda, A. Nanda, S. Lohan, R. Kaur, and B. Singh, "Nanocosmetics: Performance enhancement and safety assurance" Part of Nanobiomaterials in Galenic Formulations and Cosmetics (Elsevier Inc., 2016): 47-67, doi: 10.1016/B978-0-323-42868-2.00003-6

[28]S. W. Bennett, D. Zhou, R. Mielke, and A. A. Keller, "Photoinduced Disaggregation of TiO 2 Nanoparticles Enables Transdermal Penetration," (2012) vol. 7, no. 11: 1-7, doi: 10.1371/journal.pone.0048719

[29]J.S Tsuji, A. Maynard, P.C. Howard, J.T. James, C. W. Lam, D.B. Warheit, A.B. Santamaria, "Research Strategies for Safety Evaluation of Nanomaterials , Part IV : Risk Assessment of Nanoparticles," (2006) vol. 89, no. 1, pp. 42–50, doi: 10.1093/toxsci/kfi339

[30]J.S Tsuji, A. Maynard, P.C. Howard, J.T. James, C. W. Lam, D.B. Warheit, A.B. Santamaria, "Research Strategies for Safety Evaluation of Nanomaterials , Part IV : Risk Assessment of Nanoparticles," (2006) vol. 89, no. 1, pp. 42–50, doi: 10.1093/toxsci/kfi339

[31]V. K. M. Poon and A. Burd, "In vitro cytotoxity of silver : implication for clinical wound care," (2004) vol. 30, pp. 140–147, doi: 10.1016/j.burns.2003.09.030

[32]THE SCCS NOTES OF GUIDANCE FOR THE TESTING OF COSMETIC INGREDIENTS AND THEIR SAFETY EVALUATION (10TH REVISION)

[33]M. Kumud and N. Sanju, "Nanotechnology Driven Cosmetic Products: Commercial and Regulatory Milestones" *Bentham Science,* no.5, (2018): 112-121, doi: 10.2174/2213476X05666180530093111

[34] Guidance for Industry Safety of Nanomaterials in Cosmetic Products, U.S. Department of Health and Human Services Food and Drug Administration Center for Food Safety and Applied Nutrition (June 2014)

[35]G. Oberdörster , E. Oberdörster , and J. Oberdörster, "Nanotoxicology: An Emerging Discipline Evolving from Studies of Ultrafine Particles", No.7 (2005): doi: 10.1289/ehp.7339

[36]https://psi.ul.com/en/resources/article/china-compliance-cosmetic-products/

[37]https://www.cosmeticsdesign-asia.com/Article/2018/02/20/China-makes-strides-in-improving-safety-evaluation-and-cosmetics-technical-regulations

[38]A. Meloa, M. Seragiotto Amadeua, M. Lancellottia, Luciana Maria de Hollandaa and D. Machadoa, "THE ROLE OF NANOMATERIALS IN COSMETICS: NATIONAL AND INTERNATIONAL LEGISLATIVE ASPECTS", Quim. Nova, No. 4, (2015): 599-603, doi: 10.5935/0100-4042.20150042

[39]A. C. Santos, F. Morais, A. Simões, I. Pereira, J.A.D.Sequeira, M. Pereira-Silva, Francisco Veiga & A. Ribeiro, "Nanotechnology for the development of new cosmetic formulations", *Expert Opinion on Drug Delivery,* (2019) doi: 10.1080/17425247.2019.1585426

[40]Kai Savolainen et al. Nanosafety in Europe 2015-2025: Towards Safe and Sustainable Nanomaterials and Nanotechnology Innovations (2013)

[41]F. Geremia, "Quality Aspects for Medical Devices, Quality System and Certification Process", *Microchemical Journal*, (2016): 1-13, doi: 10.1016/j.microc.2017.04.018

