Chapter 13

Revolutionizing Nanoparticles in Veterinary Care: Classifications and Benefits

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ABSTRACT

The development of new methods for working with materials at the nanoscale has affected many medical fields. Thousands of nanomaterials exist today, and they can be categorized based on their shape, place of origin, or application. Nanotechnology offered fresh approaches to time-honored issues. They are employed in the medical sciences for either therapeutic or diagnostic objectives. They can also be used to make nano-adjuvants and nano-vaccines. Its use in cancer treatment and gene therapy ushered forth a new era in medicine. In the veterinary field, nanotechnology is now finding a lot of applications. They are quickly taking over the fields of animal nutrition, diagnostics, farm disinfectants, veterinary vaccine manufacturing, and animal breeding and reproduction. The public's health is immediately affected when they are substituted for commonly used antibiotics. These measures address the problem of residues in meat and milk and lessen drug resistance in both veterinary and human treatment. Additionally, this approach offers significant economic benefits by decreasing the quantity of milk that is wasted and reducing the number of culling of calves in dairy herds. Additionally, nanotechnology has been used in the development of sanitary products and pet care items. This chapter covers the many types of nanoparticles, the advantages of using nanomaterials over their equivalents, and the applications of nanotechnology in the field of animal care.

KEYWORDS
Nanovaccines, Nanomedicine, Nanotechnology, Nanomaterials, Nanoadjuvants

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INTRODUCTION

The term "nanotechnology" was first coined in the year 1974 to describe technical tools used to manipulate materials at the nanoscale. Materials with dimensions between 1 and 100 nanometers are considered to be at the nanoscale. Examples of naturally occurring nanoscale biological materials include protein molecules, which are approximately 5 nanometers wide, and DNA molecules, which are about 2.5 nanometers wide. In contrast, human hair is roughly 80,000 nanometers wide. Today, "nanobiotechnology" refers to the nanotechnological use in life sciences (Troncarelli et al., 2013). "Nanomedicine" provides quick and efficient medical treatments by using nanotechnology-based techniques. This field overcomes the limitations of conventional medicine. In addition, it also enhances our understanding of various processes in living organisms. These processes may be physiological and pathological. Thus, it will be leading to innovative treatment strategies (Mohantya et al., 2014). Additionally, terms like "nanotheranostics" describe advanced formulations that serve both diagnostic and therapeutic purposes. These formulations provide critical information about drug distribution, release locations, and efficacy, aiding in the customization of treatment approaches (Chapman et al., 2013; Rizzo et al., 2013).

The recent significant advancements in nanotechnology have made it possible to conduct smart chemical delivery studies for medicinal and diagnostic purposes. They have the ability to treat damaged or malignant cells while sparing healthy ones, identify illnesses before symptoms show up, provide hormones or enzymes when required, and much more. Smart delivery systems can be designed to automatically respond to changes in temperature, acidity, or specific chemicals on their own (Scott, 2007; Manuja et al., 2012).

The recently established nanobiomaterials, which have sizes ranging from 5 to 20nm, are architecturally designed to mimic different bodily receptors, DNA, ligands, and proteins. These structural commonalities enable them to interact with tissues and cellular membranes in a range of biological situations (Venkatesan and Kim, 2014; Yi et al., 2016).

Lipid-based and biodegradable carriers of nanoparticles are among the most popular types of nanobiomaterials. The long-term toxicity and the bioaccumulation of nanoparticles in cells are avoided by using these nanobiomaterials. Macrophages can easily degrade and engulf those (Dobrovolskaia et al., 2016).

The following factors play a role in selecting the proper size of nanoparticles (NPs) (Cormode et al., 2013)

- The Nature of target cells
- The planned application
- The type and quantity of payloaded agents
- The need for rapid excretion
- The preference for tissue internalization versus a longer half lifetime in the circulation
- The method of clearance (reticuloendothelial OR renal system)
- Biodistribution
- Contrast image intensity
- The desired amount of stimulation of the immune system
- The type of the immune response

While certain nanoparticle (NP) sizes are suitable for medical applications, large NPs are typically avoided because of the risk of causing embolisms. They are also quickly phagocytized and removed from the bloodstream. Conversely, very small NPs are rapidly filtered out by the kidneys. Their tiny size increases the surface area to volume ratio, making them more toxic and reactive, leading to a noticeable rise in their chemical and biological activity. This results in an increase in the number of production of free radicals and reactive oxygen species (ROS) (Jain et al., 2011; Venkatesan and Kim, 2014; Yi et al., 2016). The release of reactive oxygen species (ROS) can lead to considerable oxidative stress and inflammation. It may also lead to damage at the cellular level. When NPs accumulate in the mitochondria, they hinder the body's defense system (Meena et al., 2018).

Nanoscale materials have different physical, chemical, and biological properties than their larger material forms. The small size of the material means it has better solubility and reactivity, thereby being more bioactive. Hence, with the increased potential, they become steadier and less susceptible to inactivation by oxidation (Troncarelli et al., 2013; Swain et al., 2015). In light of this occurrence, NPs can be modified to increase solubility, improve pharmacokinetics, reduce immunotoxicity, and minimize side effects in addition to being used as carriers. For instance, patients who are vulnerable to anaphylactic reactions may experience side effects from the cancer therapy drug Paclitaxel (Cremophor-EL); however, these reactions can be eluded by utilizing Paclitaxel (Abraxane) (Dobrovolskaia et al., 2016). Therefore, if the chemical is nano-sized, new science for nanopharmacokinetics needs to be introduced in light of the newly discovered properties of the same chemical (Casals et al., 2017).

