ISSN: 2224-0616
Int. J. Agril. Res. Innov. Tech. 15(1): 22-38, June 2025 Available online at https://ijarit.online
DOI: https://doi.org/10.3329/ijarit.v15i1.82751 https://www.banglajol.info/index.php/IJARIT

Exploring the role of chitosan in enhancing poultry production: Benefits and applications

Ghulam Abbas[1]*, Abdullah Hassan Hashmi[2], Muhammad Saeed Imran[3], Muhammad Arshad[1], Ahsan Mustafa[1], Muhammad Asif Naveed[1], Umar Farooq[4], Seemab Jaffery[5], Muhammad Talha[6], Kinza Imran[7], Muhammad Naveed Alam[8], Kunwal Hafeez[1], Sundas Arif[1], Muhammad Shahzaib[1], Sundas Amin[1], Asyia Shafique[1], Ghulam Muhammad Gill[1], Asma Basharat[1], Ghulam Rasool Zahid[9], Muhammad Jamshaid[1] and Syed Muhammad Ali Hassan[1]

Received 6 February 2025, Revised 25 May 2025, Accepted 20 June 2025, Published 30 June 2025

ABSTRACT

Chitosan (a chitin derived biopolymer) has gained enormous attention due to its potential uses in broiler and layer production. Chitosan is a natural, nontoxic, and biodegradable substance that has several benefits in poultry, i.e., it improves poultry growth, health, and overall productivity. The present review paper explores the versatile roles of chitosan in poultry, exploring its use as an immune-modulator, growth promoter, and natural antimicrobial and antioxidant feed additive. Chitosan biopolymer is reported to boost the immunity FCR (feed conversion ratio) and reduce the chances of diseases working as an antimicrobial compound. Chitosan enhances nutrient digestibility and absorption, maintains gut health, and controls the intestinal microflora. Dietary use of chitosan is well documented to improve meat quality, feather growth, and egg quality as an alternative to synthetic growth promoters and antibiotics. Therefore, this review intends to gather scientific evidence about using chitosan on production performance, health and well-being of poultry and minimizing the impact of production on the environment for sustainable and environmentally healthy production. Moreover, the prospects for research on chitosan in poultry production are also discussed, stressing the need to optimize chitosan applications and address existing limitations.

Keywords: Chitosan, Antimicrobial, Feed additive, Production performance, Poultry welfare, Nutrient digestibility, Sustainability

[1]*Riphah College of Veterinary Sciences, Riphah International University, Lahore, Pakistan*
[2]*University Institute of Food Science and Technology, University of Lahore, Pakistan*
[3]*Department of Pathology, University of Veterinary and Animal Sciences, Lahore, Pakistan*
[4]*University of Agriculture Faisalabad, Sub campus Toba Tek Singh, Pakistan*
[5]*Center of Agricultural Biochemistry and Biotechnology, Faculty of Agriculture, University of Agriculture Faisalabad, Pakistan*
[6]*Department of Clinical Medicine and Surgery, University of Veterinary and Animal Sciences, Lahore, Pakistan*
[7]*Department of Food Science and Human Nutrition, University of Veterinary and Animal Sciences, Lahore, Pakistan*
[8]*Faculty of Social Sciences, University of Agriculture Faisalabad, Pakistan*
[9]*Police Service of Pakistan, Joint Director General Intelligence Bureau, Islamabad, Pakistan*

*Corresponding author's email: ghulamabbas_hashmi@yahoo.com (Ghulam Abbas)

Cite this article as: Abbas, G., Hashmi, A.H., Imran, M.S., Arshad, M., Mustafa, A., Naveed, M.A, Farooq, U., Jaffery, S., Talha, M., Imran, K., Alam, M.N., Hafeez, K., Arif, S., Shahzaib, M., Amin, S., Shafique, A., Gill, G.M., Basharat, A., Zahid, G.R., Jamshaid, M. and and Hassan, S.M.A., 2025. Exploring the role of chitosan in enhancing poultry production: Benefits and applications. *Int. J. Agril. Res. Innov. Tech.* 15(1): 22-38. https://doi.org/10.3329/ijarit.v15i1.82751

Introduction

Chitosan is a natural polysaccharide bio-polymer derived from Chitin (the major component of arthropods) derived from the exoskeleton of arthropods and crustaceans e.g., shrimps, lobsters, crabs, etc., which has currently gained significant attraction in poultry production because of its beneficial effects viz; antimicrobial, growth promoter, antioxidant, and immune-modulating effects (Khambualai *et al.*, 2009).

In poultry production, chitosan has gained significant attention as a potential alternative to antibiotics, which are being eliminated more and more due to concerns about antibiotic resistance. The chitosan contributes to faster growth and better food efficiency by promoting better nutrient digestion and absorption. Several studies have shown that chitosan improves the food conversion relationship (FCR), a critical metric in the cultivation of poultry, by increasing the efficiency with which birds turn food into body weight. A study by Yu *et al.* (2016) showed that the chitosan improves the height of the villus and the depth of the crypt in the intestines of the poultry. This increase in surface increases nutrient absorption, which leads to better growth. The study found that the birds that received chitosan in their feed significantly increased their average daily weight compared to the control group. Research by Shamsudin *et al.* (2015) reported that chitosan supplementation improved FCR, indicating that birds could utilize feed more efficiently. This resulted in reduced feed costs, as less feed was required to achieve the same weight gain, a crucial factor for improving the economic sustainability of poultry farming (Huang *et al.*, 2005). Chitosan has been found to increase the production of key immune cells such as macrophages, T-cells, and infection-fighting cells, which are responsible for protecting the body from pathogens and infections. Besides that, the controlling/adjusting of the release of cytokines by chitosan aids in the creation of a balanced unable-to-be-harmed response. Research has shown that dietary chitosan improves nutrient digestibility, eventually leading to a better feed.

Several researches found Chitosan significantly improves the immune response in poultry; therefore, it is a significant component in poultry production. Besides that, the modulation of the release of cytokines by chitosan aids in creating a balanced immune response in the poultry. The ability of chitosan to strengthen immunity leads to a lower infection rate and, hence, enhanced general flock health with minimal use of antibiotics or other synthetic growth promoters. By promoting natural immunity, chitosan allows for a more sustainable and bio-secure poultry production system. Research has shown that dietary chitosan enhances poultry's nutrient digestibility, eventually improving feed efficiency. Its capacity to enhance the absorption of key nutrients such as proteins, fats, and carbohydrates allows for nutrients in feed to be utilized more efficiently. This results in increased weight gain and growth in poultry. Moreover, its impact on digestive enzymes and gut morphology significantly improves complex nutrient digestibility in birds to maximize the nutritional value obtained from feed.

Chitosan has been shown to exhibit good effects on the gut health of poultry, including fostering beneficial gut microbiota and improving the integrity of intestines. It has also been proven to improve gut anatomy by raising the surface area for the absorption of nutrients through stimulation of villi growth in the intestines. Improving gut structure leads to better nutrient absorption and digestion. In addition, chitosan helps balance beneficial gut bacteria, keeping a healthy microbiome in place, which supports digestion, immunity, and general health. By improving gut health, chitosan boosts poultry health and productivity and supports a more sustainable and eco-friendly approach to poultry farming, reducing reliance on antibiotics and synthetic growth promoters. Therefore, the review article explores the possible benefits and applications of chitosan for enhancing poultry health, immunity, welfare growth, gut health, and overall production efficiency.

