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Popular Article

Nanotechnology-Based Therapeutic Approach in Veterinary Medicine: An Adjunct to Conventional Treatment

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Abstract

Nanotechnology is an emerging area of research interest in the present era. A number of nanomedicines has been by approved by United States Food and Drug Administration (FDA or USFDA) for clinical use in humans. Nanomedicine also has potential applications in veterinary medicine which can act as an adjunct to the conventional treatment available. Nano drug delivery system offers benefits such as low particle size, decreased drug dosage, increased solubility and bioavailability, reduced side effects and degradation of drug. There are number of nanocarriers developed overtime such as liposomes, nano emulsions, micelles, lipid nanoparticles, polymeric nanoparticles, metallic nanoparticles, dendrimers and others. The nanocarriers have proved to be beneficial in administration of drugs which are highly hydrophobic or have large particle size in a sustained release manner which thereby decreases the risks of toxicity and increases the therapeutic potential of the drug. In conclusion, the use of nanomaterials in veterinary medicine can produce beneficial effects and thus improve animal health and production

Introduction

Pet and food-producing animal populations have been gradually increasing around the world. In the present times, the field of nanotechnology has become an emerging scientific area of importance. Nanotechnology plays an important role in the development of new materials and tools that aid in the enhancement of animal health and output. In veterinary medicine, the production of effective and safe products is highly required which upon administration to the animal produces minimum pain and adverse effect which is a priority of pet owners. In the case of livestock products, ensuring the residual number of drugs is at a safe minimum level is of great importance to the veterinary pharmaceutical industries (Hill and Li, 2017). However, the drugs used for veterinary patients are similarly designed as in human medicine, regardless of the difference in the biochemical, anatomical and physiological systems between the species. For the sake of animal health and welfare, it is highly required to develop novel pharmaceutical products having the potential to enhance animal health and livestock production. Therefore, along with the conventional treatment, a new strategy has to be developed to design new products veterinary, justifying the interest nanotechnology as effective therapeutics in veterinary medicine.

The applications of nanotechnology include improved disease diagnosis and treatment, enhanced drug delivery and sustained release of the active component, increases the solubility of hydrophobic agents, reduces drug dosage and degradation of the active constituents. In order to highlight the importance of nanotechnology in veterinary medicine, the present article covers the aspect of nanomedicines, different types of nanomaterials of pharmaceutical importance and their application in various ailments.

Nanomedicine

The medicinal products which are synthesized or produced by using nanocarriers or nanomaterials are termed as nanomedicine (Tinkle *et al*., 2014) and the nanoparticles used to synthesize nanomedicine are called as nanocarriers. Nanoparticles have hydrodynamic diameter less than 1000 nanometers (nm) and their diameter have greater impact on their physical and chemical interactions with the biomolecules (Kreuter, 2007). In case of conventional therapy, due to the low bioavailability or rapid metabolism of drug, a high dose is required to produce the desired pharmacological effect which increases the risk of side effects or toxicity. Nano-compartmentalization provides benefit over conventional therapy as the time-release profile and other pharmacokinetic parameters of the therapeutic agent can be modulated without any change in the pharmacological properties of the drug (Suri *et al*., 2007). Nanomedicine includes the development of nanoparticles capable of targeted delivery of active biomolecules, synthetic drugs, nutrients, immunotherapeutic agents and natural bioactive compounds (Lombardo *et al*., 2019). The nanocarriers have two types of system i.e. reservoir

or matrix system. In the case of the reservoir system, the nanoparticles have an aqueous or oily core in which the therapeutic agent is trapped (Fig. 1A), while in the matrix system, the therapeutic agent is dispersed throughout the nanocarrier's matrix (Fig. 1B). For targeted drug delivery, the functionalization of the nanoparticle surface is carried out in which the therapeutic agent is attached to the surface of the nanoparticle (Fig. 1C) (Rani and Paliwal, 2014). Nanomedicine is targeted to the specific target in two ways: active and passive (Fig. 2). In passive nano-delivery, there is accumulation of drug at the specific site while in the active process the surface of the nanoparticle is tagged with a specific marker or therapeutic agent itself which specifically interact with the receptor at the target site (Aiacoboae *et al*., 2017). Another system of nanomedicine is the stimulus responsive nanoparticles which release the active drug in presence of a stimuli such as pH, temperature, light, electric or magnetic field, and electrolytes (Sharma *et al*., 2015). The present article addresses the nanocarriers which can/are used to synthesize nanomedicines of veterinary importance.