There are many forms of nanoparticles known today. A nanoparticle basically contains a nucleus that is surrounded by a casing or capsule. The payload is planned to be delivered by NPs in two modes: either the medicinal and diagnostic compounds are attached onto the superficial surface, or they get encapsulated or trapped inside the nanoparticles. The proper encapsulation of the NPs can enhance diffusion, disintegration, drug loading, and imaging. This coating material will then protect the particles from any attack of the immune system, catalyst-induced disintegration, and harsh conditions of pH. Certain coating materials, such as PEG, protect the particles from the immune system's attack, disintegration because of catalyst, or harsh conditions of pH (Sailor and Park, 2012).

Nanoparticles Types

Nanoparticles (NPs) can be classified on the basis of their place of origin and various other factors. There are three main categories:

1. Shape: NPs can take forms such as spheres, tubes, or liquid drops.
2. Application and Type of Payload: NPs can be used for therapeutic, diagnosis, administration of vaccine, or for nutritional purposes (Thulasi et al., 2013).
3. Origin of NPs:

i. Organic NPs: These include proteins, peptides, and lipids.
ii. Inorganic NPs: These consist of materials like gold, silver, silica, iron, magnesium, or graphene.
iii. Hybrid NPs: These are a combination of organic and inorganic materials (Torres-Sangiao et al., 2016; Riley and Vermerris, 2017)

However, the major classification will be discussed in detail later in the chapter (Fig. 1)

Currently, the most commonly used taxonomy categorizes NPs as follows:

1. Polymeric NPs: According to Torres-Sangiao et al. (2016), there are 2 categories of polymers:

i) Synthetic polymers e.g. polyethylene-glycol

ii) Natural polymers, based on polysaccharides, e.g. chitosan and inulin

They are made by sticking active molecules onto the surface of the nanoparticles, which then form intricate, tree-like shapes. The shape is somewhat dendrimer-like, but the branches radiating from the center of NPs have differing numbers of branch points (Elgqvist, 2017; Mohantya et al., 2014). What distinguishes them is their substantial loading and conjugating capacity. Additionally, these polymers can be used to create hydrogel nanoparticles (NPs), which have a large surface area and high-water content (Torres-Sangiao et al., 2016).

