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Review Article

Application of Nanotechnology Towards Improved Nutraceuticals, Pharmaceuticals and Theranostics: An Overview

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Abstract

Nanotechnology is an emerging field which has provided long lasting solutions to many areas of human health e.g., such as food, nutrition, medicine etc. To meet the increasing demand for more value-added food and pharmaceutical substances, nutritionists, scientists and researchers have turned to nanotechnology and other similar technologies to provide better solutions to persistent problems in these fields. These include efficient targeted drug delivery systems for pharmaceutical compounds and bioavailability of the nutritional compounds in food substances, etc. Already quite a few efficacious and improved nanotechnology-based pharma and dietary products are available in the market, enhancing significantly the satisfaction of the customers and manufacturers alike. This particular review deals specifically with the applications of nanotechnology towards better nutraceuticals, therapeutics, especially drug delivery systems for pharmaceutical molecules and theranostics, combining both diagnostics and therapy.

Keywords: Nanotechnology; Nanoparticles; Nutraceuticals; Theranostics; Nanoencapsulation

Introduction

Stephen L. DeFelice proposed the term 'Nutraceuticals' in 1989, combining the two separate words nutrition and pharmaceutical [1,16]. The term broadly encompasses any kind of feed and feed components providing general medicinal benefits towards prevention and/or cure from diseases, comprising of both natural and processed foods [26]. Because of this feature, the demand and market of nutraceuticals have grown enormously in the last few years [23]. In spite of this, a major hurdle in the domain of nutraceuticals has been their poor bioavailability, resulting in fast excretion of these substances from the body without providing their full health benefit to the persons consuming them [26]. Due to this lacuna, the potential of these compounds has not been fully realized, till date.

Nanoparticles are generally referred to those molecules or particles, of size less than 100 nanometres and possessing unique properties, attributable specifically to their small size, enhanced ratio of surface area to volume and high reactivity with biological specimens. These all lead to their higher uptake in the body. These properties of nanoparticles are quite distinct even from similar particles with same chemical composition, but bigger size. Because of these advantages, the nanoparticles are used along with nutraceutical compounds to enhance their bio absorption, bioavailability and sustained release *in vivo*. Nanotechnology is a latest and advanced field, wherein the unique physicochemical characteristics of nanoparticles are used to significantly alter the structure, texture and quality of food and health products. Similar applications of nanotechnology have also been sued in allied sectors like agriculture, diary, beautifying products like cosmetics etc. The wide-ranging applications of nanotechnology in various sectors of health, medicine and food has turned it to a multibillion-dollar industry worldwide. Various reports from diverse sources also confirm the far-reaching impacts of the nanoparticles in various sections, as mentioned above.

Nanomaterials used in nutraceutical formulations can be either organic or inorganic, based on the chemical nature on the main nutraceutical compound. Organic nanomaterials again, can either act as the active ingredient e.g., omega-3 fatty acids, or as the 'delivery vehicles' of the active ingredients. Inorganic materials mostly act as active ingredients only, e.g., nano silver and zinc oxide. But sometimes these exciting applications of nano materials can also be detrimental to human health, which warrants regulation of these materials to properly evaluate their safety and efficacy, for human consumption. As these are ingested orally reaching the gut, wherein they undergo various physio-chemical transformations through the process of digestion, ultimately resulting in their degradation and release of the active ingredients in the body. The European Food Safety Authority (EFSA) and European Centre for the Sustainable Impact of Nanotechnology (ECSIN) have proposed certain guidelines and protocols to test the safety of nanomaterials used in food, and also certain *in vitro* methodologies for production of safe and efficacious nutraceutical formulations.

Nanomaterial refers to a natural, incidental, or manufactured material comprising particles, either in an unbound state or as an aggregate wherein one or more external dimensions is in the size range of 1-100 nm for ≥50% of the particles, according to the number size distribution [30]. Nanomaterials can be applied to medical purposes in three different areas: diagnosis (nano diagnosis), controlled drug delivery (nano therapy) and regenerative medicine. Nanomedicine holds promising improvements in clinical practice by addressing unmet medical needs, such as (i) integrating effective molecules that otherwise could not be used because of their high toxicity (e.g., Mepact), (ii) exploiting multiple mechanisms of action (e.g., Nanomag, multifunctional gels), (iii) maximizing efficacy (e.g., by increasing bioavailability) and reducing dose and toxicity, (iv) providing drug targeting, controlled and site specific release, favouring a preferential distribution within the body (e.g., in areas with cancer lesions) and improved transport across biological barriers [30].

The achievements of this revolutionary technology are attributed to the huge surface-to-volume ratio, high porosity and different physiochemical properties of these amazing nanoparticles [31]. The ease of movement of these nanoscale sized particles in the human body compared to their larger counterparts, along with their unique structural, chemical, mechanical, magnetic, electrical, and biological properties, have been harnessed in designing nanoparticle-based therapeutic approaches. In this report, we have captured an overview of the use of nanoparticles in the field of nutraceuticals and pharmaceuticals [30].