[42]K. Jagiełło , A. Sosnowska , A. Mikołajczyk , T. Puzyn "Nanomaterials in Medical Devices: Regulations' Review and Future Perspectives", *Journal of Nanotoxicology and Nanomedicine,* No. 2(2017): 1-11, doi: 10.4018/JNN.2017070101

[43]E. Wu, C. Chan and Albert T.W. Li, "On the evaluation of the safety aspects of nanomaterials in medical devices – a regulatory perspective", *ADMET & DMPK,* No. 4 (2013):76-81; doi: 10.5599/admet.1.4.19

[44]JoAnn C. L. Schuh and K. A. Funk, "Compilation of International Standards and Regulatory Guidance Documents for Evaluation of Biomaterials, Medical Devices, and 3-D Printed and Regenerative Medicine Products", *SAGE*, doi: 10.1177/0192623318804121

[45]https://ec.europa.eu/growth/sectors/medical-devices/new-regulations_en

[46]I.Mahapatra, J. R. A. Clark, P.J. Dobson, R. Owen, I. Lynch and J. R. Lead, "Expert perspectives on potential environmental risks from nanomedicines and adequacy of the current guideline on environmental risk assessment", Environ. Sci. Nano, No.5, (2018)1873-1889¨, doi: 10.1039/C8EN00053K

[47]https://ec.europa.eu/health/scientific_committees/consultations/public_consultations/scenihr_consultation_22_en

[48]https://ec.europa.eu/growth/sectors/medical-devices/new-regulations_en

[49]Scientific Committee on Emerging and Newly Identified Health Risks (SCENIHR), Opinion on the Guidance on the Determination of Potential Health Effects of Nanomaterials Used in Medical Devices

[50]P. Aguar, "The novel EU legislative framework for medical technology", Presentation, The European Commission's science and knowledge service, Joint Research Centre

[51]https://www.emergobyul.com/resources/europe-medical-devices-regulation-mdr-ce-marking-regulatory-process

[52]https://www.fda.gov/science-research/nanotechnology-programs-fda/fdas-approach-regulation-nanotechnology-products

[53]https://www.fda.gov/regulatory-information/search-fda-guidance-documents/use-international-standard-iso-10993-1-biological-evaluation-medical-devices-part-1-evaluation

[54] https://www.iso.org/standard/68936.html

[55]A-Andrew D. Jones, G. Mi and T.J. Webster, "A Status Report on FDA Approval of Medical Devices Containing Nanostructured Materials", Trends in Biotechnology (2018): 1-4

[56]B.V. Chikkaveeraiah, S. G. Malghan, and G. Kumarb, "Physicochemical Properties of Nanomaterials Relevant to Medical Products", Part of Physicochemical Properties of nanomaterials (Pan Stanford Publishing Pte, 2018)

[57]C. Lee Ventola, "The Nanomedicine Revolution Part 3: Regulatory and Safety Challenges", P&T, No.11 (2012): 631-639

[58]http://intelligentsystemssource.com/engineered-success-the-engineers-contributions-to-fda-medical-device-market-commercialization/

[59]https://www.pacificbridgemedical.com/regulation/china-medical-device-pharmaceutical-regulations/

[60]https://www.emergobyul.com/blog/2018/02/chinese-regulators-issue-near-term-plan-medical-device-standards-development

[61]A. Migliore, "On the new regulation of medical devices in Europe", *Expert Review of Medical Devices*, (2017) doi: 10.1080/17434440.2017.1407648

[62]https://www.efsa.europa.eu/en/topics/topic/food-supplements

[63] Goncalves, R.F.S.; Martins, J.T.; Duarte, C.M.M.; Vicente, A.A.; Pinheiro, A.C. “Advances in nutraceutical delivery systems: From formulation design for bioavailability enhancement to efficacy and safety evaluation”, *Trends Food Sci. Technol.* No. 78 (2018): 270–291, doi: 10.1016/j.tifs.2018.06.011