Chitosan: a versatile biopolymer with a wide range of applications

Chitosan is a natural biopolymer derived from chitin, a major structural component of the exoskeletons of crustaceans such as crabs, shrimps, and lobsters. It is obtained by deacetylation of chitin, which removes acetyl groups from the polymer chain. Chitosan has gained significant attention in recent years due to its unique properties and wide range of applications in various fields, including medicine, agriculture, food, and water treatment.

Properties of Chitosan

Chitosan is a cationic polymer with a positive charge at low pH. This property gives it several unique characteristics:

- Chitosan is generally well-tolerated by the human body and is non-toxic. It is biodegradable and can be easily metabolized by the body.
- Chitosan is biodegradable in the environment, making it an environmentally friendly material.

- Chitosan exhibits antimicrobial activity against various microorganisms, including bacteria, fungi, and viruses. This is due to its ability to interact with the cell membranes of microorganisms, disrupting their integrity.
- Chitosan promotes wound healing by stimulating cell proliferation and migration. It also forms a protective barrier over the wound, preventing infection and promoting tissue regeneration.
- Chitosan has a high affinity for various substances, including heavy metals, dyes, and other pollutants. This property makes it useful for water treatment and waste management.

Widespread applications of chitosan in various fields

Medicine: Chitosan is biocompatible, meaning it is well-tolerated by the body and does not cause adverse reactions. Chitosan can accelerate blood clotting due to its ability to interact with platelets and promote fibrin formation. This makes it a promising material for wound dressings, particularly in cases of severe bleeding. This is why chitosan-based dressings and films accelerate wound healing and prevent infection. Chitosan can be used to encapsulate drugs, vaccine and nutrients and deliver those nutrients to particular target points in the body (Fig. 1). It is used to encapsulate drugs and deliver them in a controlled manner to particular points in the body to the desired location. This can improve drug efficacy and reduce side effects. It is also used as a scaffold material for tissue engineering applications, such as cartilage regeneration and bone repair, to enhance the immune response and stimulate the production of antibodies. Moreover, its antimicrobial properties help prevent wound infection, a crucial factor in promoting healing.

Fig. 1. Chitosan is a bioactive agent to delivers drugs, vaccines, and nutrients at target sites (Huang *et al.*,2005)

Agriculture: Chitosan can stimulate plant growth and development by promoting root elongation and enhancing nutrient uptake. Chitosan can be used as a bio-control (natural fungicide and bactericide) to protect plants from diseases. Chitosan can improve soil quality by increasing water retention and nutrient availability; hence, it stimulates plant growth and development by promoting root elongation and enhancing nutrient uptake (Fig. 2).

Food Industry: Chitosan can be used as a natural food preservative to extend the shelf life of food products. Chitosan can be used to clarify fruit juices and wines by removing suspended particles. Chitosan can be used as a thickening and gelling agent in various food products (Fig. 2).

Water Treatment: Chitosan can be used to remove heavy metals (lead, cadmium, and mercury) from contaminated wastewater to purify it as it has a high affinity for these metals and to remove dyes from textile wastewater, hence helping to reduce environmental pollution. Chitosan can remove bacteria and other microorganisms from drinking water (Fig. 2).

Other Applications: Chitosan is used in cosmetics as a hair conditioner (forming a protective film around hair strands), skin moisturizer, and sunscreen (improve skin hydration and reduce wrinkles, protect the skin from UV radiation) and to produce biodegradable plastics. Chitosan can be used in biofilters for air purification (Fig. 2).

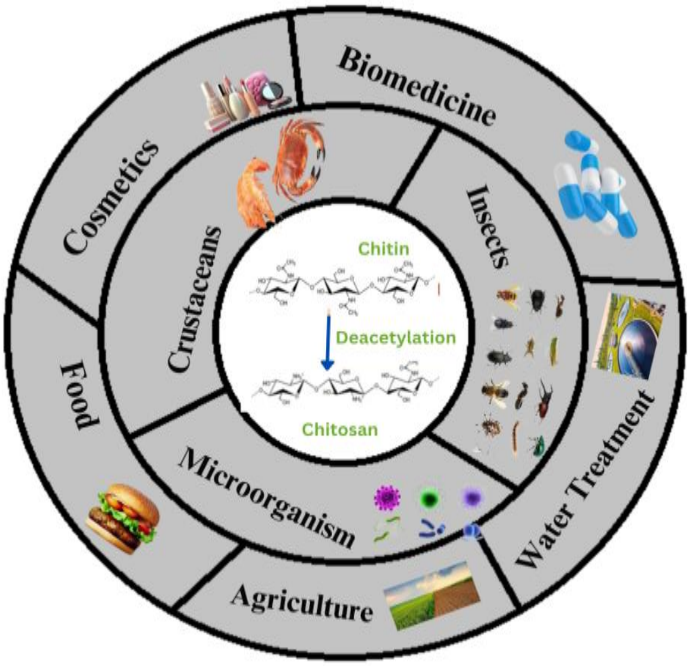

Fig. 2. Widespread application of Chitosan in different industries; adopted by Wu *et al.* (2024).

Chitosan also improves gut health by acting as a prebiotic, encouraging the growth of beneficial gut macrobiotics while suppressing harmful pathogens. This helps reduce digestive disturbances and maintain a healthy gut environment, essential for optimal nutrient absorption and overall health. Chitosan helps to reduce intestinal permeability, or "leaky gut," which can lead to nutrient loss and increased susceptibility to infections. By improving intestinal barrier function, chitosan helps poultry retain more nutrients from their feed, leading to better growth and immune defense. By controlling harmful bacteria, chitosan reduces inflammation and stress in the digestive tract, which can improve overall gut (Yu *et al.*, 2016).

With antimicrobial and anti-fungal properties, chitosan helps control harmful microorganisms and protects poultry birds from infections that could otherwise interrupt growth and production performance (Jang and Lee, 2014). Chitosan binds to the cell walls of bacteria, breaking off their structure and preventing their growth. Research has shown that chitosan is effective against several pathogens of poultry like *Escherichia coli, Salmonella, Campylobacter,* and *Clostridium Perfringens* (Xia *et al.*, 2011), hence helps to improve intestinal health, nutrient digestibility/absorption, and production (Hossain and Lim, 2014). This beneficial product also has anti-fungal properties and can help to control mold and fungal contamination in poultry feed by disrupting the cell membranes of mushrooms, preventing their growth and reducing the risk of mycotoxicosis. Studies have shown that chitosan can reduce fungus contamination by up to 50%.

The chitosan effects on the immune response of poultry is attributed to its ability to modulate immune responses, increase resistance to diseases, and stimulate function of immune cells including macrophages and lymphocytes, which are the body's main defense, hence reducing the need for antibiotics. Chitosan is also reported to increase antibody production, improving further poultry disease resistance (Zhang *et al.,* 2012). In a study, birds fed with Chitosan showed significantly improved serum levels of immunoglobulin G (IgG). Therefore, Chitosan offers a natural alternate option to address the increasing concern for traditional antibiotic resistance. This is especially important in organic poultry farming and regions with strict regulations regarding antibiotic use (Wu *et al,* 2005).

How it works in poultry

Catioonc chitosan's allows it to interact with anionic bacterial cell membranes. This interaction disrupts the membrane's integrity, leading to cell death. This antimicrobial property makes chitosan effective against various pathogens that can infect poultry, such as *E. coli, Salmonella,* and *Clostridium perfringens*. Beyond its antimicrobial effects, chitosan also influences the gut microbiome. It can selectively stimulate the growth of beneficial bacteria while suppressing the growth of harmful pathogens. This balanced gut macrobiotic is crucial for nutrient absorption, immune function, and overall bird health.