Nanotechnology in nutraceuticals

Nutraceuticals, which are feed and dietary supplements with health promoting properties, as described above, has become part of the routine diet nowadays. The nutraceutical market is booming at present, because of the high interest and demand among customers of late for new, more nutritious, healthy and immunity promoting food products. But, as discussed earlier, the main challenge still in fully realizing the potential of nutraceuticals has been their poor bioavailability after consumption. These substances are generally having low solubility, leading to their poor absorption and bioavailability in the human body. In this regard, one of the most important applications of nanotechnology in food sector has been the formulation of novel nutraceutical compounds with improved properties viz. enhanced solubility, stability, bioavailability and efficacy. This is achieved by encapsulation of nutraceuticals by nanoparticles, which modifies their pharmacokinetics (PK) and biodistribution (BD). Various studies and trials have been undertaken of late to evaluate the physiological action of these nutraceuticals, so that improved and better encapsulation methods can be developed with the nanomaterials, which would increase their potency and safety. One of the effective methods has been encapsulation with the "generally recognized as safe" (GRAS) nanomaterials which promote sustained release of the functional ingredients of the nutraceuticals in the body, simultaneously also increasing their nutritional quality and stability. Examples of such nanomaterials include plant polysaccharides e.g., pectin, starch, gum etc, microbial polysaccharides e.g., Xanthan gum, dextran etc, food proteins e.g., soy proteins, casein, gelatin etc and emulsifiers e.g., lecithin, tweens, sugar esters, monoglycerides etc.

The choice of the correct vehicle of delivery for the active nutraceutical or pharmaceutical ingredients into the human body is very crucial for facilitating the direct contact of the ingredients with the target site of action in the body. As most of these are either poorly soluble or lipophilic compounds their delivery is significantly enhanced by altering the physicochemical properties like water solubility, partition coefficient, lipophilicity, crystallinity, etc. The poor solubility of the active ingredients poses multiple

challenges to their full utilization: route of administration, transport in the physiological system and reaching the site of action. All these in turn ultimately lead to the poor bioavailability of the same in the organisms. To overcome these challenges, one of the most efficient alternatives is to nano emulsify the active ingredients. Nano emulsions are extremely small, transparent or translucent droplet emulsions. The size of the droplets varies from 50 to 200 nm, much smaller as compared to the emulsions which are otherwise used for such applications, containing a continuous phase, dispersed phase and an emulsion stabilizer, also known as the emulsifier or surfactant. These nano emulsions provide many benefits as compared with pre-existing physical methods e.g., co-solvent addition, micronizing/milling, spray drying, salt formation etc. Nano-emulsification can encompass both hydrophilic and lipophilic active ingredients, leading to more kinetic and thermodynamic stability. In addition, the small size of the droplets facilitates easy transport of the nutraceutical ingredients across the cell membranes, thus increasing their plasma concentration and also bioavailability for the host.

Bioavailability is one of the most crucial and indispensable property of any dietary or pharmaceutical ingredient, which decides what proportion of the therapeutically active component reaches the systemic circulation in the host and is available at the target site for action. Many types of nutraceuticals are available today in the market with various proclaimed health benefits. One such prominent example among nutraceuticals are the phytochemicals including plant polyphenols (curcumin, resveratrol) carotenoids (lycopene, βcarotene, lutein) etc. These are widely favoured by the researchers, food manufacturers and consumers alike, because of their multiple health benefits viz. blood pressure regulation, reducing the probability of having malignant diseases like cancer, promoting digestion, immunity and growth, regulating glucose and cholesterol levels and also reducing stress by acting as antioxidants.

Characterization of nanomaterials used for nutraceutical purposes

The nanomaterials can be characterized based on their various physio-chemical properties e.g., the composition, state of dispersion, aggregation, size distribution, surface area, porosity etc. Because of the increasing applications and new dimensions of these nanomaterials being discovered almost continuously, it is being evaluated whether the existing methods are enough to completely characterize all the nanomaterials (NMs), or newer methods are necessary. Alterations of the physio-chemical properties of the NMs can occur in two ways, one synthetically by processing in complex matrices for nutraceutical applications and also naturally, during the digestive process in the physiological system, both ways significantly altering the stability and bioavailability of active nano-ingredients. Some of the alterations in the physio-chemical properties of both organic and inorganic nanomaterials are listed in table 1. Thus, both types of characterizations i.e., primary characterization based on their pure state and secondary characterization, based on their functional properties in complex matrices e.g., food, biological tissues and fluids becomes important, so that their efficacy and safety can be analysed in the physiological systems, in entirety. Various parameters should be considered for characterizing the physio-chemical properties of NM e.g., their typology and the measurement environment have to be checked on a case-by-case basis, as envisioned in the guidelines of EFSA regarding the risk assessment of nanomaterials used in nutraceuticals and dietary supplements. Figure 1 summaries the list of EFSA mandated steps for complete analysis of nanoparticles fragments used in the food products and food additives.