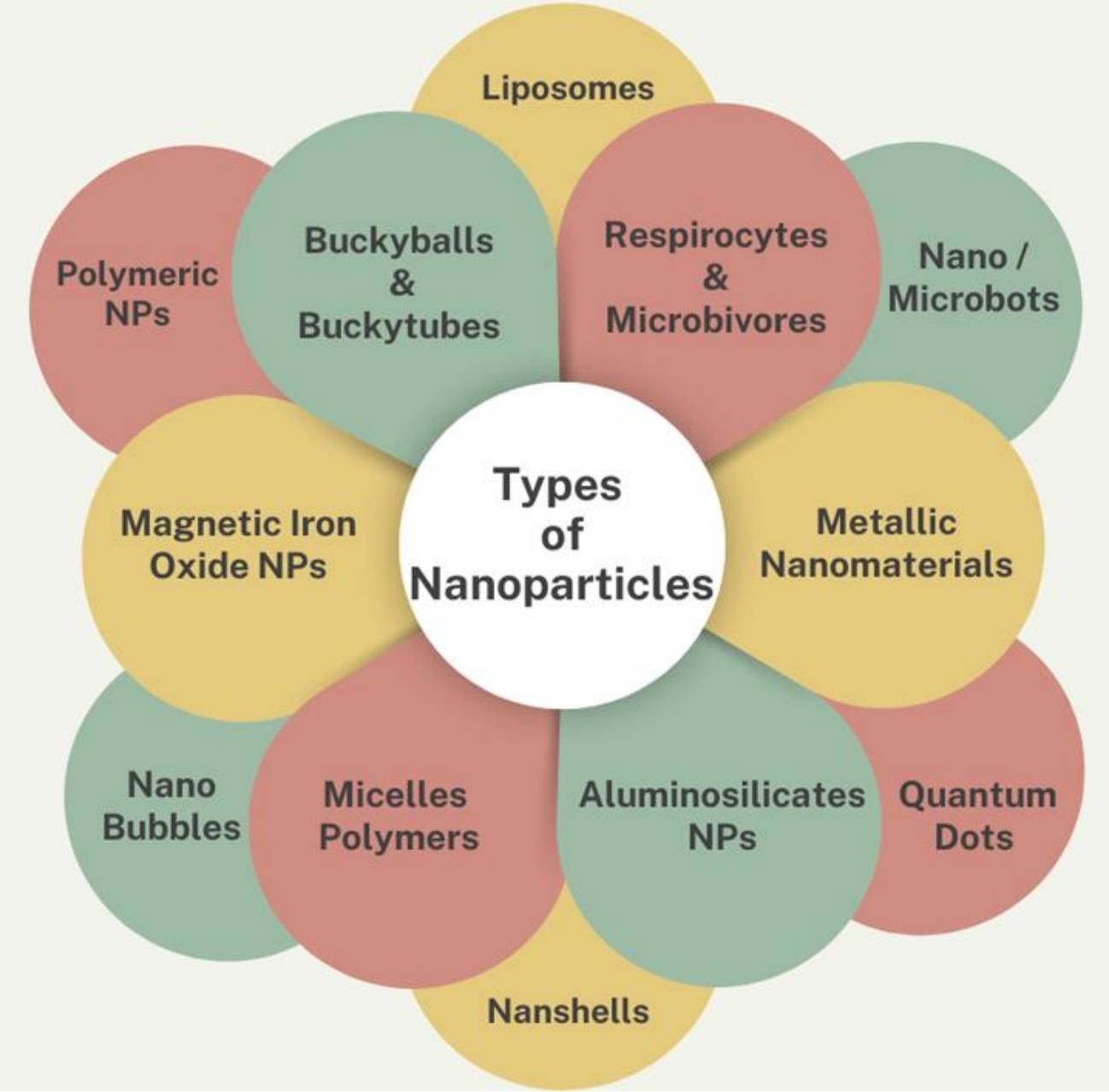

Fig. 1: Categories and Classifications of Nanoparticles.

2. Liposomes: These are the round, non-toxic, biodegradable PE glcolylated nanoparticles. Small- interference RNA (siRNA) for gene therapy, radionuclide or strings of RNA, DNA, and water-soluble drugs are transported by their aqueous core. Drugs that are fat soluble (hydrophobic) are encased in a double-layered phospholipid shell coating the particles. Various antigens, including viral envelop glycoproteins (referred to as virosomes), can be encapsulated by them (Torres-Sangiao et al., 2016). An exterior protective PEG layer covers the outside of the particle, shielding it from immune system attack. It is possible to fix chelated antibodies for imaging and targeting agents on the outside. This group's primary benefit is their capacity to administer therapeutic medicines that are both hydrophilic and hydrophobic (Elgqvist, 2017). Both hydrophobic and hydrophilic medications can be administered topically or by injection using liposomes; however,

liposomes cannot be taken orally due to the lipids' propensity to be broken down in the gastrointestinal system. Their eco-friendly structure is important in their safety. Moreover, liposomes can be attached to foreign antigens for vaccination or to antibodies known as immunoliposomes, which can bind to cancerous cells (Bakker-Woudenberg et al., 2005) (Mohantya et al., 2014). (Mohantya et al., 2014). In this regard, Torres-Sangiao et al. (2016) noted that liposome-polycation-DNA nanoparticles that can act as an adjuvant with DNA vaccines can be formulated by complexing cationic polymer-condensed DNA with cationic liposomes.

3. Fullerenes, or buckyballs, and nanotubes, or buckytubes: According to Meena et al. (2018) and Mohantya et al. (2014), buckyballs are small, spherical carbon-based nanoparticles that have a tendency to interact with proteins, cells, and pathogens. Fullerenes are typical in a number of varieties, each consisting of 20 or 60 or 100 carbon atoms. The other shape of carbon-based nanoparticles is the so-called buckytubes, which are very distinctly cylindrical and can be of one, two, or multiple walls (Elgqvist, 2017). By functionalizing these nanotubes, they can be employed as biosensors to detect immunoglobulins, glucose, ethanol, or to facilitate electrochemical DNA hybridization (Manuja et al., 2012).

4. Nanoshells: They are spherical, having an ultra-thin metal layer, usually gold, over a glass or silica core. The thickness of the gold layer can be varied to change their optical properties so that they respond to different wavelengths. Because of these properties, gold nanoparticles are mainly used for diagnosis of cancer (Mohantya et al., 2014). Infrared light can pass through blood samples. By attaching gold nanoshells with antibodies, it is possible to even detect very low levels of immunoglobulins in blood samples (Manuja et al., 2012). Because of its minute size, nanoshells can also act as therapeutic agents, as they tend to accumulate in tumor tissues. IR lasers are able to pass through healthy tissues without heating them. In tumor cells, nanoshells absorb infrared (IR) radiation and generate heat. This heat produced melts the polymer coating of the nanoshell.This releases the entrapped drug directly at the site (Freitas, 2005). Prolonged exposure to temperatures as high as 55°C can cause death to the cancerous cells. Nanoshells are chosen instead of quantum dots because gold is safe for the body and doesn't react, while quantum dots contain cadmium, which can be harmful (Hirsch et al., 2003; Krishnan and George, 2014).

5. Solid lipid Nano Particles: SLNPs are the suspensions of lipids stabilized in water. They have a hydrophobic core of lipids capable of solving lipophilic radionuclide-based pharmaceuticals used in cancer therapy, enclosed by the dehydrated tails of phosphatidylcholine lipids. An outer shell is hydrophilic that is formed to envelope the fatty core and can be coupled to many hydrophilic pharmacological drugs or antibodies. An outer hydrophilic layer improves plasma stability and, hence biodistribution of the drug, increasing its bioavailability. Solid lipid NPs (Cationic) can electrostatically associate with DNA or RNA segments, so they are suitable for gene therapy according to Elgqvist, (2017). Injections may be administered continuously over several weeks, but they may also be topically or orally applied. They get easily and quickly absorbed through mucosa because of their lipid composition and adhere to mucous membranes. Another chief advantage of solid lipid NPs is that they get efficiently delivered across the central nervous system and cross the blood-brain barrier. According to Mishra et al. (2010) and Mohantya et al. (2014), one of the main advantages of solid lipid NPs is that they can cross the blood-brain barrier. In contrast, Krishnan and George, (2014) stated that solid lipid NP formulations act as colloidal carriers, which remain in a solid state both at room temperature and inside the human body. Apart from solid lipid NPs, research into liquid lipid NP is going on at present (Elgqvist, 2017).