Nanocarriers

Number of nanocarriers are used for the delivery of drugs, as of veterinary importance, the nanocarriers which are used and addressed in the present article include liposomes, micelles, lipid nanoparticles, nanoemulsions, polymeric nanoparticles, dendrimers and metallic nanoparticles. With the help of these nanocarriers, reformulation of the traditional dosage forms can be done. Many therapeutic agents have great pharmacological potential, but due to their large particle size and toxic effect, the dosage of that particular drug is reduced, leading to a decrease in its therapeutic effect. On the other hand, some drugs are highly hydrophobic in nature and insoluble in aqueous solution and require toxic solvents for its administration. Nanocarriers have proved to be beneficial in the administration of such drugs. One such example is the Abraxane®, approved by the FDA in 2005. The anti-tumor drug paclitaxel is capable of inducing an anaphylactic reaction in susceptible patients, while the nanoparticle albumin-bound paclitaxel formulation has proved to be more tolerable than the conventional paclitaxel therapy (Caster *et al*., 2017). The principal objective of using nanocarriers in Veterinary medicine is that it enables the delivery of therapeutic agents with dose reduction, decreased risk of adverse reactions or toxicity and discomfort in chronic therapy.

Potential application of different nanocarriers in Veterinary medicine

Liposomes

Liposomes have a double lipid layer consisting of phospholipids and have a varying hydrodynamic diameter ranging from 25nm to 1000nm. Liposomes are useful in the delivery of both hydrophilic and lipophilic compounds; its surface charge and size can be modulated easily along with its functionalization capacity.

This nanoparticulate system is widely described and used in the veterinary application. B and T cell epitope peptides were entrapped in the liposome having hydrodynamic diameter of 127-141nm and zeta potential of -25.1 to -31.8 millivolts (mV) were used as vaccine against the influenza virus (H1N1) in pigs (Dhakal *et al*., 2018). The liposomes improved haemagglutination, IgA response and protected from adverse effects caused by influenza A virus H1N1, such as fever and lesions. There are reports where liposomes were used as a nanoparticulate system for the treatment and prevention of tumors.

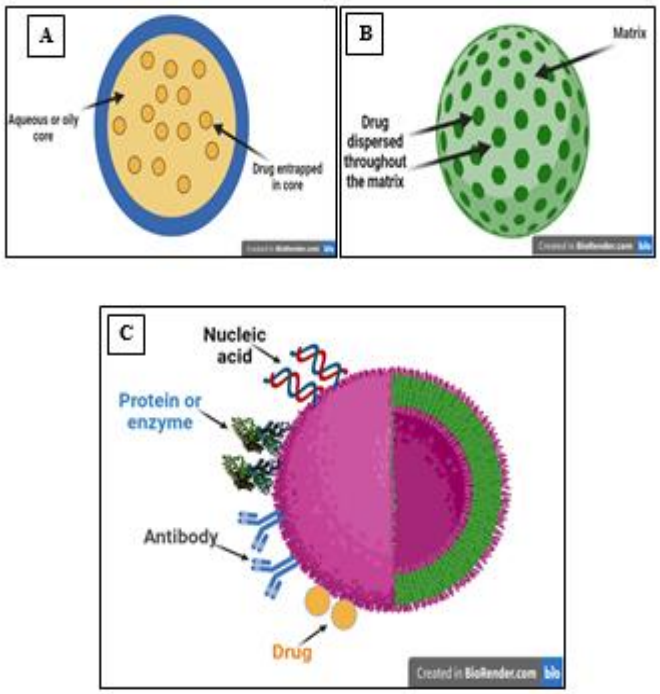

Fig 1. Nanoparticulate system: A. Reservoir system, B. Matrix system, C. Functionalization of the nanoparticle surface [Pictorial representation is designed manually by using an online tool (Biorender)]

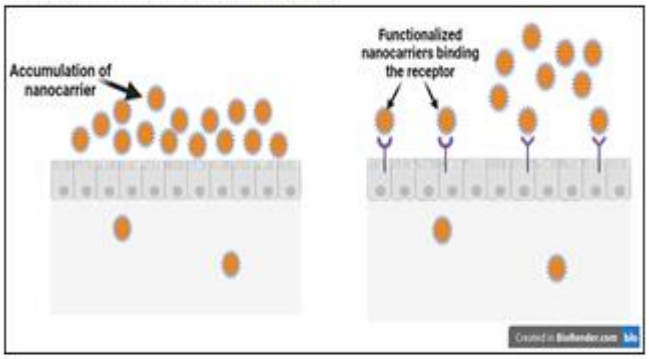

Fig 2. Nano-delivery system: Passive process (left), Active process (Right) [Pictorial representation is designed manually by using an online tool (Biorender)]

Curcumin was encapsulated in liposome and its effect was studied against naturally induced canine cancer. The liposomes exhibited good anti-tumor effect in *in vitro* as well as in *in vivo* studies (Withers *et al*., 2018).