Figure 1: EFSA mandated steps for complete analysis of nanoparticles used in the food industry (Verleysen., *et al*. 2021).

Still, it is very difficult to pinpoint a single standard method to completely characterize or quantify the different NMs used in various food matrices. The main hurdle is to differentiate between inorganic and organic NM with a single method. Inorganic NMs are mostly metal in nature, and traditional techniques e.g., atomic

absorption spectrometry (AAS), plasma mass spectrometry (ICP-MS) etc. in simple matrices or some advanced techniques e.g., single particle (SP-ICP-MS) in complex matrices, can be used. On the other hand, the organic NM are quite difficult to characterize by traditional methods, as their composition itself is quite similar with matrices in which they are used for food and nutraceuticals, and consequently very few methods are available for their detection, characterization or enumeration. Due to this, it is required to use a combined approach of separation methods and detection systems e.g., HPLC and ultraviolet-visible (UV-Vis) spectroscopy.

But only compositional characterization is not enough to fully describe a NM. Data about certain other physical parameters are needed for that e.g., the physical form, morphology, particle size distribution etc, which can be obtained through imaging techniques like scanning and transmission electron microscopy (SEM and TEM). Light scattering techniques e.g., Dynamic light scattering (DLS) can provide additional information about particle size distribution of NMs in liquid samples, like average size and even size distribution of NMs, both during the synthesis and in physiological processes like digestion. Stability, which is another very basic and important property of any of the active ingredients inside the body, like the various nutraceutical components, can be measured by laser Doppler micro-electrophoresis method over a period of time, as zeta potential.

Finally, it can be said that no unique and verified methods exist for the characterization of NM in complex nutraceutical matrices, and a combination of methods is needed to properly evaluate all the important physio-chemical properties. This would facilitate its complete characterization and an understanding of its fate over the entire life-cycle, i.e., right from the synthesis, its physiological action in the body and excretion.

Organic nanomaterials	Inorganic nanomaterials
Alteration in surface charge	Alteration in surface charge
Small molecular interaction	Small molecular interaction
Protein adsorption	Protein adsorption
Enzymatic degradation	Surface passivation
Release of active component	Release of ions
Structure destabilization	Dissolution rate in presence of oxygen

Table 1: Various modifications of organic and inorganic nanomaterials due to physiological processes (Source: Zanella., *et al.* 2015).

Use of nanoengineering to produce nutraceuticals

Various nanotechnology platforms are explored to create these NM based delivery systems to increase the solubility and bioavailability of the NM. This is mainly achieved by integrative nano bioengineering to compile the different aspects of nanotechnology such as material sciences and microsystems technology with the biological sciences, ultimately leading to the development of innovative nutraceutical products with far reaching implications.

Materials used in nanoencapsulation

Different types of nanomaterials can be used to encapsulate the nutraceutical substances. These may be intentionally added to food components as delivery vehicles etc or accidently get their way into food e.g., leaching from packaging material in to the food components. However, the guidelines for using additives in food is quite strict and only those qualifying as the "generally recognized as safe" (GRAS) materials, according to the safety requirements EFSA or USA Food and Drug Administration (FDA), can be used. Examples of some compounds which qualify these requirements, viz. polysaccharides (alginic acid, xanthan gum, chitosan), proteins (albumin, zein, casein, or beta-lactoglobulin), lipids and low molecular surfactants, are generally used for encapsulating food components, because of their unique properties of biodegradability, biocompatibility and non-toxicity. Various nutraceutical compounds have different molecular and psychochemical requirements e.g., size, stability and surface properties and correspondingly need different delivery systems to address these. However, still any fixed standards are not available for most of these parameters and regulatory scientists are working in this regard to develop uniform guidelines and parameters for the assessment of the quality and safety of nanotechnology-based nutraceutical products. The cell membrane is the most preferred target for the synthesis of nanostructured biomaterials. Some examples of nanomaterials commonly used for encapsulation of various nutritious and therapeutic substances e.g., drugs, proteins, enzymes, nutraceutical compounds, in addition to facilitating their sustained release in the body, are 'alginate' and 'calcium alginate'.

