Use of Nanoparticles in Elimination of Ectoparasites in Companion Animals

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ABSTRACT

Companion animals, including pets and working animals, are susceptible to various ectoparasites. The companion animals are mostly affected by fleas, lice, mites, ticks, and flies. These parasites present a significant challenge for the health and well-being of animals. These parasites also pose the zoonotic risk by spreading various zoonotic pathogens from animals to humans. The use of different chemical (organophosphates, carbamates, pyrethriods, macrocyclic lactones etc.), mechanical (grooming) and environmental methods (biological control, sanitation, and environmental modifications) are considered as traditional methods in control of ectoparasites. However these control methods exhibit limitations such as variable efficacy and toxicity concerns. Similarly, the repeated use of these acaricides has led to environmental pollution, resistance development and increase in the treatment cost. The promising approaches to ectoparasite management can be found in the growing field of nanotechnology. Owing to their small size, nanoparticles provide opportunity for the targeted distribution and continuous release of active substances. Different metallic and polymeric nanoparticles employs mechanisms such as cell membrane damage and oxidative stress induction against ectoparasites. Various formulations, including topical treatments, environmental applications, and controlled-release formulations demonstrate effectiveness in ectoparasite elimination. Overall, nanoparticle-based ectoparasite control shows promise in providing safer, more effective, and sustainable solutions for companion animal healthcare, enhancing their health and well-being globally.

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INTRODUCTION

Companion animals, ranging from beloved household pets to essential working animals, play a significant role in the lives of millions of people globally (Overgaauw et al., 2020). Ensuring their health and well-being is vital, yet they are often affected by a variety of ectoparasites, including fleas, ticks, mites, and others. These ectoparasites not only cause discomfort and irritation to the animals but also pose significant health risks, including the transmission of diseases to both animals and humans (Rafiqi et al., 2016). The current methods of ectoparasite control, such as topical and oral medications, environmental management, and mechanical methods like grooming, although effective to some extent, are not without limitations. These limitations include variable efficacy, potential toxicity, and the development of resistance among ectoparasites, presenting ongoing challenges in the management of ectoparasitic infestations (Leone and Han, 2020). In recent years, the emerging field of nanotechnology has offered promising solutions to these challenges. Nanoparticles, defined as particles with dimensions typically ranging from 1 to 100 nanometers, possess unique properties that make them highly suitable for applications in animal healthcare. With their small size and large surface area, nanoparticles offer enhanced capabilities for targeted delivery, sustained release of active ingredients, and improved efficacy in ectoparasite elimination (Najahi-Missaoui et al., 2020).

This chapter aims to explore the potential of nanoparticles in ectoparasite control in companion animals. This chapter will delve into the properties and mechanisms of action of nanoparticles, categorize different types of nanoparticles being explored for this purpose, and discuss the advantages and limitations of each type. Furthermore, it will examine the factors influencing the effectiveness of nanoparticle-based treatments and explore their application in various formulations, including topical treatments, environmental applications, and controlled-release formulations. By shedding light on the

innovative potential of nanotechnology in ectoparasite control, this chapter seeks to pave the way for the development of safer, more effective, and sustainable solutions for the management of ectoparasitic infestations in companion animals, ultimately enhancing their health and well-being.

Types of Ectoparasites in Companion Animals

Companion animals are affected by ectoparasites such as fleas, ticks, lice, and mites, which can lead to various health issues. Fleas, belonging to the order *Siphonaptera* with over 2500 species (Bourne et al., 2018), include cat fleas (*Ctenocephalides felis*) and dog fleas (*Ctenocephalides canis*) that transmit bacterial pathogens like *Bartonella* and *Rickettsia* (Durden and Hinkle, 2019). They can cause diseases like flea-borne spotted fever, murine typhus, and cat scratch fever (*Bartonella henselae*) (Rust, 2017), as well as flea allergy dermatitis (FAD) (Farrell et al., 2023). Fleas also carry *Mycoplasma haemoplasma* species causing hemolytic anemia (Bourne et al., 2018) and can transmit *Yersinia pestis*, the plague bacterium (Rust, 2017). Additionally, fleas are intermediate hosts for the tapeworm *Dipylidium caninum*, causing pruritis and digestive issues in pets (Bourne et al., 2018). Ticks are haematophagous invertebrates with over 900 species in the families *Ixodidae* and *Argasidae* (Boulanger et al., 2019). Species like *Rhipicephalus sanguineus*, *R. haemaphysaloides*, and *Haemaphysalis longicornis* affect pets (Nelder et al., 2021), transmitting diseases such as Lyme disease, Q fever, spotted fever rickettsioses, and babesiosis, and can cause tick paralysis and allergic reactions (Kopsco et al., 2021; Buczek et al., 2020).

