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Nanotechnology, an alternative to prevent and treat bovine mastitis

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ABSTRACT

Objective: to describe the importance of nanoparticles in the prevention, diagnosis and treatment of bovine mastitis.

Design/Methodology/Approach: nanotechnology in the field of veterinary medicine has revolutionized scientific research. As it aims to access a new era in the diagnosis and treatment of disimile diseases such as bovine mastitis. In regard to it, this disease was characterized in its most common origin and diagnosis. Then, we examined state-of-the-art papers on the use of nanoparticles as an alternative treatment to verify results and effectiveness.

Results: reviewed reports on the use of nanotechnology as treatment did show favorable, faster and more effective results. Also, it is discussed how nanotechnology has innovated the field of prevention of this disease that can occur in cattle in a clinical or subclinical form, in order to offer innovative solutions to current problems, such as the prevention, diagnosis, and treatment of this common disease in dairy cattle.

Limitations/implications of the study: despite its significant importance in the treatment of bovine mastitis. applied nanotechnology still has certain limitations to be noted. These include the lack of a single, standardized dose; doses used must be tested through clinical studies assessing some aspects such as nanoparticle type, drug load, and infection severity. In addition to other aspects like toxicity, distribution, bioavailability, immunological interactions, and costs.

Findings/Conclusions: the use of nanoparticles in veterinary medicine as treatment of bovine mastitis allows transforming delivery vehicles and assimilation paths for drugs and vaccines, which enhances treatment effectiveness. Therefore, the use of nanomaterials positively impacts animal health and well-being.

Keywords: bovine mastitis, nanotechnology, veterinary medicine.

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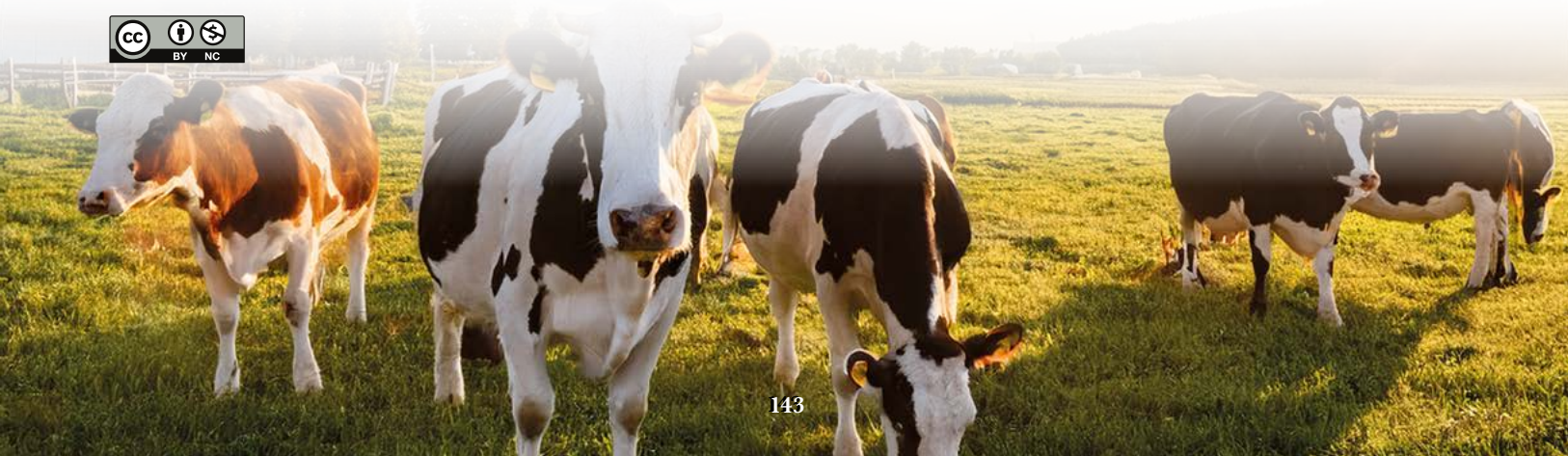
INTRODUCTION

Milk is one of the most sought-after products worldwide due to its nutritional properties and the variety of derivatives obtained from it. However, one of the main challenges facing its production is the incidence of diseases such as bovine mastitis.



In the production chain of bovine milk, mastitis is considered the most important economic and productive disease (Alfonso *et al.*, 2020). Because it causes losses related to decreased milk yield, increased clinical treatments for animals, and the culling of affected cows (Bedolla *et al.*, 2020). This disease also produces changes in the protein and lipid composition of milk that affect its quality (Aguilar and Álvarez, 2019). For this reason, it can also threaten the health of consumers through the presence of pathogenic microorganisms in the milk, when is obtained from cows affected by the disease.

In preventing mastitis, proper hygiene and good husbandry practices during milking are essential factors. Antimicrobials are used as the drugs of choice to achieve its control. However, some new, more sustainable and environmentally friendly alternatives have recently emerged, such as the use of nanoparticles.