6. Micelles polymeric: Their hydrophobic cores allow them to carry materials that are insoluble in water, unlike solid lipid nanoparticles. They have a hydrophilic layer covering their hydrophobic core, which makes them extremely water soluble. Four types of polymeric micelles can be distinguished based on the characteristics of their shells:

i) Phospholipid
ii) Poly-amino acid
iii) Biocompatible polyester
iv) Pluronic micelles (Mohantya et al., 2014)

7. Dendrimers: Dendrimers are an artificially created polymer that is minute, even compared to human cells—thousands of times smaller. These NPs are well-known for the following properties: high biocompatibility, water solubility, and polyvalency. (Chakravarthi and Balaji, 2010, Jurj et al., 2017). Their small size and chemical properties make them unlikely to trigger unwanted immune responses upon entering the bloodstream (Chakravarthi and Balaji, 2010). Dendrimers are basically three-dimensional molecules with a branching structure, much like trees. Drugs can bind to the dendrimers at the functional groups available on their surface, or they can be encapsulated within the dendrimers' core according to Mohantya et al. 2014. In that, the dendrimers' epoxy cores can load a wide variety of hydrophobic and hydrophilic drugs through non-covalent interactions, chemical bonding or physical entrapment. The covalent conjugation of dendrimers with drugs improves the therapeutic efficiency and increases the stability of the drug as reported by Jurj et al. 2017.

8. Metallic nanomaterials: These materials consist of different metals, primarily gold. Mohantya et al. (2014) provide evidence that the main application for these materials is treating cancer. Other metals, which are occasionally used for making these materials, are manganese, silver, gadolinium, and platinum. The iron cores are contained in biocompatible

capsules, which have protective shells. The capsules may be functionalized to bind different medical or imaging agents, such as chelated radionuclides or targeted antibodies. Polyethylene glycol (PEG) coats the particles to help them avoid detection by the immune system and reduce unwanted sticking (Elgqvist, 2017). Lately, nanoparticles made from two metals, like silver-gold, silver-selenium, and gold-platinum, have been used in cancer treatment (Mittal et al., 2014; Alshatwi et al., 2015; Fakhri et al., 2017).

9. The great advantage of magnetic iron oxide NPs is the fact that they can be guided toward target cells by the help of an external magnetic field applied from outside, and due to this property, they have been proved very useful for imaging, thermal therapy, and drug delivery. However, there are fears that they might get accumulated in tissues (Manuja et al., 2012; Mohantya et al., 2014). These nanoparticles feature an iron core, either Fe_3O_4 or Fe_2O_3, surrounded by a silica shell embedded with chemotherapeutic agents. The outer polymeric shell, which is also functionalized with antibodies targeting tumor antigens, provides extra stability and must be dark to prevent intervention with fluorescence. Due to their magnetic nature, these nanoparticles are employed as multifunctional theranostic complexes in various MRI applications for diagnosis of cancer and treatment. Normally, the nanoparticles are PEGylated for preventing agglomeration and protecting them from immune attacks. However, silica is used as a coating material of choice for its application in contrast agents intended for light absorption applications in cancer imaging (Elgqvist, 2017).

10. Ceramic nanoparticles are straightforward to design and have several advantages over other types. They are also fully inert. They are easily molded into a variety of shapes, sizes, and porosities. According to (Mohantya et al., 2014), they provide protection for their burden against extreme temperatures and pH values.

11. Quantum dots are small, semiconductive nanomolecules that can be light-activated and are in the size range of 2-10 nm. Developed initially for optoelectronic applications, these semiconductor components normally comprise CdS, CdSe, CdTe, ZnS, and ZnSe materials (Patil et al., 2009; Torres-Sangiao et al., 2016). A typical quantum dot includes an inorganic nucleus and capsule (covering), with an aqueous coating that may be attached to various proteins. The size of the crystal changes the color of the light emitted. QD have been tailored to be inexpensive, simple, and durable probes, which emit light for an average of several hrs or days (Manuja et al., 2012). They can be labeled with biomarkers like DNA or proteins; hence, they are useful in screening blood samples for particular proteins, infections, and tumor markers. Thus, they find extensive applications in immunodiagnostics and diagnostics, respectively (Mohantya et al., 2014). QD physical properties make them a very suitable tool in imaging applications, and their advanced tools have been applied in pharmacological studies, medical diagnostics, and genetic tests (Meena et al., 2018). The fact that they are able to trace drugs and biomolecules in the body and image cellular pathways makes them very useful in studies (Meena et al., 2018).

12. A nano-emulsion is a type of mixture where tiny oil droplets are evenly distributed in water, and it's stabilized to keep the droplets from separating. Using co-surfactant and surfactant, a thin layer is applied to the oil droplets to stabilize them physically rather than chemically. Water-in-oil (W/O) and oil-in-water (O/W) are considered as the two varieties of nano-emulsions. Various techniques are used to prepare nano-emulsions, primarily high- or low-energy techniques. Low-energy generated nano-emulsions have greater stability over a two-month period at 4°C and 25°C (Rodríguez-Burneo et al., 2017).

13. Nanobubbles: When subjected to ultrasonic vibrations, stability is maintained by the micro-bubbles at room temperature, but they are collapsed into micro-bubbles with minimal heating. They are typically used for drug delivery and to target tumor tissues directly with the drug (Rapoport et al., 2007). Gene therapy applications also utilize the liposomal nanobubbles (Mohantya et al., 2014).