[64] Oehlke, K.; Adamiuk, M. Behsnilian, D. Graef, V. Mayer-Miebach, E. Walz, E. Greiner, R. “Potential bioavailability enhancement of bioactive compounds using food-grade engineered nanomaterials: A review of the existing evidence”, *Food Funct*, No. 5, (2014): 1341–1359, doi: 10.1039/c3fo60067j

[65] Khorasani, S. Danaei, M. Mozafari, M.R. “Nanoliposome technology for the food and nutraceutical industries”, *Trends Food Sci. Technol.*, No. 79, (2018): 106–115, doi: 10.1016/j.tifs.2018.07.009

[66] Esfanjani, A.F. Assadpour, E. Jafari, S.M. "Improving the bioavailability of phenolic compounds by loading them within lipid-based nanocarriers", *Trends Food Sci. Technol*., No. 76, (2018): 56–66, doi: 10.2147/IJN.S206350

[67] J. Jampilek, J. Kosand Katarina Kralova, "Potential of Nanomaterial Applications in Dietary Supplements and Foods for Special Medical Purposes", Nanomaterials, No.9, (2019): 1-42, doi:10.3390/nano9020296

[68] https://www.mdpi.com/2079-4991/9/2/296/pdf

[69] A. Anadón, M.R Martínez-Larrañaga, I. Ares and M. A Martínez, "Evaluation and Regulation of Food Supplements: European Perspective", *Nutraceuticals*, (2016): 895-623, doi: 10.1016/B978-0-12-802147-7.00063-2

[70]https://www.efsa.europa.eu/en/topics/topic/food-supplements

[71]https://www.efsa.europa.eu/en/topics/topic/novel-food

[72]K. Rasmussen, H. Rauscher, S. Gottardo, E. Hoekstra, R. Schoonjans, R. Peters, K. Aschberger, "Regulatory Status of Nanotechnologies in Food in the EU", Nanomaterials for Food Applications (2019): 381-407, doi: 10.1016/B978-0-12-814130-4.00013-0

[73]Guidance on risk assessment of the application of nanoscience and nanotechnologies in the food and feed chain: Part 1, human and animal health, EFSA Scientific Committee (2018)

[74]https://ods.od.nih.gov/factsheets/DietarySupplements-HealthProfessional/

[75]R.S. Pawar and E. Grundel, "Overview of regulation of dietary supplements in the USA and issues of adulteration with phenethylamines (PEAs)", Drug Test. Analysis, No. 9, (2017): 500–517, doi: 10.1002/dta.1980

[76]https://www.nowfoods.com/now/nowledge/nanotechnology-nutritional-supplements-scientific-and-regulatory-review-and-update

[77]http://burdockgroup.com/roadmap-for-claims-a-guide-to-establish-a-claim-for-food-food-ingredients-or-dietary-supplements/

[78]http://www.cirs-reach.com/news-and-articles/health-food-registration-and-filing.html

[79]https://www.cfs.gov.hk/english/multimedia/multimedia_pub/multimedia_pub_fsf_54_02.html

[80]Leo Bey Fen, Ahmad Hazri Abd Rashid, Nurul Izza Nordin, Mohd Rafie Johan, "Applications and impacts of nanomaterials in food safety and quality", *Preparation and Processing of Religious and Cultural Foods*, (2018): 131-161, doi: 10.1016/B978-0-08-101892-7.00007-9

[81]RacliffeW. S. Lai & KatieW. Y. Yeung & Mana M. N. Yung & Aleksandra B. Djurišić & J. P.Giesy & K. M. Y. Leung, "Regulation of engineered nanomaterials: current challenges, insights and future directions", Environ Sci Pollut Res, (2018): 1-18, doi: 10.1007/s11356-017-9489-0

[82]T. Singh, S. Shukla, P. Kumar, V. Wahla, and V. K. Bajpai, “Application of Nanotechnology in Food Science: Perception and Overview,” (2017): 1–7, doi: 10.3389/fmicb.2017.01501