Nanoencapsulation process

Encapsulation is the method of entrapping or coating different biological or chemical entities, so that they do not undergo degradation and their sustained regulated release can be obtained, under the prescribed physiological conditions. For

this, the primary step is to evaluate which kind of material is suitable for the encapsulation of the active ingredients. The main objective behind this, is to shield the bioactive component from the external destructive conditions at the target site, while permitting simultaneously the transport of the NMs into the neighbouring membranes and organelles. The main micro/nano capsule properties of the nutraceuticals which are targeted for alteration by the nanoencapsulation process to improve the efficacy, include configuration, mode of release, particle size, ultimate physical architecture, etc. Recently, considerable progress has been achieved towards increasing the oral bioavailability of the nutraceutical compounds by these processes. Generally bioactive molecules can be entrapped in a polymeric matrix in the form of a nanosphere or by a protective wall to form a nano capsule, and there are a number of techniques to achieve this encapsulation process. Since the encapsulating compounds are usually present in liquid form, many encapsulation techniques are based on drying methods *viz,* spray drying, spray-bed-drying, fluid-bed coating, spray-chilling, spray-cooling and melt injection. Even the choice of the appropriate drying procedure is crucial, as many nutraceutical compounds are liable to degrade or specifically lose their bioactivity, due to the heat and high temperature used during the encapsulation process.

Proteins, lipids and carbohydrates used as nano materials for encapsulation

Proteins

Most of the techniques used in nanoencapsulation use carbohydrates (gums) for the production of capsules and coating materials. However, same can be achieved using proteins as well. The stability of the nanoparticles in the gastrointestinal system is largely affected by pH, proteases, gastric juices and other food components present there. Some of the unique functional properties of proteins have made them as ideal target for study, to convert them into natural biodegradable polymers to function as delivery systems. These include gel, film and emulsion formation ability of the proteins providing the opportunity for developing delivery systems for both hydrophilic and lipophilic bioactive compounds, enhancing their biocompatibility, biodegradability, absence of toxicity, nutritional advantages and also because they are generally recognized as safe for applications in food. Examples of methods which are generally employed for encapsulation with proteins are precipitation at the isoelectric point, ion pair formation, cross-linking reactions, spray drying, extrusion, enzymatic gelation, gelation by acidification and heat treatment.

Lipids

Lipids are widely used in various encapsulation methods as bioactive agents in nutraceuticals. Prominent among them are nano-emulsion based delivery systems, utilizing the fact that lipophilic bioactive components can be easily entrapped within the hydrophobic core of the lipid droplets. Nano-emulsified biomaterials have greater stability against separation and aggregation than materials emulsified by conventional methods, because of their higher liquid droplet interface area, transparent systems and higher encapsulation efficiency. Liposomes are phospholipid vesicles, composed of a lipid bilayer having an inner aqueous phase, separated from an external continuous hydrophobic phase and are one of the most preferred lipid vesicular carriers in the production of nutraceuticals. Some of the methods usually employed for the production of liposomes include high pressure homogenization, micro fluidization, electro-spraying etc. Moreover, liposomes exhibit the exquisitely unique property of encapsulating all kinds of bioactive compounds, hydrophobic, hydrophilic and amphiphilic simultaneously. However, their rapid commercialization has been impeded to an extent due to certain shortcomings e.g., high cost, low physicochemical stability, drug leakage and fast release.

Carbohydrates

Polysaccharides (carbohydrates) also possess many unique properties to this regard e.g., biocompatibility, biodegradability and flexibility, making them preferred candidates as delivery systems for various industrial applications. They can interact with a wide range of bioactive compounds through their functional groups, thus providing them the ability to bind and entrap both hydrophilic and hydrophobic bioactive food ingredients. Moreover, they are also considered more temperature tolerant than lipid or protein-based delivery systems, thus again being mostly suitable for processes involving higher temperatures. The techniques used to synthesize carbohydrates-based delivery systems are diverse viz., spray drying, coacervation, electro spinning, electro road, supercritical fluid, emulsion-diffusion, inverted micelles, coalescence of emulsion droplets, evaporation of emulsion solvents, salting, ultrasonication and homogenization high pressure.

Limiting factors in the nanoencapsulation process

Certain factors can actually degrade the nutraceuticals at various stages of their life cycle e.g., synthesis, processing, storage, transport and delivery. These include certain food ingredients, pH, light, temperature, oxygen, undesirable interactions with other items etc. And delivery systems should be such that these harmful effects on can be minimized. Some of the principal factors are elaborated in the following section.

Emulsions and solvents effect

If preparation of emulsions by using solvents is needed in the particular technique, the correct choice of solvent and evaporation/ extraction method has a great influence on the final properties of the capsules obtained. To achieve good internalization of the active ingredient, the stability of the water-in-oil emulsion becomes crucial. If the first emulsion is low, leading to aqueous phase emerging with the continuous phase of another capsule, the overall potency of the microencapsulation becomes inefficient. Table 2 below illustrates the solubility characteristics of the final nano emulsions obtained by the encapsulation process.