14. Respirocytes and microbivores: Respirocytes and microbivores present similar functions to red and white blood cells, respectively. Whereas respirocytes are nanorobots designed for efficient oxygen delivery to the tissues while removing the accumulated CO2 through specialized regulatory sensors for very precise control, the microbivores are circulated traps and phagocytose pathogens in the circulatory blood, functioning like macrophages. Their enzymatic digestion results in essential building blocks, such as nucleotides, fatty acids, and amino acids (Mohantya et al., 2014).

15. Micro/nano robots: Nanobots are tiny, computerized, and programmable robots capable of doing a wide variety of jobs, from scanning one's body for malignant cells to introducing nanocameras which provide real-time monitoring during surgery. Freitas remarks that the most recent research in developing cytobots and karyobots is focused on building devices that can work wirelessly inside cells (Freitas, 2005)

16. Aluminum-silicon nanoparticles (NPs), which are silica nanoparticles with a short chain of polyphosphate attached, might be used to help stop bleeding faster by speeding up the body's natural clotting process (Kudela et al., 2015).

The NPs can also be Separated into the following Categories

1. Inorganic nanoparticles (NPs): Due to their rigid structure, inorganic NPs are non-degradable. In the course of their

production, they are capable of producing different shapes. Out of these, the most well-known are silica-based NPs, which have already been proven as biocompatible (Torres-Sangiao et al., 2016; Zhao et al., 2014).

2. Immuno-stimulating complexes: Such complexes consist of antigens to be immunized, associated with supra-molecular particles of the saponin adjuvant Quil A. The presence of hydrophobic interactions renders these complexes competent to fuse antigens with viral envelope proteins. Cage-like structures can capture multiple viral antigens. (Grgacic and Anderson, 2006; Noad and Roy, 2003).
3. These particles measure between 20 and 800 nm. They have a structure similar to that of viruses but lack nucleic acids. While they are not infectious, they elicit an aggressive host immune response (Pushko et al., 2013).
4. Self-assembling systems and proteins: The role of such systems extends to generating quaternary protein complexes of higher order used in vaccinating humans and animals for immunization processes (Kanekiyo et al., 2013).
5. Polymeric nanoparticles: As it has been mentioned above, some of the natural polymeric NPs originated from polysaccharides are inulin and chitosan. They have often been utilized for production of the vaccines like DNA and Newcastle disease vaccines (Fig.2) (Zhao et al., 2014).

(Riley and Vermerris, 2017) proposed an additional classification scheme, grouping the NPs employed in gene transfer into the following categories:

We also covered carbon-based NP and natural- and synthetic-polymer-based nanomaterials, apart from organic and inorganic nanomaterials.

1. Inorganic Nanomaterials

They typically have low cytotoxicity and are biocompatible and reasonably safe. Their distinct optical and electrical characteristics are easily adjustable during the manufacturing process. According to (Erathodiyil and Ying, 2011), this group includes a variety of inorganic materials like gold, silver, iron oxide, and calcium phosphate.