Nanoemulsion

Nanoemulsion is a nanoparticulate system which consist of at least oil, water and an emulsifier and have the hydrodynamic diameter ranging from 50nm to 1000nm (Salvia-Trujillo *et al*., 2017). It consist of colloidal dispersion of droplets which are kinetically stable but thermodynamically unstable (Gupta *et al*., 2019). Nanoemulsion for canine cancer chemotherapy was synthesized (Lucas *et al*., 2015). Carmustine containing lipid nanoemulsion was used for canine lymphoma therapy using ultrasonication for reducing the size of the nanoparticles. Another study reported use of photodynamic therapy along with the aluminium-chloride-phthalo-cyanine nanoemulsion for the treatment of cutaneous hemangiosarcoma in dogs which lead to complete disappearance of tumor (Rocha *et al*., 2019). A study reported anesthetizing tilapia fish by using clove oil nanoemulsion having hydrodynamic diameter of 64 ± 1.0nm and it was observed that nanoemulsion enhanced the efficacy of clove oil as compared to eugenol ethanolic solution with respect to anesthesia of tilapia (Kheawfu *et al*., 2017).

Solid lipid nanoparticles (SLN)

Solid lipid nanoparticles have hydrodynamic diameter ranging from 50nm to 1000nm and are used as an alternative system to liposomes (Muller *et al*., 2011). SLN consist of solid lipids and surfactants are used as stabilizer. In contrast to nanoemulsion, the SLN have solid lipid replacing the liquid lipid used in nanoemulsion. This improves the stability and release profile of the SLN under severe environmental condition (Gordillo-galeano and Mora-huertas, 2018). For the treatment of leishmaniasis, oryzalin has proved to be a promising drug, but due to its less bioavailability and toxicity, it is not well tolerated in animals. To overcome this, a study reported development of SLN encapsulating oryzalin which consisted of mixture of soy lecithin, tween® 20 and sodium deoxycholate as surfactant and tripalmitin as solid lipid and the synthesized SLN had hydrodynamic diameter of less than 140 nm and zeta potential of -35mV with encapsulation efficiency of more than 75%. The cytotoxic effect of this SLN was analysed revealing that the nanoparticulate system allowed increased tolerability of the drug (Lopes *et al*., 2012).

An important Veterinary application of SLN is the delivery of antibiotic for the treatment of mastitis. Tilmicosin was encapsulated in SLN containing hydrogenated castor oil, which enhanced activity of tilmicosin with reduction in side effects related to tilmicosin (Wang, 2014; Ling, 2016). The SLN has proved to be cost effective as the tilmicosin encapsulated SLN were incorporated into a sodium alginate chitosan nanogel used for the treatment of the cow mastitis, such formulation required low concentration of the drug and had long post antibiotic effect (Zhou *et al*., 2019).

Nanostructured lipid carriers (NLC)

Nanostructured lipid carriers are similar to the SLN consisting of a structural solid lipid and stabilized by a surfactant which is amphiphilic in nature (Garcês *et al*., 2018). However, there is a minor difference between NLC and SLN, as NLC contains solid lipid which contains a small fraction liquid lipid which enhances more accommodation and encapsulation of the drug by the NLC and prevents the untimely release of the drug (Garcês *et al*., 2018). Nanotoxicological studies have shown that both SLN and NLC are biodegradable and have greater biocompatibility (Doktorovova *et al*., 2014). Buparvaquone was encapsulated in the NLC. The NLC were developed using Softisan® 154 and Miglyol® 812 as lipids and surfactants used were Kolliphor P188 as well as tween 80. High-pressure homogenization method was used for the synthesis of NLC, forming nanoparticles having hydrodynamic diameter around 350nm and encapsulation efficiency close to 100%. Another study involved synthesis of functionalised NLC, where the surface of NLC was modified with chitosan and dextran to co-deliver buparvaquone and polymixin B against *L. infantum*. The functionalisation and co-delivery enhance leishmanicidal activity in *in vitro* test up to 3-fold compared to free buparvaquone (Monteiro *et al*., 2019).