Benefits of using chitosan in poultry farming

Studies have shown that supplementing poultry feed with chitosan can enhance feed conversion efficiency, leading to faster growth rates and increased body weight. Chitosan can stimulate the immune system of poultry by activating macrophages and increasing the production of antibodies. This can help birds better resist infections and diseases. By controlling pathogens and improving gut health, chitosan can help reduce mortality rates in poultry flocks. Chitosan may also improve meat quality by enhancing muscle growth and reducing fat deposition. As a natural alternative to antibiotics, chitosan can help reduce the reliance on these medications in poultry farming.

Chitosan can be incorporated into poultry feed, including powder, pellets, and premixes. The optimal dosage and administration method will depend on factors such as the age and health status of the birds, the specific pathogens of concern, and the desired outcomes. Whilst chitosan offers several benefits, it's important to note that research on its use in poultry is ongoing. More studies are needed to fully understand its mechanisms of action and optimize its application in different poultry production systems. Chitosan can also extend the shelf life of poultry products by preventing microbial growth during storage. This has economic implications for the poultry industry, as it helps maintain the quality and safety of poultry meat during transportation and retail distribution (Chen *et al.,* 2013). Chitosan can be applied to poultry meat as a natural preservative. Research has shown that when chitosan is used as a coating for poultry products, it inhibits the growth of spoilage microorganisms and extends shelf life by up to 50% by direct impact reducing the forging and improving the safety of meat, especially when the climate is hot for faster bacterial growth (Ouattara *et al.,* 2000). Being an antioxidant Chitosan helps protect birds from oxidative stress caused by free radicals, which can damage cell structure and deteriorate immune function, Chitosan, however helpful to neutralize these free radicals, and its use in poultry production is in line to address the increasing demand of sustainable and organic agricultural practices (Badawy and Rabea, 2011) by cutting antibiotics requirement, improving natural feed quality and extending the shelf life of poultry products (Kim *et al.,* 2011). Therefore, Chitosan is a promising solution for economic and environmental goals in modern poultry production (Zhang *et al.,* 2012; Kong *et al.,* 2010).

Table 1. Antimicrobial and antioxidant applications of chitosan.

Application Field	Mode of Action	References
Antimicrobial	The positively charged chitosan molecules interact with negatively charged microbial cell membranes, causing the membrane to rupture. This gives chitosan its antibacterial properties.	Wu *et al.* (2024)
Antioxidant	Chitosan demonstrates antioxidant properties by neutralizing hydroxyl, superoxide, and DPPH radicals.	Yang and Chen, 2014)

In a study (Huang *et al.,* 2005) Chitosan supplementation in poultry diets led to a momentous improvement in the weight gain by increasing the surface area of the intestine to facilitates better absorption of nutrient as it causes improved villus height and crypt depth of the intestines leading to an increased growth and production performance hence helpful to reduce the feed costs and increase the profitability of poultry production (Zhuang *et al.,* 2015).

Chitosan acts as a pre-biotic and promotes the growth of beneficial bacteria (lactobacillus and bifid bacteria, etc.), inhibiting the growth of harmful pathogens (Hassan *et al.,* 2016), causing intestinal micro-flora balance that is necessary for proper digestion and absorption of nutrients. The Zhang study investigated that Chitosan can lessen intestinal permeability and prevent leakage of harmful microorganisms and toxins into the bloodstream. As an anti-inflammatory substance, Chitosan helps to reduce bowel inflammation caused by enteritis, bacterial infections, and/or digestive disorders (Jang and Lee, 2014).

Key benefits of chitosan in poultry farming

Chitosan improves the digestibility of numerous nutrients in feed which improves bioavailability of feed in cost effective mode. This is achieved by modulating the gut macrobiotic, promoting the growth of beneficial bacteria, and improving nutrient absorption. Chitosan has been shown to stimulate the immune system of poultry, making them more resistant to various diseases. It activates immune cells, increases antibody production, and enhances the immune response. Chitosan's antimicrobial properties can help reduce the reliance on antibiotics in poultry farming (Khalil *et al.,* 2020). By controlling pathogens and improving gut health, chitosan can help maintain a healthy gut environment and reduce the risk of infections. Chitosan can improve meat quality by enhancing muscle growth, reducing fat deposition, and improving meat tenderness. This is achieved by optimizing nutrient utilization and regulating metabolic processes.

Mechanism of action

Chitosan's positive charge interacts with the negatively charged cell membranes of bacteria, fungi, and viruses, disrupting their integrity and leading to cell death. Chitosan can activate macrophages, a type of white blood cell that plays a vital role in the stress response. It can also stimulate the production of cytokines, which are signaling molecules that control stress function. Chitosan can control/adjust the gut macrobiotic, the growth of helpful bacteria and stop the growth of harmful. This helps maintain a healthy gut (surrounding conditions), which is extremely important for something that acts as food and is unable to be harmed function.

Applications in poultry farming

Chitosan can be incorporated into poultry feed, including powder, pellets, and premixes. It can also be used as a coating for feed or as a water additive. The optimal dosage and administration method will depend on the specific needs and goals of the poultry farm. Beyond its prebiotic effects, chitosan is a potent antimicrobial agent. It has been shown to reduce harmful bacteria such as Salmonella, Escherichia coli, and Clostridium perfringens, all common pathogens that can cause illness and affect poultry performance. Chitosan works by binding to the bacterial cell walls and disrupting their integrity, which prevents the bacteria from proliferating. Studies by Liu demonstrated that chitosan supplementation significantly reduced bacterial loads in the intestines and helped maintain a healthier gut macrobiotic, leading to fewer digestive problems and better overall health. This reduction in harmful bacteria means less energy is spent by the birds fighting infections, which in turn allows more energy to be directed towards growth (Liu *et al.,* 2010).

Chitosan's anti-mycotoxin properties are also valuable in farming, especially in preventing the contamination of feed with molds and fungi that produce mycotoxins (Dai *et al.,* 2015). Molds such as *Aspergillus* and *Fusarium* are commonly found in feed, and the mycotoxins they produce can lead to serious health issues, including liver damage, reduced feed intake, and damaged/weakened unable to be harmed function. Research has shown that chitosan stops fungal growth and spores, which helps reduce the risk of mycotoxicosis (Al-Marzooqi *et al.,* 2012). A study by Tavares showed that chitosan could reduce fungal contamination in feed by up to 50%, by that/in that way lowering the possible exposure to harmful mycotoxins and improving feed quality (Liu *et al.,* 2012).

In addition to its antimicrobial benefits, chitosan has been found to control/adjust the disease-fighting system, increasing their resistance to infections. Chitosan boosts the activity of macrophages and infection-fighting cells, and improves resistance against diseases. Research by Yang *et al.* (2016) and

Yang *et al.*, (2009) showed that birds added with chitosan had higher levels of immunoglobulin G (IgG), a key marker of unable to be harmed response, compared to those not increased/added.

This enhanced immune response improves disease resistance and reduces the need for antibiotics, which is particularly important in light of the growing concerns over antibiotic resistance in agriculture. By improving the poultry's natural defenses, chitosan can reduce the frequency and severity of infections, helping to maintain healthy flocks and reduce veterinary costs (Zhou *et al.*, 2009).