Term	Parts of solvent required for 1 part of solute	Solubility defined in g/L
Very soluble	Less than 1 part	> 1,000
Freely soluble	1-10 parts	1,000-100
Soluble	10-30 parts	100 to 33.3
Sparingly soluble	30-100 parts	33.3 to 10
Slightly soluble	100-1,000 parts	10 to 1
Very slightly soluble	1,000-10,000 parts	1 to 0.1
Insoluble*	> 10,000 parts	< 0.1

Table 2: Solubility Terms of nanoparticles as defined by JECFA (2006) and European and US Pharmacopoeias (Council of Europe (2019); USP38 and USP38 NF33**- Source: More, S., *et al*. EFSA Guidance Document 2021).

*: The European Pharmacopeia terms it as 'practically insoluble.

**: The United States Pharmacopeia-National Formulary (United States Pharmacopeial Convention, 2016).

pH effect

The encapsulation efficiency and the final size of the capsules is also influenced by pH. It has been observed if the value of pH changes during conservation process, the size of the particles also change, since the gel charge gets altered leading to the modification of the density, in turn leading to either expansion or contraction of the molecules due to intra molecular repulsion forces. The ionic gelation technique allows for possible modification of the protein solubility by pH leading to a better interaction with the polymer and obtaining encapsulated hydrolysate in enhanced quantity.

Polymer

The problem with polymers like starch is that it is hydrophilic and also does not have emulsifying property, limiting its use for encapsulation of hydrophobic bioactive compounds. So, their chemical structures are altered employing chemical, biochemical, physical and/or enzymatic methods leading to generation of modified starches, such as cross-linked, oxidized, acetylated, hydroxy propylated. These have been reported to possess enhanced functionality and commercial applicability.

Temperature

Controlling the temperature effect on the gelling solution can lead to heat-set or cold-set gelation of certain biopolymers. This method can be utilized by injecting a hot-gelatine solution into a cold environment for generation of gelatine hydrogel beads. Generally, at high temperatures, the gelatine molecules tend to form random coils but on appropriate cooling, can turn into helical regions that form hydrogen bonds and can cross-link different molecules. Conversely, sufficiently heating above the thermal denaturation temperatures can make globular proteins e.g., those obtained from whey, egg, or soy, turn to gel by unfolding and associating through hydrophobic attraction and disulphide bond formation.

The temperature used in the nanoencapsulation process is not fixed and varies with the particular technique used in the process and the specific nutraceutical or pharmaceutical product being encapsulated. It has been observed that many of the nanoencapsulations processes for nutraceutical substances are achieved optimally even at room temperatures i.e., 25°C,

e.g., nanoencapsulation of bioactive eicosapentaenoic acid rich oil within whey protein microparticles, using emulsion electro spraying assisted by pressurized gas (EAPG) technology [44].

Nanotechnology in therapeutics and theranostics

Therapeutics

Nanomedicine is a fast-developing branch of science where nanomaterials are used for the diagnosis or treatment of diseases, by specific and controlled delivery of drugs at the target site. This has led to a revolution in the field of personalized and precise medicine. For example, of late nanoparticles and materials, in various applications, as chemotherapeutic, biological, and immunotherapeutic agents etc, to treat a multitude of diseases. In biosciences, especially medicine and pharmacy, the are many promising areas of application for nanotechnology viz. nanobiotechnology, nano-pharmaceuticals, nanomedicine, nano-pharmacology, nanotoxicology etc. for the improvement of drug delivery systems. For example, by employing nanotechnology; it's possible to: Enhance the delivery efficiency and controlled release of the targeted drug; minimise the dose and consequently increase the safety level, Increase the bioavailability of the oral administration medicaments, Extended drug existence in biological fluids, Reduced the toxicity and side effects of drugs, Increase the drug solubility and dissolution rate, Improve the stability of drugs in the biological system and protect drugs from early biodegradation before reached to the pathological sites, Increased the surface area to improve pharmacodynamic and pharmacokinetic properties and overcome the cancer drug resistance and other microbial and infection diseases [76].

A recent trend also has been the practise of using lot of compounds for various therapeutic purposes, to the tune of almost one-fourth of the all the various pharmaceutical agents today are based on natural substances. This is owing to their various intrinsic advantages reduced side effects and negligible toxicity, in addition to possessing high therapeutic potential and low cost [46,47]. Still due to the issues of biocompatibility and requirement of higher doses of the natural compounds, their full therapeutic potential has not been realized.