Lice are wingless insects in the orders *Mallophaga* (chewing lice) and *Anoplura* (sucking lice) (Shiferaw, 2018). Dogs are infested by *Trichodectes canis*, *Linognathus setosus*, and *Heterodoxus spiniger*, while cats are infested by *Felicola subrostratus* (Little, 2021). Infestations cause dermatological lesions, anemia, and alopecia due to scratching and biting (Kumsa et al., 2019). Mites belong to orders *Astigmata* and *Prostigmata*, with families like *Sarcoptidae*, *Psoroptidae*, *Psorergatidae*, *Demodicidae*, and *Cheyletiellidae* (Benti et al., 2020). Dogs can be infested with *Cheyletiella yasguri*, *Demodex canis*, *D. injai*, *Sarcoptes scabiei*, and *Otodectes cynotis*. Cats may host *Cheyletiella blakei*, *Demodex cati*, *D. gatoi*, *Notoedres cati*, and *O. cynotis* (Little and Cornitas, 2021). Mite infestations can result in dermatitis, pruritis, skin allergies, and severe illness by invading internal organs (Olivry and Mueller, 2019).

Current Methods in Ectoparasites Control Chemical Methods

The drugs and chemicals which are used against ectoparasites are known as ectoparasiticides (Sharma et al., 2021). Various types of chemicals are used for this purpose including organophosphates, carbamates, pyrethroids, insect growth regulators, macrocyclic lactones, phenylpyrazoles, chloronicotinyl nitroguanidines, spinosad and the isoxazolines (Staffored and Coles, 2017). They are utilized in a variety of ways, including macrocyclic lactones (Ivermectin, Doramectin-subcutaneous injection), organophosphates (Malathion, Dichlorvos as dip, spray, pour on), carbamates (Carbaryl, Propoxur as dip, dust, spray), formamidines (Amitraz as dip, spray), and pyrethroids (Cypermethrin, Permethrin, Deltamethrin, etc. as dip, spray, spot on, and pour on). Synthetic hormones and arthropod enzymes, such methoprene and pyriproxyfen, can slow the development and proliferation of parasites. These are used in oral preparations as well as in combination with other drugs (Rust, 2020; Jamil et al., 2022). Isoxazolines, a recently developed class of insecticides, have the ability to strongly inhibit GABA-gated channels and have an impact on L-glutamate-gated chloride channels. In addition to killing adult cat fleas, they can also destroy triatomine bugs, ticks, mites, lice, mosquitoes, biting flies, and sea lice (Jamil et al., 2022). Fipronil containing collars are also available for dogs and cats which protect them from ticks, mites and lice (Paily et al., 2021).

Environmental Control

Environmental control includes the strategies like biological control, sanitation, and environmental modifications (Rust, 2020). Biological controls are used in the form of essential oils, entomopathogenic fungus, and Spider venom peptides. Azadirachtin a naturally occurring compound is used as an insecticide against agricultural pests which is very effective in biological controls (Jamil et al., 2022). Sanitation of carpets and surfaces can be done by treating with 0.4% dimeticone spray, IGRs and pyrethroids to control immature stages of insects. Vacuuming can also remove eggs and larva from the surface and carpets (Elsheikha, 2017; Rust, 2020). Environmental modification involves provision of adequate nutrition, removal of manures, hygienic food provision and keeping the animal stress free (Sharma et al., 2021).

Mechanical Methods

Many animals use mechanical defenses such as grooming to avoid or to mitigate the occurrences of infection by ectoparasites (Horn and Luong, 2019). Various types of grooming practices are done by domestic animals such as stimulus-response grooming (immediate immune response) and programmed grooming (response in form of bouts), or scratch and scan grooming (Kupfer and Fessler, 2018). Self-grooming may be energetically costly, comprises future resistance and other energy costs (Horn and Luong, 2019). Animals self-groom themselves by scratching, picking with their digits, or by using the mouth to remove ectoparasites (Kupfer and Fessler, 2018). Another mechanical method is bathing

which can be done with water, shampoo and chemicals (Frontline[®] Plus, Boehringer Ingelheim). Bathing can be performed every seven days and involves wetting the animal and apply the shampoo along the back, neck, belly and limbs (avoiding the eyes, nostrils and mouth). After waiting for five minutes clean it with warm water and dry the animal with the help of towel (Cruz et al., 2020).