Table 1. Applications and Benefits of Chitosan Nanoparticles in Poultry Nutrition and Immunity.

Application	Mechanism	Benefits	References
Nutrient Delivery	Encapsulation of essential nutrients for targeted release.	Enhanced nutrient bioavailability and absorption; reduced nutrient wastage.	Burt (2004)
Immunomodulation	Activation of macrophages and stimulation of cytokine production.	Improved immune responses; enhanced resistance to infections.	Lan *et al.* (2019)
Antimicrobial Activity	Disruption of bacterial cell membranes due to chitosan's cationic nature.	Reduction of harmful gut pathogens; promotes a healthier gut microbiome.	Sharma *et al.* (2020)
Antioxidant Properties	Neutralization of free radicals and reduction of oxidative stress.	Improved overall health and performance; better meat and egg quality.	Zhang *et al.* (2018)
Improved Feed Efficiency	Enhancement of gut villi structure, improving nutrient absorption.	Reduced feed costs; higher weight gain with less feed.	Lan *et al.* (2021)

Production of free radicals during oxidative stress can damage cells and immunity, however, Chitosan are reported to neutralize these free radicals thus protect cells from being damaged (Zhang *et al.*, 2018) contribute to better health, disease resistance, and improved production performance, during heat stress and oxidative stress (Lan *et al.*, 2019). It also preserves poultry meat preventing microbial spoilage during storage conditions and extends the shelf life of poultry products. In a study, poultry products coated with chitosan showed significantly longer shelf life and healthier microbial quality when compared to non-treated products (Sadeghi and Motlagh, 2013). As the industry continues to search sustainable alternatives to chemical additives and traditional antibiotics, chitosan offers an auspicious solution (Al-Murrani and Al-Mashhadani, 2015; Dong *et al.*, 2016).

Chitosan help poultry's resistance to pathogens, which is especially beneficial in large-scale poultry operations where diseases can spread quickly. It stimulating both the innate and adaptive immune responses and increase key immunity markers e.g. IgA, IgG, IgM and cytokines, which are necessary to defend against pathogens (Shahidi *et al.*, 2010). In addition, it also boosts macrophage activity playing a critical role in phagocytosis as Researchers have shown that chitosan can boost the function of macrophages (Hassan and El-Hamid, 2012). Moreover, chitosan helps to reduce inflammation by balancing pro-inflammatory and anti-inflammatory responses (Xie *et al.*, 2017).

Chitosan boost the activity of antioxidant enzymes i.e. superoxide dismutase (SOD) and catalase (CAT) and protect cells and tissues from oxidative damage and improve the meat quality by maintaining muscle tissue integrity, color, texture, and shelf life (Yan and Zhang, 2019; Yang *et al.*, 2016). This can assist lessen post-slaughter losses and improve the quality of stored meat. It also reported to improve the strength of eggshells by improving calcium retention making the eggs stronger and sound and protects egg yolk from oxidation (improve the lipid profile of meat and eggs, potentially lowering cholesterol levels), maintaining its color and nutritional worth thus help orienting with consumer demand for healthier food choice (Wang *et al.*, 2017; Zhao *et al.*, 2014). By doing so, chitosan not only helps to produce healthier poultry flocks but also supports antibiotic-free poultry production, which is becoming increasingly important in both traditional and eco-friendly organic poultry farming (Zhang *et al.*, 2018).

Table 2. Immune response, anti-inflammatory, and anti-cholesteremic applications of chitosan.

Application Field	Mode of Action	References
Immune Response	Complement system activation, improvement in humoral immunity and stimulation of CD4+ cells are main immune responses produced by chitosan.	Jang and Lee (2014)
Anti-inflammatory	Chitosan has powerful anti-inflammatory properties by modulating the inflammatory response and reducing its severity in chronic illnesses.	Wang *et al.* (2005)
Anti-cholesteremic	Chitosan helps lower LDL cholesterol and triglyceride levels when used in the form of chitosan oligosaccharides.	Wang *et al.* (2017)

The benefits of chitosan in poultry farming are also important for environmental healthy poultry farming. Chitosan also improves FCR, reduces resource input use and minimizes the environmental impact of feed production (Khateeb and Al-Ajmi, 2011). In addition to improving feed efficiency, chitosan can enhance waste management in poultry farms. By reducing the levels of harmful nutrients such as nitrogen and phosphorus in poultry manure, chitosan can help mitigate pollution and improve the sustainability of waste disposal practices. Moreover, chitosan is biodegradable and has a minimal environmental footprint compared to synthetic additives and chemicals commonly used in poultry farming, making it a more eco-friendly option (Liu *et al.*, 2020). Its ability to enhance poultry health while reducing antibiotic use aligns with the growing trend toward more sustainable and natural farming practices. Additionally, its environmental benefits—such as improved feed efficiency, reduced antibiotic residues and better waste management—make chitosan an important component of sustainable poultry farming. As the industry seeks alternatives to synthetic chemicals and antibiotics, chitosan provides a safe, effective, and environmentally friendly solution for improving poultry health, productivity, and sustainability (Zhang and Luo, 2017). Chitosan has been shown to have a broad range of positive effects on environmental health and poultry performance by its impact on nutrient absorption promoting healthy gut environment cut-down the antibiotics usage thus reduces the environmental impact of poultry farming (Wang *et al.*, 2020; Yang *et al.*, 2009).

In addition to its antimicrobial activity, chitosan has been found to reduce intestinal inflammation and increase intestinal integrity. By enhancing gut barrier function, chitosan helps prevent conditions like leaky gut, where harmful substances can pass through the intestinal wall and cause systemic inflammation. Studies have shown that chitosan can increase villus height in the intestine, which is indicative of a healthier, more efficient digestive system (Zivanovic *et al.*, 2005).

Chitosan's ability to modulate immune cells such as macrophages and lymphocyte helps poultry better defend against a range of viral, bacterial, and fungal infections helps to reduce the need for vaccines and antibiotics shifting toward sustainable organic poultry farming practices addressing common issues of small stack poultry farms by supporting the innate immune response (Liu *et al.*, 2019).

Furthermore, chitosan is a promising alternative to synthetic chemical additives in poultry feed. As concerns over chemical residues in food and the environment continue to grow, chitosan offers a natural and biodegradable solution. Its use can reduce the accumulation of harmful substances in the environment, particularly in poultry manure. By lowering the levels of nitrogen and phosphorus in poultry waste, chitosan helps mitigate the environmental impact of poultry farming, particularly in terms of nutrient runoff and water pollution. This makes chitosan a valuable tool in sustainable farming practices, where the goal is to reduce waste and improve the overall environmental footprint of animal production (Rioferio *et al.*, 2021).

Several studies have also proved that chitosan can help to progress the lipid profile of both poultry meat and eggs, which is in line with consumer demand for healthier food options. By reducing cholesterol levels in the meat and eggs, chitosan can help produce healthier poultry products. This is especially valuable in meeting the growing market demand for low-cholesterol and heart-healthy foods. Additionally, chitosan's effect on reducing oxidative damage may play a role in improving the nutritional quality of poultry products by preserving essential fatty acids and other sensitive nutrients in the meat and eggs (Yin *et al.,* 2008).