A lot of problems exist for using large sized materials for drug delivery in the body, viz. stability, poor solubility, bioavailability, proper absorption and distribution in the body, target-specific delivery, efficacy and even potential toxicity or side-effects. Thus, adopting an improved, precise and targeted delivery system to direct the drugs to the target organs is a need of the hour. This gap is being uniquely filled by nano-medicines, employing nanomaterials coated drugs and therapeutic agents to be specifically targeted to the diseased site with sustained release, significantly improving the success rate of the treatment [48,49]. Based on its efficiency, the application of nanomedicine has been further extended to the advanced areas e.g., biosensors, microfluidics, microarray tests, to tissue engineering etc. [50-52]. Because of their the atomic or molecular sizes, mostly in the form of nanospheres, they possess the liberty of free movement and distribution in the body, which is difficult for the large sized molecules. They also have the extra advantage of possessing distinct and characteristic structural, chemical, mechanical, magnetic, electrical, and biological properties etc. [53]. The significance of Nanomedicines has become prominent in the recent years due to the reason that nanostructures could be utilized for encapsulating or attaching therapeutic agents or drugs, making them as ideal delivery agents for the same, with enhanced precision and better, controlled release [54,55]. This has ushered its application in many advanced and emerging area of clinical therapy, especially disease prevention and remediation, utilizing various nanoscale tools like nanorobots, nano-sensors for diagnosis, delivery and for live cells imaging.

One of the initial examples and applications of nanomedicine includes the use of FDA-approved liposomes and micelles, which can encompass both inorganic nanoparticles e.g., gold or silver and magnetic nanoparticles. This actually has facilitated the extensive use of inorganic nanoparticles for therapeutics and diagnostics [56,57]. Nanostructures have demonstrated multiple advantages as delivery vehicles as compared to the traditional and large molecules, viz., they protect the drugs from being degraded in the acidic environment of the gut and even aid in hydrophobic drugs in reaching their target. The oral bioavailability demonstrated by the nano encapsulated drugs is also enhanced. As discussed earlier, nanomaterials enable control and sustained release of drugs in the circulatory system, increasing the treatment efficacy, which is again due to their property of extended persistence in the physiological system. Interestingly, this has also led to the reduction in the blood drug concentration fluctuations and the corresponding side-effects in the patient. Moreover, due to their extremely small sizes, they can easily penetrate the tissues and organs, facilitating increased

uptake of drugs by the cells present at the target site leading to better therapeutic outcome [58,59]. One of the outstanding features of nanoparticles is that various therapeutic proteins can be attached to its surface, gold nanoparticles used in that fashion as biomarkers and tumour labels for cancer diagnostics.

The choice of the nanomaterials used in drug delivery, etc depends on the physical and chemical properties of the drugs. The amalgamated use of nanoparticles with natural bioactive compounds is expanding on a rapid pace, because of the combined uniqueness of natural substances to treat various kinds of diseases, enhanced by the drug delivery potential of nanomaterials. The pharmacokinetic properties of the natural molecules were also observed to be enhanced by emulsification with nanomaterials. Though the same results have been observed in case of synthetic drugs also. Various kinds of polymeric nanostructures e.g., metallic, organic, inorganic, dendrimers, micelles, liposomes etc are utilized for tagging drugs and designing target-specific delivery systems, with a special focus on the drugs exhibiting reduced solubility and absorption ability [58,60]. Still the potential of these nanostructures as drug delivery vehicles, also depends on the inbuilt physiochemical properties of the nanomaterials, size, shape etc. [61]. For fabrication of nanoparticles to effective nanomedicine vehicles, various kinds of both natural (e.g., alginate and chitosan) and synesthetic (e.g., polyvinyl alcohol, poly-l-lactic acid, polyethylene glycol, and polylactic-or-glycolic acid) polymers with pronounced biocompatibility and biodegradability characteristics are used. These polymer fabricated nanomaterials are again categorized into two types, nanospheres and nano-capsules, both serving as excellent drug delivery agents. Similarly, lipid-based nanostructures and phospholipids like liposomes and micelles have also found extensive application in precise therapeutic drug delivery [62-65].

One more application of nanomaterials for treating malignant tumours is the permeation and retention effect (EPR-effect). The EPR effect signifies the enhanced capability of various nanosystems (liposomes, micelles, nanoparticles, nanovectors, nanotubes, quantum dots, etc.) for delivering anticancer therapeutic agents to the target organs. "Nanovectors" is the umbrella term referring to the combination of various types of organic and inorganic nanoparticles, nanowires, and nanotubes, employed in the fight against cancer. One of the prominent examples being liposomes, used for delivering different kind of drugs to the target. The following list of FDA approved nanotechnology based drugs only illustrate the current importance of nanomedicines for the treatment and cure of various diseases, namely, Doxil® (Caelyx) – manufactured by Ortho Biotech Schering-Plow, AbraxaneTM - manufactured by American Biosciences, Diprivan - manufactured by Zeneca Pharma, Estrasorb - manufactured by Novavax, TriCor®-Abbott Laboratories USA, Rapamune® - manufactured by Elan Pharmaceuticals, Megace ES - manufactured by PAR Pharmaceutical, Elestrin- manufactured by BioSante, Marqibo®-Talon Therapeutics, Inc etc. Some of the largest pharmaceutical brands globally are involved in producing nanomedicines on a regular basis, viz. GlaxoSmithKline (GSK), Merck, Johnson and Johnson, Novartis, Pfizer, etc. [76].