Challenges and Limitations Associated with Existing Methods

Although there are different ways of fighting against parasitism, but these methods exhibit various limitations as well. These imitations include anthelmintic resistance and its prolonged use leads to worm refugia (Maqbool et al., 2017). Anthelmintic resistance is emerging as an important issue which develops due to irrational use of anthelmintic drugs, lack of knowledge about drug usage and lack of molecular diagnostic tests to detect anthelmintics resistance (Kotze et al., 2020). Use of biological control method has some limitations as it is not effective enough to remove infection. In addition, it is very slow acting, time consuming, gives unpredictable results, effective at one place and not effective at another similar place (Maqbool et al., 2017).

Nanoparticles

The use of technology at the nanoscale for the useful application in everyday life is known as nanotechnology. This includes manipulating physical, chemical, or biological systems down to the atom's submicron size and incorporating the resulting nanostructures into larger systems (Molento and Arenal, 2020). Nanoparticles (NPs) are particles characterized by dimensions typically falling within the range of 1 to 100 nanometers. Their properties vary based on size and surface characteristics. Due to their small size and expansive surface area, nanoparticles have wide range of applications in fields including cosmetics, electronics, as well as diagnostic and therapeutic medicine (Najahi-Missaoui et al., 2020). The practical application of nanotechnology has greatly accelerated the development of several sectors, most notably biomedical applications such as tissue engineering, drug transport, bio-imaging, and nano-diagnostics (Hikal et al., 2021).

There are major two categories of nanomaterials being used against the parasites i.e. Metallic and Polymeric Nanoparticles. Metallic NPs include silver, gold and copper nanoparticles. They exhibit antimicrobial and insecticidal properties due to their surface chemistry and small size. On the other hand, polymeric NPs include polymeric micelles, dendrimers, and nanocapsules. They are biocompatible, biodegradable and act as versatile carriers for controlled release of active ingredients (Begines et al., 2020; Hikal et al., 2021). The nanoparticles mainly work by entering the body of parasite resulting into cell membrane damage, ribosome disassembly, protein denaturation and oxidative stress. The oxidative stress and denaturation of the protein results in mitochondrial and DNA damage. This leads to the death of the parasite (AlGabbani, 2023). However, nanoparticles also offer various other ways to eliminate the parasites from the body of the host. These particles offers the methods to address the shortcomings of conventional drug delivery systems (Aljabali et al., 2018). In order to fight ectoparasites, nanoparticles use a variety of strategies, the most common of which are controlled release of active chemicals and physical disruption. Sharp-edged or spiked nanoparticles cause structural damage to the cellular membranes of ectoparasites when they come into direct contact with them. Moreover, nanoparticles are effective dispersants of insecticidal or acaricidal substances, allowing for their slow release over time (Banu et al., 2023).



Fig. 1: General Mechanism of Nanoparticles against ectoparasites

Application of Nanoparticles in Ectoparasite Control

The use of nanoparticles in diverse formulations for direct and environmental treatments is a viable route for controlling ectoparasites (Zaheer et al., 2022).

A. Topical Formulations

i) Nanoparticles in Spot-on Treatments: Spot-on treatments represent a common method for administering ectoparasiticides directly onto the skin of companion animals (Ferrer, 2021). Nanoparticles integrated into spot-on formulations enhance efficacy through several mechanisms. Firstly, nanoparticles can serve as carriers for active ingredients, protecting them from degradation and facilitating their controlled release over time. This sustained release ensures prolonged exposure of ectoparasites to the insecticidal or acaricidal compounds, thereby enhancing efficacy while reducing the frequency of application. Additionally, nanoparticles with specific surface modifications can improve adhesion to the skin, prolonging the duration of action and minimizing product runoff (Pavoni et al., 2019; Prasad et al., 2021).

ii) Nanoparticles in Shampoos and Sprays: Shampoos and sprays are alternative topical formulations commonly used for ectoparasite control in companion animals (Gorman, 2016). Integration of nanoparticles into these formulations offers several advantages (Pereira-Silva et al., 2022). Nanoparticles can enhance the penetration of active ingredients through the ectoparasite's cuticle, increasing their bioavailability and efficacy. Moreover, nanoparticles can confer stability to formulations, preventing degradation of active compounds and ensuring consistent performance over time (Molento and Arenal, 2020).

B. Environmental Applications

Nanoparticles are embedded in fabrics, beddings and household sprays. Ectoparasites often reside in the environment surrounding companion animals, including their bedding and living areas (Kocoń and Nowak-Chmura, 2017). Nanoparticles can be incorporated into fabrics, household sprays and bedding materials to create a hostile environment for ectoparasites (Chatha et al., 2019). Metallic nanoparticles, such as silver nanoparticles, exhibit potent antimicrobial properties and can be impregnated into textiles to inhibit the growth of ectoparasites and prevent transmission between animals (Qu et al., 2023). Additionally, sprays can be applied to surfaces frequented by companion animals, such as floors, carpets, and furniture, to eliminate ectoparasites and their eggs (Elsheikha et al., 2018).