Chitosan's versatility and effectiveness in poultry farming extend beyond health benefits to also include economic advantages. The reduced need for antibiotics and other chemical additives can lead to lower veterinary costs, and the improvement in feed efficiency directly translates to cost savings. Furthermore, the enhanced growth rates and healthier flocks can increase production yields and overall profitability for poultry farmers. As the demand for antibiotic-free and sustainably produced poultry products increases, chitosan offers a way to meet these consumer preferences while improving both the productivity and sustainability of poultry farming (Zhou *et al.,* 2012).

Chitosan in Animal Feed

Chitosan offers a range of advantages for animal health and productivity. Chitosan exhibits strong antimicrobial properties against a wide range of bacteria, fungi, and viruses. This is due to its positively charged structure, which interacts with the negatively charged cell membranes of microorganisms, disrupting their integrity and leading to pathogen death. Chitosan also reported to enhance nutrient digestibility, and absorption/utilization in poultry birds by enhancing the gut health and improving gut micro-flora causing better FCR and growth performance. Chitosan stimulates the immune system by activating macrophages, other immune cells, enhancing the antibodies production, and strengthening the animal's defense against infections/diseases. Possessing antioxidant properties, Chitosan helps to neutralize free radicals produced in the body and protect cells from the oxidative damages hence help to reduce the risk of oxidative stress-in poultry. This can help to reduce the dependence of poultry industry on antibiotics and other synthetic feed additives and addressing the growing problem of antibiotic resistance in humans and animals.

Effect of chitosan on production performance, body organs weight, blood profile, and immune response of poultry

The use of chitosan in poultry feed has been widely studied at varying concentrations, and the results regarding its effects on carcass and organ weight and slaughter characteristics are reported. Tufan and Arslan (2020) reported that supplementation at 50 or 100 mg/kg of chitosan has improved the carcass percentage and enhanced the leg and wing relative weights, but decreased the carcass-to-liver body weight ratio. However, in a following study carried out by Tufan *et al.* (2015), dietary addition of chitosan at 75 or 150 mg/kg had no significant effect on carcass weight, heart, liver, or gizzard percentages in Japanese quail. Sirsat Shraddha *et al.* (2017) investigated the dietary of chitosan and neem leaf meal at 0.025% and 0.05% to lower abdominal fat contents in broilers. Whereas, results of the experiment conducted by Arslan and Tufan (2018) who used chitosan oligosaccharides and L-carnitine both in a combination at 100 mg/kg reported no significant changes in carcass weight, carcass ratio, or heart, spleen, and gizzard ratios to carcass weight. Similarly, Miao *et al.* (2020), did not find the effects of 200 mg/kg of chitosan on dressing percentage, eviscerated carcass percentage, and half-eviscerated carcass percentage in Huoyan geese. Other sources of chitin (studies on cricket and shrimp) have also been explored for their impact on poultry. Lokman *et al.* (2019) reported that cricket chitin and chitosan supplementation at 0.5 g/kg or 0.5 g/kg of shrimp chitin and shrimp chitosan did not improve carcass or organ characteristics in broilers.

Several studies have explored the effects of chitosan and its derivatives on various biological processes in poultry. Li *et al.* (2009) found that supplementation with chitosan at concentrations of 0.05-0.10% increased nitric oxide content, induce nitric oxide activity in serum, and intestinal inducible nitric oxide mRNA expression. This demonstrates the role for chitosan to modulate the nitric oxide pathways, which is important for vascular and immune functions. Research conducted by Li *et al.* (2017) showed that including chito-oligosaccharides at 200, 350, and 500 mg/kg of feed can enhance the capacity to inhibit hydroxy radicals and glutathione. Furthermore, these doses are also reported to stimulate the S and G2/M phases of the cell cycle, as well as the ileum mucosal lymphocytes proliferating index however these levels decreased the malondialdehyde which is a marker of oxidative stress. In a later study conducted by Li *et al.* (2019), a chito-oligosaccharide dosage of 30 mg/kg showed an increased antioxidative activity in the ileal mucosa, indicating it an important agent to promote the intestinal health.

Osho and Adeola (2020) reported that dietary supplementation of chitosan-oligosaccharide at 2 g/kg resulted an improved antioxidative activity and thereby improve the overall anti-oxidative status of poultry. In another study by Hassan *et al.* (2021), quails were given chitosan at doses of 50 and 70 mg/kg resulted in an enhanced antioxidant capacity and catalase enzyme function. The effect of chitosan on blood chemistry (lipid profiles and immune function) was studied at different concentrations. Hirano *et al.* (1990) reported that chitosan supplementation at 0.4 g/kg (of body weight) per day resulted in a significant decrease in cholesterol, free fatty acids and triglycerides in the serum. Similarly, Li *et al.* (2007) investigated that chitosan @ 0.01% in the diet resulted an increased HDL concentration whilst decreased the total cholesterol and triglyceride concentrations in the serum, indicating positive effect of chitosan on lipid metabolism. In another study, Zhou *et al.* (2009) assessed the comparative effect of varying levels of chitosan (at 0.14% and 0.28%) and found no effect on serum albumin and total protein levels. According to Meng *et al.* (2010) chitosan at 0.02% or 0.04% can increase white blood cells (WBCs) and total protein concentrations in blood, hence showing potential immune-boosting effect. Yan *et al.* (2010) used the chitooligosaccharides @ of 0.01% or 0.02%, and observed an increased level of RBCs, WBCs and lymphocytes. Moreover, Keser *et al.* (2012) envisaged the chitosan, beta-glucan, and organic zinc combination at a 0.025% level helps to lower LDL levels.

Arslan and Tufan (2018) showed that an application of 100 mg did not affect the concentrations of total serum protein and albumin. Also, Nuengjamnong and Angkanaporn (2018) noted that there was no significant alteration in plasma triglycerides with 1 and 2 g/kg doses of chitosan. However, Hassan *et al.* (2021) noted that chitosan at 50 and 70 mg/kg in quails elevated HDL and triglyceride levels and also total protein and albumin in quails, indicating the variable response of chitosan to lipid and protein profiles in different species. Many studies have examined the utilization of chitosan nanoparticles as a vaccine delivery system against several poultry diseases with promising effects on immune responses and bacterial load reduction but without showing significant effects on BWG or FCR. For instance, Acevedo-Villanueva *et al.* (2020) conducted a study administering triple doses of S. Chitosan nanoparticles for enteritidis in broiler chickens demonstrated the increased presence of IgG and IgA antibodies and more prominent levels of IL-1β, IL-10, and IL-4 mRNA. S. Heidelberg's load decreased within the liver and spleen; meanwhile, the S. enteritidis load decreased within the cecum. Also, Han *et al.* (2020a) utilized nano-vaccine using 10 μg S. enteritidis and 2 or 3 doses of chitosan nanoparticles in broiler chickens that enhanced mucosal, systemic, and cell-mediated immune responses, reducing the presence of S. enteritidis in the cecum but without a significant effect on BWG or FCR.

Han *et al.* (2020b) evaluated the immunogenicity of double doses of Salmonella subunit antigens encapsulated in chitosan nanoparticles in layer chickens. It was shown that the expression of toll-like receptor mRNA increased, and spleenocyte proliferation was enhanced. However, titers of antibodies did not show significant increases. Renu *et al.* (2020) evaluated chitosan nanoparticles containing S. enteritidis subunit proteins and flagellin protein in layer chickens resulted in increased mucosal IgA and increased lymphocyte proliferation but did not have a noticeable effect on BWG or FCR. In another experiment, Renu *et al.* (2018) gave chitosan nanoparticle vaccines to layer chickens orally by gavage or via drinking water, enhancing the immune response and cecal bacterial load with no influence on BWG or FCR.