In spite of all this advantages of nanomedicine, certain drawbacks of it, especially the toxicity exhibited by certain particles in the patient's body cannot be completely ignored. Some of the potential risks of using nanoparticles and nanomaterials for various applications are as follows. Apart from the issue of toxicity, use of nanotechnology has also ushered other challenges related to it, in terms of its environment, social, ethical, legal, and cultural implications [66]. As the scope of nanotechnology has extended to various industrial sectors like healthcare, agriculture, energy, automobiles, materials etc, people employed in the sectors and even the consumers buying the end products stand a high chance of exposure to ENPs (engineered nanoparticles) through multiple routes e.g., ingestion, inhalation, absorption via skin, and even directly like injection [67,68]. The study into the side-effects of inhalation of nanoparticles viz. titanium dioxide (TiO_2) and accumulation in the body through food, sunscreen and carbon nanotubes has revealed significant scarring and damage in the lung wall, in addition to both reduced oxygen and carbon dioxide levels, ultimately resulting in breathing problems in the person. The effect is more pronounced on long-term exposure [69-71].

It is reported that upon absorbtion through the skin, nanoparticles can exter detrimental effects both at the cellular and subcellular levels, e.g., damage the DNA of the host [72]. Similarly, if copper nanoparticles, entering the human body through food, water, drugs, cosmetics, etc, exceeds the normal levels, it can lead to haemolysis, jaundice and even death [73,74]. Research into their toxic effects have demonstrated that due to their nanosizes, ENPs can even cross the blood-brain barrier and directly impact

the central nervous system of the host, like damaging the neuronal cells, causing neuroinflammation, prooxidant changes etc. [68]. The nanoparticles used in packaging also has the ability to leach into food items, instigating safety and health concerns [75]. At present, it is challenging to correctly verify the relation between toxicity and exposure levels of nanomaterials, due to lacunae in the proper evaluation and detection methods. Therefore, in parallel of developing the field of nanotechnology, it is imperative to also upgrade the assessment technology and methods to establish a direct correlation between the nanomaterial exposure levels and the resulting toxicity.

This has further accentuated the use of a combination of nanomaterials and natural products to obliterate the problems related to toxicity. Presently the designing of nanoparticles customized for nanomedicine, is mostly preferred through the green chemistry route, as it generates considerably less amounts of hazardous constituents, in turning also diminishing the side-effects of treatment process [55]. In addition, the modification of the physio-chemical properties of the nanostructures e.g. the size, shape, hydrophobicity, and surface changes can boost their bioactivity. Thus, the treatment of various clinical diseases can be significantly improved by the application of nanotechnology, mainly by enhancing the target-specific and controlled delivery of various therapeutic agents, though more studies and methods are need of the hour to increase its safety and efficacy simultaneously.

Theranostic nanoparticles

Presently even one step furher application of nanoparticles is the field of "Theranostic" nanoparticles. These are multifunctional systems, combining both diagnostic and therapeutic abilities and leading to highly specialized and personalized medicinal therapy. Generally, these are composed of biocompatible and biodegradable nanomaterials. There are certain intrinsic properties necessary in a theranostic nanoparticles, viz. safety, targeted delivery, fulfilling the diagnostic applications e.g. assessment and reporting of biochemical and morphological parameters, rapid translocation to target and the ability to deliver the required dose of drug when needed, selective action without affecting healthy organs, being biodegradable and effective clearance from the body within specified time [77-80]. There are in fact many kinds of organic and inorganic theranostic nanoparticles presently used in oncology, but few have fulfilled all the criteria, as mentioned above. Various preclinical and clinical trials have demonstrated the potential of peptide- or antibody-conjugated theranostic nanoparticles for tumor imaging and therapy [80,81]. Still, substantiated efforts are needed to generate theranostic nanoparticles which are biocompatible and importantly possessing tumor active targeting ability *in vivo* with highest specificity.

Synthetic pathway and translational applications of theranostic nanoparticles

There are various ways to engineer theranostic nanoparticles. One route of synthesis is by tagging or loading) therapeutic drugs onto imaging nanoparticles viz. quantum dots, iron oxide nanoparticles (IONPs), gold nanocages, etc. it can also be synthesized exactly in the opposite way, i.e., by conjugating or loading detection agents e.g., like fluorescent dyes, optical or magnetic nanoparticles and radioisotopes onto the therapeutic agents. Other mechanism and quite efficient, is using by using nanoplatforms which are biocompatible such as polymeric nanoparticles, ferritin nanocages and porous silica nanoparticles for encapsulating both imaging and therapeutic agents together. Even unique nanoparticles have been constructed possessing potential imaging and therapeutic properties e.g. porphysomes, [64Cu]CuS, gold nano-shells and cages etc. A new and advanced approach for further improving the half-life of the nanoparticles in circulation and ability to actively target tumor, has been surface modifications post-synthesis with polyethylene glycol and various targeting ligands. Still, as already discussed before, the potential of the nanoparticle-based theranostic agents have also been impeded of late by toxicity related issues. Despite the fact, it is heartening to observe, that on account of the extensive studies and efforts to reduce the toxicity, Food and Drug Administration (FDA) has so far approved more than 35 imaging or therapeutic nanoparticles for clinical trials [82].