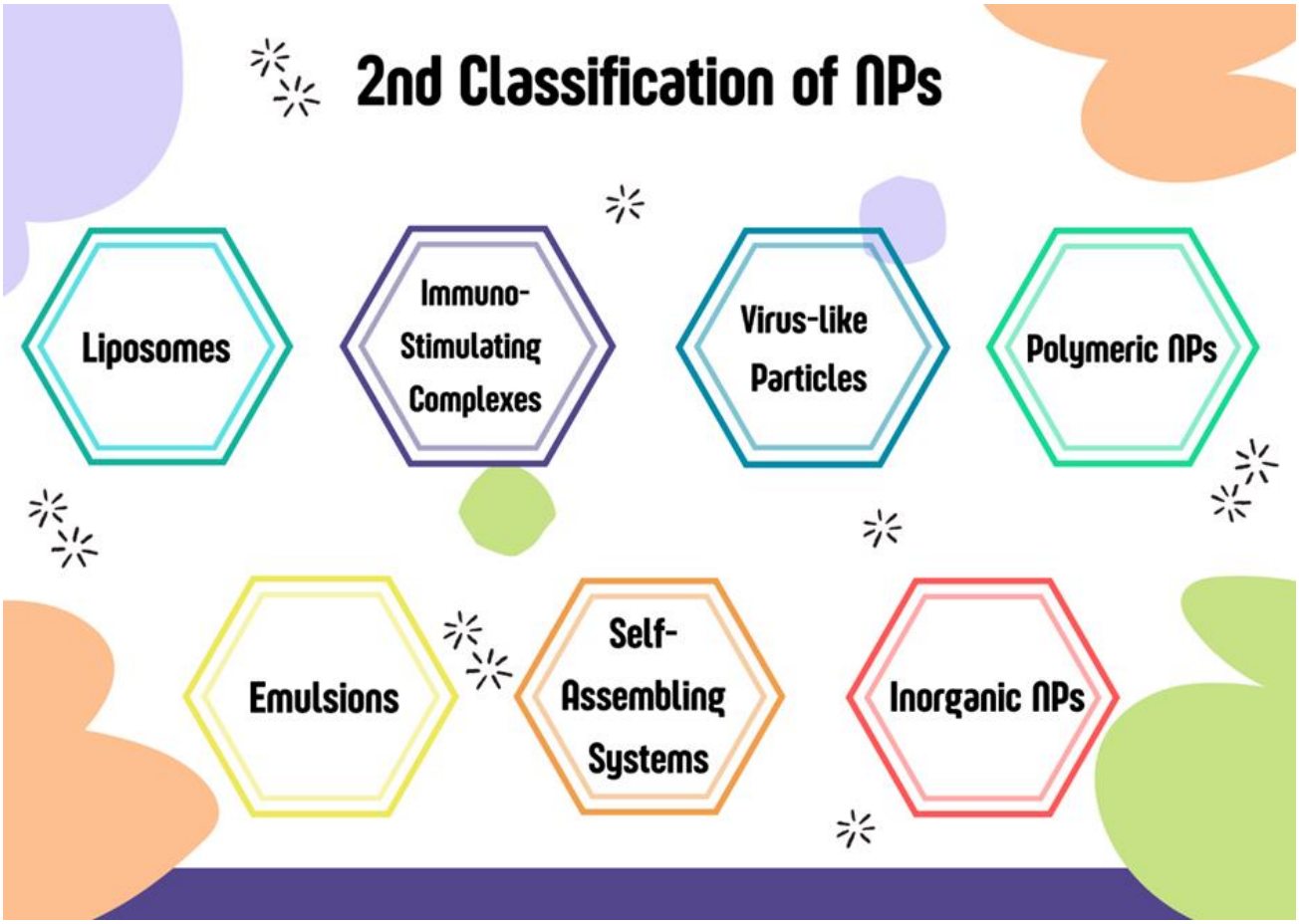

Fig. 2: Second Classification of NPs.

2. Organic Nanoparticles

A) **Proteins and peptide NPs:** They are mostly used in gene transfer experiments due to their good biodegradability, inexpensive cost of manufacture, minimal toxicity, and biocompatibility (e.g., gelatin). They can interact with a range of payloads according to their amphiphilic nature.

B) **Lipid-based nanomaterials:** It sets them apart due to their hydrophobic or amphiphilic character, making them able

to form vesicles and membranes. Catalytic lipids are more effective in delivering genetic elements than any other neutral or anionic lipids because they have the capability to attach to the cell membrane which is anionic in nature (de Ilarduya et al., 2010). Another family of small lipid molecules is lipidoids, which have recently grown further attention for their potential use as siRNA carriers. Research has been done on their possibility of siRNA transport for the therapy of HCV and cancer (Knapp et al., 2016; Moon et al., 2016).

3. **Hybrid NPs:** These include various kinds of nanoparticles; for instance, the polymer-lipid hybrid systems are developed by combining the liposomes with the polymeric NPs. In these NPs, usually the biodegradable and hydrophobic polymer core encases the water-soluble drugs to facilitate the release of drugs in a continuous manner. In order to prevent the reactions of the immune system, a coating of a hydrophilic shell covers the monolayer of lipid on the aforementioned core. This lipid layer regulates the penetration of water into the nanoparticles, thereby controlling the release of the drug contained within them. By modulating how water interacts with the nanoparticle surface, the lipid layer ensures a controlled and gradual release of the drug over time (Prabhu et al., 2015). For the administration of siRNA, further hybrid systems were also created (Bellocq et al., 2003). As with micellar hybrid nanosystems, therapeutic and diagnostic NPs can also be combined to create hybrid nanoparticles. According to (Liao et al., 2011) and (Yang et al., 2010). This approach integrates hydrophobic functional nanocrystals into the core of the nanoparticles for imaging purposes, while hydrophilic therapeutic agents are attached to the outer surface. This configuration allows for effective imaging and targeted therapy, utilizing the distinct properties of the nanocrystals and agents to enhance overall treatment efficacy. Recently, viral hybrid nanoparticles have found an application in monitoring biological functions. Such viruses exhibit a number of advantages that have been artificially redesigned as nanoparticles: uniform size and shape, specificity for delivering therapeutic nucleic acids into target tissues, improved protection of the nucleic acid payload, and the ability to encapsulate other non-nucleic acid drugs within the viral capsid itself (Steinmetz, 2010; Steinmetz et al., 2011). Attachment of magnetic nanocrystals on the capsid surface could enable MRI imaging. Very often, several functions are included in one hybrid nanosystem—for example, in cancer surgery, optical fluorescent quantum dots and superparamagnetic iron oxide NPs. Quantum dots help surgeons see the edges of tumors more clearly during surgery with special glowing images, while iron oxide nanoparticles make MRI scans more detailed, helping doctors find tumors before surgery (Sailor and Park, 2012).

Applications of Nanotechnology in Veterinary Medicine

Thanks to nanotechnology, veterinarians have access to the same range of options as physicians, including advanced disinfectants, tissue engineering, diagnostics, medications, and tissue engineering. The disciplines of animal nutrition, animal breeding and reproduction, and animal health and productivity are already using nano-applications, as Fig. 3 illustrates (Manuja et al., 2012). This technology enables the targeted delivery of drugs to specific cells, allowing for the use of very small doses. This precision means there is less leftover medicine in the animals and less time needed before they can be safely used again (Troncarelli et al., 2013).