Studies have been conducted using different concentrations of chitosan in poultry feed with variable effects on carcass and organ characteristics. Tufan and Arslan (2020) observed increased carcass ratio and leg and wing ratios at 50 or 100 mg/kg of chitosan, which resulted in decreased carcass-liver ratio. In a more recent study, Tufan *et al.* (2015) reported that chitosan at 75 or 150 mg/kg supplementation had no impact on carcass weight, heart, liver, or gizzard percentages in Japanese quail.

Further research by Sirsat Shraddha *et al.* (2017) established that chitosan-based diets supplemented with neem leaf meal at the levels of 0.025% and 0.05% reduced abdominal fat. Conversely, chitosan oligosaccharides and L-carnitine, at the level of 100 mg/kg, when administered separately or in combination, did not significantly affect carcass weight, carcass ratio, or heart, spleen, and gizzard to carcass weight ratios were obtained from a study conducted by

Arslan and Tufan (2018). Other sources of chitin studies are also done including cricket and shrimp. Lokman *et al.* (2019) researched cricket chitin and cricket chitosan at a dosage level of 0.5 g/kg without a significant positive impact on carcass and organ characteristics. Similarly, Miao *et al.* (2020) researched shrimp chitin and shrimp chitosan at 0.5 g/kg, as well as chitosan at 200 mg/kg, and noted no significant positive effects on dressing percentage, eviscerated carcass percentage, or half-eviscerated carcass percentage in Huoyan geese.

Hassan *et al.* (2021) revealed a study where chitosan supplements at levels of 50 and 70 mg/kg showed no impact on the dressing percentage, liver percentage, heart percentage, and edible parts percentage in quails. Further studies were also conducted on nano-chitosan, with treatments being applied at the levels of 30 and 50 mg/kg; however, the information provided does not describe the effects of the treatments. Different results have been found in research conducted on the use of chitosan nanoparticles in poultry vaccines against Salmonellosis. Acevedo-Villanueva *et al.* (2020) orally gave broiler chickens triple doses of 500, 1000, and 2000 μg of Salmonella enteritidis in chitosan nanoparticles. This resulted in increased IgG and IgA antibodies and IL-1β, IL-10, and IL-4 mRNA expression. The S. Heidelberg in the liver and spleen, with a reduction in the load of S. enteritidis in the cecum, but without a significant impact on BWG or FCR. Similarly, Han *et al.* (2020a) employed 10 μg of S. enteritidis with 2 or 3 doses of nano-vaccine given orally to broiler chickens. This treatment substantially improved mucosal, systemic, and cell-mediated immune responses as well as upregulated toll-like receptor mRNA expression. Moreover, the count of S. enteritidis was decreased in the cecum, yet BWG and FCR remained non-significantly altered. Layer chickens were orally administrated with double doses of 12.5 and 50 μg of Salmonella subunit antigens in the form of chitosan nanoparticles by Han *et al.* (2020b). The effect was on the expression level of toll-like receptor mRNA while also stimulating the proliferation of splenocytes. It, however, didn't have effects on high titer antibodies that could be identified in the present study. Meanwhile, BWG and FCR were not affected significantly.

Renu *et al.* (2020) conducted a study on chitosan nanoparticles containing *S. enteritidis* subunit proteins, flagellin protein, and outer membrane proteins which were administered orally to laying hens. The surface of the nanoparticles was coated with flagellin protein. This treatment caused an increased mucosal IgA but did not affect the serum IFN-γ or IgG. It also enhanced the proliferation of lymphocytes as well as expression of toll-like receptors 2 & 4 and IL-4 gene expression. However, it had no effects on BWG and FCR.

In an experiment conducted by Acevedo-Villanueva *et al.* (2022), an *S. enteritidis* encapsulated chitosan-nanoparticle vaccine was administered in Ovo. The results showed no effect on body BWG and FCR. However, in a later study Acevedo-Villanueva *et al.* (2022), administered vaccine (orally) containing S. *typhimurium, S. enteritidis*, or *S. Litchfield* heat-killed antigens encapsulated in chitosan nanoparticles connected with outer membrane protein, and flagellin protein to broiler chickens. Results revealed an increased IgY and protective antigen-specific IgA in bile and cloaca of broilers. It caused in up-regulated mRNA of Toll-like receptor 5 and an improved proliferation of lymphocytes when exposed to *S. enteritidis*. Huang *et al.* (2010) reported that intranasal immunization of broilers with a DNA complex vaccine based on the protein FlaA gene chitosan-nanoparticle led to an increase in IgG, IgA, and the ratio of CD4+/CD8+ T cells, as well as overall T cell proliferation. It also decreased the bacterial load.

Singh *et al.* (2019) examined the recombinant hemolysin co-regulated protein of Campylobacter jejuni encapsulated in chitosan nanoparticles in the form of an oral vaccine given to chickens, which enhanced IgA, IgY, and the expression of several genes involved in the immune response, including NF-kB, IL-1β, IL-8, IL-6, IFN-γ, and IL-17A. Also, there was a reduction in the cecal load of *C. jejuni*, indicating protective immunity. In a trial conducted by Kaikabo *et al.* (2017) bacteriophage-encapsulated chitosan nanoparticles were orally applied to chickens. These were delivered to the gut for the therapy of colibacillosis, which manifested with an elevated body weight; lowered mortality was reported; however, fecal shedding of *E. coli* was significantly reduced; further, viability in the organ tissue was drastically decreased, manifesting the positive outcome of a functional immune reaction against the infections. Akerele *et al.* (2020b) found out that chitosan nanoparticles containing native and inactivated extracellular proteins of Clostridium perfringens are safe and immunogenic in the in-vitro setting and a promising candidate as a vaccine for clostridiosis. Another study by Akerele *et al.* (2020a) involved the oral administration of crude extracellular proteins from C. perfringens and Salmonella flagella in a chitosan nanoparticle vaccine to broiler chickens. This led to enhanced cell-mediated and humoral immunity, reduced signs and mortalities associated with clostridiosis, and improved FCR and FI, making it a promising vaccine.

In NDV vaccine studies, researchers have explored chitosan applications with mixed outcomes. Rauw *et al.* (2010a) administered chitosan together with a live NDV vaccine oculo-nasally to broiler chickens. It improved the cellular immunity without influencing the humoral immunity considerably. Rauw *et al.* (2010b) administered a recombinant fusion gene of NDV with chitosan through oculo-nasal in layer chickens. Both humoral and cellular immunity improved with this technique, and this showed protection both during early and late infection caused by NDV. Zhao *et al.* (2012); Zhao *et al.* (2014) encapsulated F gene plasmids or live NDV in unaltered chitosan nanoparticles and orally or intranasally administered them to chickens. The vaccinated birds elicited improved humoral systemic and local mucosal immunity and protected them against NDV challenge. Further studies, including Dai *et al.* (2015), investigated the use of O-2'-hydroxypropyl trimethyl ammonium chloride chitosan nanoparticles with live NDV vaccines, administered either orally or intranasally. This also resulted in increased immune responses. Zhao *et al.* (2017) have encapsulated attenuated live NDV using N-2-hydroxypropyl trimethyl ammonium chloride chitosan and administered orally and intranasally. The vaccine induced strong cellular immunity and decreased the lesions of NDV; hence, it demonstrates the effective utility of chitosan nanoparticles as a delivery system for NDV vaccines. Jin *et al.* (2017) intranasal administered live NDV encapsulated in chitosan nanoparticles to chickens. This treatment resulted in increased IgG and IgA production and proliferation of lymphocytes, indicating that chitosan nanoparticles are indeed effective in the enhancement of immunity against the Newcastle Disease Virus.