Based on the above, this paper aims to present the latest advances in this pathology and the new strategies proposed for its prevention and control. The objective was to describe the importance of nanoparticles in the prevention, diagnosis and treatment of bovine mastitis.

Bovine mastitis

Mastitis is the inflammation of the mammary gland (udder) and its secretory tissues. Mastitis incidence decreases milk production, alters the composition and flavor of this product; it also increases the normal bacterial load (Gasque, 2015), in addition to affecting animal well-being (Medrano *et al.*, 2020). It is classified according to its symptoms into clinical and subclinical mastitis (González y Vidal del Río, 2021). Clinical symptoms include increased volume, redness, and pain in the mammary gland. Fever, loss of appetite, and weakness may be seen. Alterations in milk quality such as lumps, changes in color, and an increase in bacterial content are also observed. In the subclinical form, no apparent changes are observed in the udder or in the milk. However, milk production decreases and its composition is altered by the pathogens that may be present (Córdova *et al.*, 2019).

Etiology

The causes of this pathology are related to aspects of cow welfare, health, and hygiene (Córdova *et al.*, 2019). Infectious sources (Benić *et al.*, 2018; Bedolla *et al.*, 2020) and non-infectious sources, such as injuries, high ambient temperatures and stress in animals, are cofactors that cause the incidence of bovine mastitis (Brisuela *et al.*, 2018).

Mastitis is classified according to its causative agents as contagious or environmental. The infected mammary gland is the primary reservoir for contagious agents (García *et al.*, 2018). These infectious source includes bacteria such as staphylococci, streptococci, or coliforms. As well as mycoplasmas, fungi, and yeasts, all of which are pathogens responsible for mastitis (Zoetis, 2023). In studies, contagious and environmental agents have been isolated as causes of the disease. Among those, the following are oustanding, *Staphylococcus aureus* and *Enterobacter* spp. (García *et al.*, 2018); staphylococci coagulase-negative (SCN) and enterobacteria. (Alfonso *et al.*, 2020); *Bacillus* spp., *Streptococcus agalactiae*, *Streptococcus* sp., *Corynebacterium bovis* and *Escherichia coli* (Rodríguez & Espinosa, 2020; Valdivia *et al.*, 2022).

Diagnosis

Different methods are used to detect mastitis, such as the evaluation of physical and chemical changes in milk through conventional bacteriological methods and molecular techniques. Other tools are currently used, ranging from field techniques to sophisticated applications such as biosensors, microchips, and nanotechnology (Aguilar and Álvarez, 2019; Ajose *et al.*, 2022).

Nanotechnology

Nanotechnology (NT) refers to structures whose size is measured in *nm*, which is equivalent to the billionth of a metre (1×10^{-9} m), this is, the millionth of a milimitre (1×10^{-6} mm). Within this scale, nanomaterials and nanostructures are found between 0.1 to 100 nanometers (Cervera-Villaseñor *et al.*, 2023).

Following the contributions from different disciplines, such as biology, chemistry, physics, engineering, and medicine, nanoparticles evolved in recent years and strategies are combined that allow the design, synthesis, and manufacture of nanomaterials (Lira *et al.*, 2018). In fields such as the diagnosis, control, and treatment of diseases, research with these structures has demonstrated great potential (Ianiski *et al.*, 2022).

Nanotechnology is a scientific and technological advance that has an important application in different fields of knowledge. In livestock industry, in particular, NT is considered one of the most influential technologies of the 21st century (Feugang, 2019).

Classification of nanoparticles

Nanoparticles are classified according to their origin and the materials they are made of. According to their origin, there are naturally occurring nanoparticles, which are made of organic or mineral materials, and anthropogenic nanoparticles, which are produced by human operations during industrial processes (Frejo *et al.*, 2011).

Regarding the materials that compose them, there are different types; carbon-based nanoparticles , which are mostly made of this material. Metal-based nanoparticles, which are composed of different heavy metals, can in turn be grouped into four categories, metallic nanoparticles (0D), metallic nanowires and rods (1D), metallic sheets and plates (2D), and metallic nanostructures (3D). Dendrimer-based nanoparticles, which are formed by synthetic polymer macromolecules (such as peptioides, lipoids or polysaccharide-modified particles). Finally, composite nanoparticles, which are combinations of similar nanoparticles or nanoparticles mixed in larger materials (Zhang *et al.*, 2016).

Among the most commonly used nanomaterials are metallic nanoparticles such as silver (AgNPs), gold (AuNPs), platinum (PtNPs), copper (CuNPs), selenium (SeNPs); or based on iron oxide (Fe_2O_3NPs), titanium dioxide (TiO_2 NPs), zinc oxide (ZnONPs), among others (Arana *et al.*, 2021).

Nanotechnology in veterinary medicine

Nanotechnology is notable for its potential to revolutionize the veterinary science sector, covering different areas ranging from animal breeding and reproduction, in the field of

animal nutrition, as well as disease diagnosis and the administration of drugs and vaccines (Ali *et al.*, 2021).