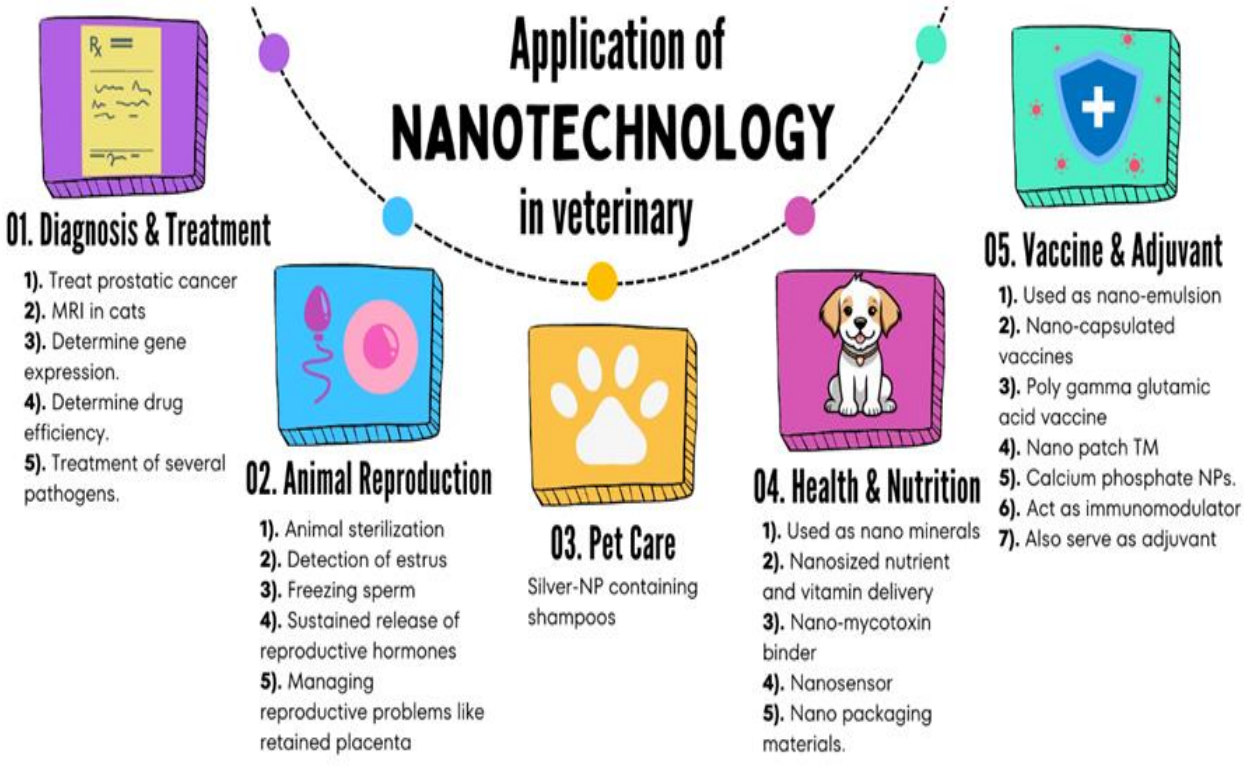

Fig. 3: Application of Nanotechnology in Veterinary Medicine

Use of Nanotechnology in the Detection and Treatment of Animal Diseases

The use of nanotechnology offers a number of innovative solutions for several problems in veterinary medicine, including treatment of infections caused by intracellular pathogens, brucellosis, FMD, and MRSA. Research is underway to exploit the potential of nanodrugs in targeting mastitic udders (Greenwood et al., 2008; Kruubi et al., 2010; Troncarelli et al., 2013). Contrary to the conventional drugs, nanodrugs can be engineered to trigger themselves in the presence of desired conditions only. For example, gentamicin attached to a hydrogel by the action of a peptide linker does not become active until the linker is cleaved by a *Pseudomonas aeruginosa* enzyme. It allows the drug to act only in the availability of *Pseudomonas aeruginosa* (Suzuki et al., 1998; Soppimath et al., 2002). Furthermore, NPs targeted against toxins of bacteria and receptors have been engineered to interact with pathogenic microorganisms in the gastrointestinal tract before they are excreted (Latour et al., 2003; Kim et al., 2010; Underwood and Van Eps, 2012). Other applications of NPs include formulations with nucleic acids or antibodies for easy, accurate, and field-based diagnosis. Nano- and biochips have been found effective in the detection of infections and genetic risk factors. High-density nano-array chips let researchers test many genes, antigens, or disease markers all at once. Protein and DNA-microarrays are used for determining drug efficiency and measuring gene expression. Using improved LOC technology, it is possible now to detect the target protein or DNA in very small sample volumes (Manuja et al., 2012). Apart from that, the NPs are used as the imaging agents in veterinary diagnostics like the MRI scan for cats (Kim et al., 2010; Underwood and Van Eps, 2012).

In the United States, gold nanoparticles replaced conventional invasive modes of treatment for canine prostate cancer. The method has the edge of not being toxic to healthy tissues and the dose it requires for treatment is comparatively much lesser than chemotherapy (Troncarelli et al., 2013).

Nanoadjuvants and Nanovaccines

Nanoparticles are now being used more and more to make vaccines for animals. They boost immune responses by helping the immune system recognize and react to invaders. Nanoparticles can also be used to slow down the release of the vaccine's ingredients, which makes the vaccine more effective (Kim et al., 2010; Underwood and Van Eps, 2012; Awate et al., 2013; Torres-Sangiao et al., 2016). Hence, targeting lymph nodes by antigen-loaded NPs can further enhance the efficacy of vaccination.