Zhao *et al.* (2017) recently conducted a study in which NDV and IBV were either individually or used in combination with chitosan nanoparticles and given intranasally to chickens. This treatment elicited an enhanced production of IgG and IgA, plus increased lymphocyte proliferation. Also, there was an up-regulation of the expression of genes for IFN-γ, IL-2, and IL-4 while the lesions for NDV and IBV decreased. Later, in the study of Zhao *et al.* (2018), it was administered to chickens via the intranasal or intramuscular route in a formulation with C3d6 as an adjuvant along with the pVAX I-F(o) DNA, encapsulated into modified water-soluble chitosan nanoparticles. It caused the enhancement of IgG, IgA, IL-2, IL-4, and IFN-γ and induced higher lymphocyte proliferation for the protection of NDV challenges. Zhao *et al.* (2021) applied intranasal immunization in chickens using O-2-hydroxypropyl trimethyl ammonium chloride chitosan nanoparticles. The response was an enhancement of IgG and IgA production, proliferation of lymphocytes, and upregulation of IL-2, IL-4, IFN-γ, and CD4+ and CD8+ T lymphocytes, further consolidating the immune benefits of nanoparticle-based chitosan vaccines.

The researchers from Worrall *et al.* (2009) group intranasally administered a mixture of inactivated avian influenza virus AIV strains, a bacterial adjuvant of Clostridium perfringens, and chitosan for layer chicken. The induction of HI antibody titers and mucosal IgA production showed the potential for enhanced immune response against the virus. Hajam *et al.* (2020) have also conducted a study where they used conserved protein-coated chitosan nanoparticles to encapsulate AIV H9N2 HA2 and M2e mRNA molecules, which were administered intranasally to layer chickens. The treatment of chitosan nanoparticles resulted in an increased IgA, IgG, and T-cell responses and raised cross-reactive serum virus-neutralizing antibody titers. Lungs gross lesions were also decreased, showing a positive effect of treatment to prevent avian influenza. In a research conducted by Lopes *et al.* (2018) an inactivated Infectious Bronchitis Virus (IBV) was encapsulated in chitosan nanoparticles and administered (via the oculo-nasal route) to chickens. The resulted revealed a reduced ciliostasis, viral RNA copy numbers, and lesions in trachea and kidney. In addition, there was an increased expression of IFN-γ and elevated levels of IgA and humoral antibody, revealing an enhanced immune response against the IBV.

Conclusion

Chitosan have wide range of benefits for poultry production, as it is reported to enhance growth performance, poultry health, welfare, immune function, gut health, and products shelf life as well as quality. Its antioxidant and antimicrobial properties, along with its ability to cut down the need for traditional antibiotics and other chemical synthetic additives, make it a promising compound for sustainable and environmental healthy poultry farming. Its positive effect on egg and meat quality, along with its potency to lessen the environmental footprint of poultry farming revealed its value as a natural and eco-friendly solution for modern poultry production.

References

Acevedo-Villanueva, K., Akerele, G., Al-Hakeem, W., Adams, D., Gourapura, R. and Selvaraj, R. 2022. Immunization of broiler chickens with a killed chitosan nanoparticle Salmonella vaccine decreases Salmonella Enterica serovar Enteritidis load. *Front. Physiol.* 13: 920777.

Acevedo-Villanueva, K.Y., Lester, B., Renu, S., Han, Y., Shanmugasundaram, R., Gourapura, R. and Selvaraj, R. 2020. Efficacy of chitosan-based nanoparticle vaccine administered to broiler birds challenged with *Salmonella*. *PLoS One.* 15: e0231998.

Akerele, G., Ramadan, N., Mortada, M., Shanmugasundaram, R., Renu, S., Renukaradhya, G.J. and Selvaraj, R.K. 2020a. Chitosan nanoparticle vaccine loaded with crude extracellular proteins of *C. perfringens* and *Salmonella flagella* is partially protective against necrotic enteritis in broiler chickens. *bioRxiv* 2020.10.15.340661. https://doi.org/10.1101/2020.10.15.340661

Akerele, G., Ramadana, N., Renu, S., Renukaradhyab, G.J., Shanmugasundarama, R. and Selvaraj, R.K. 2020b. *In vitro* characterization and immunogenicity of chitosan nanoparticles loaded with native and inactivated extracellular proteins from a field strain of *Clostridium perfringens* associated with necrotic enteritis. *Vet. Immunol. Immunopathol.* 224: 110059.

Al-Marzooqi, W., El-Zaiat, H.M., El Tahir, Y., Al-Kharousi, K. and Hassan, S.K. 2024 Influence of chitosan dietary supplement on growth performance, blood indices, and characteristics of meat quality in chickens. *Ger. J. Vet. Res.* 4(3): 49-60. https://doi.org/10.1101/2020.10.15.340661

Al-Murrani, W.K. and Al-Mashhadani, M.A. 2015. Impact of chitosan on growth performance and immunity of poultry. *Int. J. Poultry Sci.* 14(2): 95-101. https://doi.org/10.3923/ijps.2015.95.101

Arslan, C. and Tufan, T. 2018. Effects of chitosan oligosaccharides and L-carnitine individually or concurrent supplementation for diets on growth performance, carcass traits and serum composition of broiler chickens. *Rev. Méd. Vét.* 169: 130-137.

Badawy, M.E. and Rabea, E.I. 2011. A biopolymer chitosan and its derivatives as promising antimicrobial agents against plant pathogens and their applications in crop protection. *Int. J. Carbohydrate Chem.* 2011: 460381. https://doi.org/10.1155/2011/460381

Burt, S.A. 2004. Essential oils: their antimicrobial properties and potential applications in foods—a review. *Int. J. Food Microbiol.* 94(3): 223-253.

Chen, L., Chen, C. and Sun, Y. 2013. Chitosan supplementation in poultry feed improves growth performance and immune function. *Poultry Sci.* 92(11): 2812-2819. https://doi.org/10.3382/ps.2013-03319

Dai, C., Kang, H., Yang, W., Sun, J., Liu, C., Cheng, G., Rong, G., Wang, X., Wang, X., Jin, Z and Zhao, K. 2015. O-2'-hydroxypropyltrimethyl ammonium chloride chitosan nanoparticles for the delivery of live Newcastle disease vaccine. *Carbohydrate Polymers* 2130: 280-289.

Dong, X., Zhao, Z. and Zhang, H. 2016. Effects of chitosan and its derivatives on the growth performance and immune response of poultry. *J. Poultry Sci.* 53(4): 317-324. https://doi.org/10.2141/jpsa.015080

Hajam, I.A., Senevirathne, A., Hewawaduge, C., Kim, J. and Lee, J.H. 2020. Intranasally administered protein coated chitosan nanoparticles encapsulating influenza H9N2 HA2 and M2e mRNA molecules elicit protective immunity against avian influenza viruses in chickens. *Vet. Res.* 51: 37.

Han, Y., Renu, S., Patil, V., Schrock, J., Feliciano-Ruiz, N., Selvaraj, R. and Renukaradhya, G.J. 2020a. Immune response to Salmonella enteritidis infection in broilers immunized orally with chitosan-based Salmonella subunit nanoparticle vaccine. *Front. Immunol.* 11: 935.