For the treatment of infectious diseases of bacterial, fungal (Hassan *et al.*, 2015), parasitic or viral origin (Bogdanchikova *et al.*, 2016) and non-infectious neoplasias (Wójcik *et al.*, 2015), a wide range of metallic nanoparticles are evaluated in veterinary medicine. Research on these nanoparticles is focused on improving anti-inflammatory responses and the wound healing process; As well as in vaccines development, drug delivery, innovation of diagnostic methods for biomolecules detection (DNA, lipids, proteins, metabolites), and for the identification of pathogens and adulterants in food (Kuswandi *et al.*, 2017; Yaqoob *et al.*, 2020).

Nanoparticles in the prevention, diagnosis, and treatment of bovine mastitis

Silver and copper nanoparticles (AgNPs, CuNPs) have shown to have an effect against *Staphylococcus aureus*, *Escherichia coli*, *Streptococcus uberis*, *Candida albicans* and *Candida krusei*, in evaluations with Au, Ag, Cu and Pt nanoparticles, which were tested *in vitro* against microorganisms involved in bovine mastitis (Wernicki *et al.*, 2014). One of the most notable advantages of nanobiotechnology in disease diagnosis is the ability to detect specific biomarkers with high sensitivity and specificity. For the capture and detection of molecules that indicate the presence of diseases in milk samples, nanomaterials (carbon nanotubes, metallic nanowires, and polymeric nanomaterials) can be functionalized with antibodies or other biological recognition molecules (Martínez & Mera, 2024).

Another important advantage is the possibility of developing multiplexed tests, which can reveal multiple mastitis biomarkers in a single sample, providing more specific information about the type of current infection and the severity of the infection. This is beneficial because the precise determination of the causative agent is essential for effective treatment, since there are a variety of bacterial pathogens that can cause mastitis (Singh *et al.*, 2017).

In addition to their high sensitivity and specificity, techniques and devices based on nanobiotechnology provide other significant benefits in the diagnosis of bovine mastitis. For example, some of these methods are fast and portable, which supports a better clinical decision-making and reduces the time required to obtain results (Jensen, 2016; Elsanad, 2020).

Administration of drugs and vaccines

To achieve direct drug delivery to the site of infection, nanotechnology enables encapsulation and controlled release. For the reduction of frequent dosing, antibiotic-loaded nanoparticles easily penetrate the affected tissues, increasing local drug concentration. This minimizes systemic drug exposure, which can reduce bacterial resistance and side effects (Mahendra *et al.*, 2017).

One of the innovative therapies for the treatment of bovine mastitis is nanoparticles loaded with anti-inflammatory agents or immune system modulators that can help reduce inflammation and cause the recovery of the affected mammary gland (Cuca, 2018). An example of this is silver nanoparticles (AgNPs), which penetrate the bacterial cell

membrane, alter its function and cause its death. These are used as delivery vehicles for antibiotics, increasing their efficacy and reducing bacterial resistance. They can be used in concentrations between 10-50 50 μg mL^{-1} (Chen, 2022).

On the other hand, gold nanoparticles (AuNPs) have anti-inflammatory and antioxidant properties. By modulating animal immune response and reducing the production of inflammatory mediators, AuNPs decrease inflammation in the mammary gland, improving the bioavailability and efficacy of drugs by acting as vectors for their delivery (Elsanad, 2020).

Likewise, polymeric nanoparticles (nanohydrogels and polymeric micelles) are used as drug delivery systems. They can encapsulate antibiotics and release them in a controlled manner at the site of infection, extending their therapeutic action and reducing frequent dosing (Sun *et al.*, 2019).

Also, lipid nanoparticles (nanoemulsions and liposomes) are used for drug delivery in the treatment of bovine mastitis, the dose is 1-10 mg of nanoparticles per affected quarter in the udder. This varies varies according to the type of encapsulated drug and the specific formulation. They can encapsulate hydrophilic and lipophilic drugs, making them versatile in the administration of different types of antibiotics and therapeutic agents. In addition, they improve the solubility and stability of drugs, increasing their bioavailability and efficacy (Chen, 2022).

Finally, magnetic nanoparticles are a promising strategy for the treatment of bovine mastitis. These nanoparticles are functionalized with specific molecules that recognize and bind to bacterial pathogens. Using external magnetic fields, they can be selectively directed to the site of infection, improving local drug concentration and reducing systemic exposure (Buzea *et al.*, 2007; Martínez & Mera, 2024).

CONCLUSIONS

Bovine mastitis is a disease that affects dairy herds, causing significant economic losses to the dairy sector. In addition to adopting good practices and maintaining adequate hygienic conditions during the milking routine, innovative and sustainable alternatives are currently being used to prevent, diagnose, and treat this disease.

The use of nanoparticles in the diagnosis and treatment of bovine mastitis offers an innovative approach. They are an important alternative for the prevention of this disease and also enable the development of a new drug delivery route. Therefore, further research is needed in this area, which has proven to be of great importance in veterinary medicine.

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