Theranostic nanoparticles are still in the nascent stages, with majority of them limited to the level of preclinical studies only. To facilitate their use in clinical research and possible therapeutic potential later, FDA-approved imaging and therapeutic nanoplatforms have been used in the recent years to engineer theranostic nanoparticles. Some examples of such platforms include biodegradable polymeric nanoparticles, IONPs, gold nanoparticles or nano-shells (NCT00356980, NCT00848042), silica nanoparticles (NCT02106598), and silica-gold nanoparticles

(NCT01270139). Another very latest interest of selected scientists working in the area has been to create 'activatable theranostic nanoparticles' for even better cancer imaging and therapy. For example, SN-38 (a topoisomerase inhibitor) has been a conjugated with piperazinerhodol fluorophore with the help of a self-immolative linker based on disulfide bonds, for synthesizing an activatable theranostic prodrug. It has facilitated, in the presence of intracellular thiols, real-time monitoring of the delivery and release of the SN-38 molecules [83].

A very recent area of interest has been to study the potential of certain nanoparticles, e.g., gold and silver, to be used as superconductors, where certain groups of researchers have already reported some progress. Superconductors are essentially used in some of the advanced diagnostic equipment used in medicine, e.g. MRI, to boost the efficiency and avoid the grid loss during power transmission, but its utility gets limited by the requirement of extreme low temperatures or high pressure or sometimes both, making them both difficult to operate and even costly. Both gold and silver nanoparticles, on the other hand, have exhibited superconductivity at ambient temperatures, obliterating the need of the extreme conditions. This ability of the nanoparticles, when fully validated, would bring about a revolution and markedly enhance the clinical diagnosis of various diseases.

Figure 2: Summary of the applications, concerns and ultimate goals of nanotechnology in therapeutics and diagnostics.

Conclusion

In the recent past, lot of efforts have been directed towards the development of improved food, nutraceutical and pharmaceutical products. One such application is nanotechnology, which confer unique properties to food and nutraceutical products, making food and dietary products tastier, healthier, nutritious, in addition to generating new food products, improved methods of food packaging and storage. Engineered nanoparticles-based delivery systems have led to the enhancement of various aspects of the nutraceuticals viz. bio accessibility, solubility, absorption, stability in the gastrointestinal tract, ultimately boosting their bioavailability and potentially increasing their health benefits in humans.

However, still some challenges exist e.g., most of these applications are at an elementary stage, and have been used only for high-value products. Moreover, importantly, the use of nanomaterials in nutraceuticals have raised concerns on their safety for human health. EFSA has developed certain guidelines and framework to assess the safety and efficacy of the nanotechnological applications in food and nutraceuticals. Regulatory scientists and administrators are making great effort to evolve different legislative frameworks based on these guidelines, requiring certain vital information for the appropriate assessment of the quality and safety of nanotechnology-based products.

The application of nanotechnology has significantly advanced and revolutionized the medical field also, especially in the areas of therapeutics and diagnostics, combinedly known as 'theranostics'. Some of the marked advantages are evident in the aspects of improved efficacy of nanomedicines, controlled drug delivery and regenerative medicine. Many of the nanomaterial based medicinal products approved by FDA are already in the market, e.g., liposomes-based drugs, facilitating improved treatment of chronic diseases like cancer and leading to better human life. The promise of nanomedicine looks to be even greater than other advanced and latest therapeutics interventions, e.g., stem cells, gene therapy or even RNA interference (RNAi). Various studies have also indicated to the safety of the nano material-based drugs, meeting all the required standards, in addition to their enhanced efficacy. Due to this unique property, the NM applications are being envisioned in complex areas like treating even multidrug resistant (MDR) bacterial infections. The application of nanomaterials in diagnostics have also exhibited to amplify the sensitivity of detection of diseases and pathogens by many folds, leading in turn, again to better prognosis and treatment of diseases. The potential of gold and silver nanoparticles, to be used as superconductors in advanced medical diagnostics, e.g., MRI, is being presently explored.

Finally, it can be said that like any other new innovations and technological applications, the success and outreach of the applications of nanotechnology in nutraceuticals and theranostics would depend upon the utility, safety, public confidence and acceptance.

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