Some of the new developments in the area of veterinary nanovaccines are the following:

1. Nano-emulsion vaccines Examples include recombinant influenza viruses and Bacillus anthracis spores, which elicit mucosal immunity after intranasal administration.
2. Some of these targeted pathogens are Helicobacter pylori, Bordetella pertussis, tetanus toxoid, and rotavirus capsid, while others are bovine parainfluenza type 3 that elicited both IgG and IgA immune responses following oral administration.
3. African Horse Sickness Virus Vaccines: Vaccines for African horse sickness are made using a special virus (baculovirus) that helps produce particles similar to the virus. These particles include important proteins from the virus, which help the vaccine protect horses from the disease. These vaccines elicited only a mild immune response, indicating that better vaccine designs are required (Cher et al., 1998; Illum et al., 2001; Conway et al., 2001; He et al., 2002; Greenwood et al., 2008; Florindo et al., 2009; Fernando et al., 2010; Chen et al., 2010; Danesh-Bahreini et al., 2011; Hamouda et al., 2011)

Nanotechnology for the Feeding and Health of Animals

Nanotechnology has some valuable advantages in the animal feed industry with the use of nanominerals. Nanominerals are cost effective, used in smaller dosages, and have the eminent capability to improve immune function and to provide growth potential. They may also help in regulating rumen fermentation and control of pathogens in feed along with various problems related to reproduction in animals (Swain et al., 2015). Examples include nano-ZnO, an immunomodulator that can enhance immune response, growth rates, and reproductive health in poultry and other farm animals. It reduces the incidence of diarrhea in young pigs (Mishra et al., 2014; Yang and Sun, 2006). Studies also indicate that nano-ZnO is able to lower SCC in cows with subclinical mastitis and increase milk production in dairy herds (Rajendran, 2013).

Nanotechnology is also utilized in the production of liquid vitamins for poultry feed, where, due to the nanosize, nutrients are improved in their bioavailability as they pass through a hen's digestive tract. According to Thulasi et al., in 2013, these nano-enabled vitamins enable better nutritional dispersibility, extend the life of the feed, and reduce any undesirable flavoring with reduced use of stabilizers (Thulasi et al., 2013).

Microencapsulation helps protect food constituents from light, oxidation, and digestive enzymes. This technology enables finer dispersion and mixing of fat-soluble additives with various pH levels and temperatures, thus maintaining their stability and extending shelf life (Meena et al., 2018).

Through nanotechnology, newly effective MgO-SiO2 nanomycotoxin binders targeting the common mycotoxin, aflatoxins (normally found most in animal feeds, especially in developing countries), are being created in order to fight mycotoxicosis—a serious condition both in humans and animals (Moghaddam et al., 2010).

Besides, nanomaterials have made a foray into the field of packaging with special features like antibacterial traits—nano-zinc oxide; protection against UV and environmental exposure—nano-titanium dioxide; and high strength factor—nano-titanium nitride. Nanosensors can detect very small amounts of chemical and biological contaminants, making it

easier to find tiny traces of these substances (Manuja et al., 2012).

Animal Reproduction and Nanotechnology

Nanotechnology advances many aspects of animal reproduction, right from diagnosing and treating reproductive disorders to monitoring estrus and managing sperm sorting, freezing, etc. Nano devices can directly influence calving and address the reproductive challenge of retained placenta (Swain et al., 2015).

Moreover, some reproductive hormones are used for prolonged release purposes with nanoparticles. Nanoparticles guard the hormones and vitamins against oxidation, for example, vitamins and steroid hormones as well as hydrolysis of for instance gonadotropic hormones, hence not undergoing degradation thereby remaining effective (A Joanitti and P Silva, 2014).

High sensitivity is achieved with nanosensors that have movable probes made of biomolecules; thus, they are mainly used in medical diagnostics. These nanoscale devices can detect viral infections in the vaginal tract, estrus, and hormonal and metabolic imbalances. When inserted under the skin of cattle, these nanotubes are able to light up, thus indicating estrus (Scott, 2007; Saragusty and Arav, 2011).

Estradiol sensors check hormone levels in the blood and send live updates to a computer to keep track of livestock health. Nanocapsules with bull semen can be directed to fertilize eggs, and nanotechnology helps sort sperm and eggs. Biochips are also being created to determine the sex of the fetus, making it possible to choose the calf's gender (Patil et al., 2009).

Using Nanotechnology in Pet Care

The pet health care market is on the rise in a world where nanotechnology is playing a vital role in coming up with new products for pets. With nanotechnology, it enhances the functionality of surface deodorizers and disinfectants. In the area of pet care, one of the key products developed from this technology is shampoos containing silver nanoparticles (Troncarelli et al., 2013) (Sharif et al., 2024).

Security

While most NPs are considered safe, there are still a few risks involved in their use. For example,

1. Workers in the Pharmaceutical Industry: Prolonged exposure to carbon nanotubes, specifically through the route of inhalation, may cause reproductive abnormalities (Johansson et al., 2017).
2. Risks of injury because of the accumulation in the body of magnetic iron oxide nanoparticles or from unstable interactions between therapeutic agents and nanoparticles could occur, in which the therapeutic agent is released at times in areas of the body other than intended. This can lead to toxicity of the healthy tissue involved and also result in an insufficient therapeutic dose at the desired site.

Nanoparticle applications can also have disastrous consequences on the body and the environment. For example, the high demand for radionuclides could be hazardous or the depletion of the ozone layer by carbon nanofibers, which cannot be ruled out. The ability of nanoparticles to cross all biological barriers, such as the BBB, adds to the concerns (Manuja et al., 2012; Mohantya et al., 2014; Wu et al., 2018).

Conclusion

Recent improvements in nanoparticle design have created many options for targeted medical treatments. Nanotechnology has greatly improved veterinary medicine, making it better for diagnosing and treating animals, creating vaccines, and improving their nutrition and overall health.

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