C. Controlled-Release Formulations

Nanoparticles offer the protection by controlled release formulations. These products maintain therapeutic concentrations for an extended duration by encapsulating insecticidal or acaricidal chemicals within nanoparticles, therefore ensuring a continuous supply of the active ingredient. This long-lasting impact lessens the need for frequent treatment, improving animal owners' convenience and guaranteeing efficient ectoparasite management (Danyaro et al., 2023; Najitha Banu et al., 2023).

Efficacy and Safety of Nanoparticle based Ectoparasite Control

Nano-particles are very efficient in the control of ectoparasites as repeated use of acaricides has led to environmental pollution, resistance development and increase in the treatment cost (Abdel-Ghany et al., 2022). Different types of nano-particles have developed for this purpose such as silver NPs, titanium dioxide NPs, zinc oxide NPs, nickel NPs, copper NPs and magnesium NPs (Benelli et al., 2017). Zinc Oxide nanoparticles and cypermethrin-coated nanoparticles of ZnO (C-ZnO NPs) and ZnS (C-ZnS NPs) are very effective against many *Rhipicephalus* ticks as well as *Hyalommma* ticks (Zaheer et al., 2023). Zein nano-particles associated with cypermethrin (CYPE) + chlorpyrifos (CHLO) + a plant compound (citral, menthol or limonene) have high efficacy against *Rhipicephalus microplus* ticks as compared to other acaricides. This formulation lowers the nematodes toxicity and has long period of residual activity (Figueiredo et al., 2023). Similarly, iron oxide and iron sulphides NPs coated on pyrethroids (cypermethrin and deltamethrin) can control the major life stages of *Hyalomma* ticks (Zaheer et al., 2024). Hence, nano-particles have low efficacy and short shelf-life but nanopaticles-based drug delivery system effectively targets the acaricidal drugs into the sites of infection as well as enhances the efficacy of the acaricides (Najitha Banu et al., 2023).

The nanoparticles have no safety concerns (safe in use) and also reduce the adverse effects of various drug but some nanoparticles have toxic profile (Bajwa et al., 2022). Treatment with silver nanoparticles has caused DNA damage and morphological abnormalities in a variety of vertebrate and invertebrate creatures. It has also negatively impacted the enzymatic activities of a number of non-target species (Benelli, 2018). The toxicological evaluation of these nanoparticles is an important factor to reduce its negative effect on non-target species (Figueiredo et al., 2023). However, these NPs should have excellent chemical and physical structures, should be non-toxic and should be evaluated at every cellular level during their preparation. This evaluation will help to reduce their toxic effects (Bajwa et al., 2022).

Compared with traditional method (use of chemical, environmental managements, biological and mechanical means), nano-particles have more environmental safety and low pesticide resistance (Nie et al., 2023). Nano-particles coated on acaricides have led to development of drug delivery system which could increase the efficacy and performance of previous acaricidal drugs by targeting the site of infections. These NPs are also water soluble and have no effect on the environment as compared to various chemicals used in traditional methods (Athanassiou et al., 2018). NPs based acaricidal formulations

will enhance stability, action and duration of insecticidal activity. It will also remove harmful organic solvents which are not removed in commonly used acaricides (Zaheer et al., 2022).

Conclusion

In conclusion, the field of nanotechnology presents a promising avenue for revolutionizing the control of ectoparasites in companion animals. Ectoparasites pose significant health risks to both animals and humans, and current control methods are often limited by variable efficacy, toxicity concerns, and the development of resistance. The application of nanoparticles in various formulations, including topical treatments, environmental applications, and controlled-release formulations, offers diverse and effective strategies for combating ectoparasites. Nanoparticles integrated into spot-on treatments, shampoos, and sprays enhance efficacy by improving adhesion to the skin and penetration through the ectoparasite's cuticle. Furthermore, nanoparticles embedded in fabrics and household sprays create a hostile environment for ectoparasites in the animal's surroundings. Despite the numerous advantages of nanoparticle-based ectoparasite control offers a promising solution to the ongoing challenges faced in companion animal healthcare. By leveraging the innovative potential of nanotechnology, we can develop safer, more effective, and sustainable strategies for managing ectoparasitic infestations, ultimately enhancing the health and well-being of companion animals worldwide.

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