Micelles

Micelles are particulate system which consist of the polymers which are amphiphilic in nature. Micelles having anti-leishmanial activity were developed in which amphotericin B was encapsulated in the pluronic F127 micelles coated with chitosan. The hydrodynamic diameter of the synthesized micelle was 102.23 ± 11.14nm with encapsulation efficiency around 60%. The micelles were 21.97 times more internalized by macrophage as compared to the free drug in flow cytometric studies (Singh *et al*., 2017). Pluronic F127 encapsulated bilirubin nanoparticles were synthesized to enhance the wound healing activity in Wistar rats (Kamothi *et al*., 2022). The synthesized bilirubin nanoparticles had hydrodynamic diameter ranging from 100-150 nm and showed greater wound healing activity than the free drug by stimulating the expression of VEGF, TGF-β and IL-10 levels.

Polymeric nanoparticles

Polymeric nanoparticles can be prepared using number of polymers which are either natural, semi-synthetic or synthetic in nature (Sur *et al*., 2019), having hydrodynamic diameter ranging from 1 to 100nm and the polymeric nanoparticles are further divided into two types i.e. Nanocapsules and nanospheres (Ferreira *et al*., 2018). Natural polymers consist of chitosan, alginate, hyaluronic acid, dextran, carboxymethyl cellulose and pectin (Ferreira *et al*., 2018) and are biodegradable, biocompatible and non-toxic in nature (Jin *et al*., 2019). A study reported the synthesis of platin-M encapsulated polymeric nanoparticles for the treatment of canine brain tumors (Feldhaeusser *et al*., 2015). The nanoparticles exhibited a good anti-cancerous effect by disrupting mitochondrial energy production in glioblastoma and canine glioma cell lines.

To treat and prevent the diseases related to poultry breeding, sodium alginate-polyvinyl alcohol was used to encapsulate amoxicillin having hydrodynamic diameter of 513nm and surface charge of -45mV encapsulating around 43% of the drug. The amoxicillin encapsulated sodium alginate-polyvinyl alcohol nanoparticles exhibited increased bioavailability and plasma half-life as compared to the free drug, thus increasing the mean residence time in the intestinal and circulatory system (Güncüm *et al*., 2018).

Another study reported a cream prepared by synthesis of nano composite consisting of chlorhexidine, calcium phosphate nanoparticles mixed with polyethylene glycol polymer for wound healing (Viswanathan *et al*., 2016).

Metallic nanoparticle

Metallic nanoparticles are mainly used in biosensing, bioimaging, drug delivery, gene delivery and cellular labelling (McNamara and Tofail, 2017). Metal nanoparticles mainly include silver, iron oxide, gold, zinc and copper nanoparticles. These are mainly used as antimicrobial and antiviral agents in veterinary. In a study, silver nanoparticles were synthesized to evaluate its antimicrobial effect against drug-resistant strains of *Pseudomonas aeruginosa* and *Staphylococcus aureus* isolated from mastitis-infected goats (Yuan *et al*., 2017). The synthesized nanoparticles had hydrodynamic diameter of 10-50nm and surface charge of 37.7mV and exhibited anti-bacterial effect by generating ROS against bacterial cells.

Dendrimers

Dendrimers are branched structure consisting of number of functional groups allowing the delivery of hydrophilic or hydrophobic therapeutic agents, internalized or attached to the surface. Dendrimer based classical swine fever vaccine was developed and it was observed that the vaccine elicited better immune response against the virus due to the inclusion of dendrimer (Tarradas *et al*., 2012).

In a study, generation six dendrimer cyanine 5 conjugates were prepared targeting hypothermic circulatory arrest (HCA) induced brain injury in a canine model (Grimm *et al*., 2016). It was found that the administration of dendrimer cyanine 5 conjugates systemically leads to penetration of blood brain barrier by the conjugates and its accumulation in areas of the brain most affected with HCA.

Conclusion

In the present era, there is a need for the development of novel pharmaceutical products for Veterinary medicine. The field of nanotechnology holds great potential for the synthesis of novel formulations which can enhance animal health and productivity. Nanomedicine can be used along with

the conventional treatments available in order to enhance the efficacy of the therapy and decrease the time required for recovery of the animals, thus improving animal health and livestock production.

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