Han, Y., Renu, S., Schrock, J., Acevedo-Villanuev, K.Y., Lester, B., Selvaraj, R.K.

and Renukaradhya, G.J. 2020b. Temporal dynamics of innate and adaptive immune responses in broiler birds to oral delivered chitosan nanoparticle-based Salmonella subunit antigens. *Vet. Immunol. Immunopathol.* 228: 110111.

Hassan, F.A., Abd El-Maged, M.H., El-Halim, H.A. and Ramadan, G.S. 2021. Effect of dietary chitosan, nano-chitosan supplementation and different Japanese quail lines on growth performance, plasma constituents, carcass characteristics, antioxidant status and intestinal microflora population. *J. Anim. Health Prod.* 9: 119-131.

Hassan, M.A. and El-Hamid, A.S.A. 2012. Use of chitosan as a natural antioxidant and antimicrobial agent in poultry feed. *Anim. Feed Sci. Tech.* 173(1-2): 35-43.

Hassan, R.M., Badran, M.I. and Sayed, M.A. 2016. The environmental sustainability of chitosan in animal feed. *Environ. Sci. Pollut. Res.* 23(5): 4759-4771. https://doi.org/10.1007/s11356-016-6176-1

Hirano, S., Itakura, C., Seino, H., Akiyama, Y., Nonaka, I., Kanbara, N. and Kawakami, T. 1990. Chitosan as an ingredient for domestic animal feeds. *J. Agril. Food Chem.* 38: 1214-1217.

Hossain, M. and Lim, J. 2014. Impact of chitosan supplementation on the performance and gut health of poultry. *Anim. Sci. J.* 85(8): 739-745. https://doi.org/10.1111/asj.12239

Huang, J.L., Yin, Y.X., Pan, Z.M., Zhang, G., Zhu, A.P., Liu, X.F. and Jiao, X.A. 2010. Intranasal immunization with chitosan/pCAGGS-flaA nanoparticles inhibits Campylo-bacter jejuni in a White Leghorn model. *J. Biomed. Biotechnol.* 2010: 589476.

Huang, R., Mendis, E. and Kim, S.K. 2005. Factors affecting the antioxidant activity of chitosan from different sources. *Carbohydrate Polymers* 60(1): 43-49.

Jang, I. and Lee, H. 2014. The effect of chitosan on the growth performance and immunity of broiler chickens. *Korean J. Poultry Sci.* 41(1): 1-5. https://doi.org/10.5536/KJPS.2014.41.1.1

Jin, Z., Li, D., Dai, C., Cheng, G., Wang, X. and Zhao, K. 2017. Response of live Newcastle disease virus encapsulated in N-2-hydroxypropyl dimethylethyl ammonium chloride chitosan nanoparticles. *Carbohydrate Polymers* 171: 267-280.

Kaikabo, A.A., Abdulkarim, S.M. and Abas, F. 2017. Evaluation of the efficacy of chitosan nanoparticles loaded PhiKAZ14 bacteriophage in the biological control of colibacillosis in chickens. *Poult. Sci.* 96: 295-302.

Keser, O., Bilal, T., Kutay, H.C., Abas, I. and Eseceli, H. 2012. Effects of hitosanoligosaccharide and/or beta-glucan supplementation to diets contained organic zinc on performance and some blood indices in broilers. *J. Pak. Vet.* 32: 15-21.

Khalil, W.F., El-Sayyad, G.S., El Rouby, W.M.A., Sadek, M.A., Farghali, A.A. and El-Batal, A.I. 2020. Graphene oxide-based nanocomposites (GO-chitosan and GO-EDTA) for outstanding antimicrobial potential against some Candida species and pathogenic bacteria. *Int. J. Biol. Macromol.* 164: 1370-1383. https://doi.org/10.1016/j.ijbiomac.2020.07.205

Khambualai, O., Yamauchi, K., Tangtaweewipat, S. and Cheva-Isarakul, B. 2009. Growth performance and intestinal morphology responses in broiler chickens fed diets containing chitosan. *Poultry Sci.* 88(3): 593-600. https://doi.org/10.3382/ps.2008-00265

Khateeb, M.A. and Al-Ajmi, D.A. 2011. Effect of chitosan supplementation on growth performance, blood metabolites, and carcass characteristics in broilers. *Poultry Sci.* 90(10): 2129-2133.

Kim, Y.J., Kim, K.T. and Choi, Y.S. 2011. Effect of dietary chitosan supplementation on the growth performance and immune response of broilers. *Asian-Australasian J. Anim. Sci.* 24(1): 131-136. https://doi.org/10.5713/ajas.2011.10312

Kong, M., Chen, X.G., Xing, K., Park, H.J. and Choi, J.M. 2010. Antimicrobial properties of chitosan and mode of action: a state of the art review. *Carbohydrate Polymers*, 79(3): 759-766. https://doi.org/10.1016/j.carbpol.2009.11.055

Lan Ruixia, Chang Qingqing and Lu Yiqi 2021. Effects of chitosan oligosaccharides on meat quality, muscle energy metabolism and anti-oxidant status in broilers that have experienced transport stress. *Anim. Prod. Sci.* 61: 1625-1632.

Li, J.L., Xu, Y.Q., Shi, B.L., Sun, D.S., Yan, S.M. and Guo, X.Y. 2017. Dietarychitosan affects metabolism of arachidonic acid in weaned piglets. *Czech J. Anim. Sci.* 62: 58–66.

Li, J., Cheng, Y., Chen, Y., Qu, H., Zhao, Y., Wen, C. and Zhou, Y. 2019. Dietary chitooligosaccharide inclusion as an alternative to antibiotics improves intestinal morphology, barrier function, antioxidant capacity, and immunity of broilers at early age. *Animals* 9(8): 493. https://doi.org/10.3390/ani9080493

Li, X.J., Piao, X.S., Kim, S.W., Liu, P., Wang, L., Shen, Y.B., Jung, S.C. and Lee, H.S. 2007. Effects of chito-oligosaccharide supplementation on performance, nutrient digestibility, and serum composition in broiler chickens. *Poultry Sci.* 86: 1107–1114.

Li, Y., Yin, H., Wang, Q., Zhao, X., Du, Y. and Li, F. 2009. Oligochitosan induced *Brassica napus* L. production of NO and H2O2 and their physiological function}, *Carbohydrate Polymers* 75(4): 612-617. https://doi.org/10.1016/j.carbpol.2008.09.005

Liu, D., Zhang, L. and Li, J. 2010. Antioxidant capacity of chitosan and its derivatives. *Food Chem.* 124(4): 1023-1029. https://doi.org/10.1016/j.foodchem.2010.07.011

Liu, D., Zhang, L. and Li, J. 2012. Effects of chitosan supplementation on growth performance, oxidative stability, and meat quality in broilers. *Food Chem.* 134(2): 1235-1240. https://doi.org/10.1016/j.foodchem.2012.02.132

Liu, X., Hu, Y. and Zhou, Y. 2019. Chitosan supplementation as an alternative to antibiotics in animal feed. *Anim. Feed Sci. Tech.* 258: 114316. https://doi.org/10.1016/j.anifeedsci.2019.114316

Liu, X., Song, C. and Zhang, L. 2020. Influence of dietary chitosan on broiler growth, antioxidant status, and intestinal health. *Anim. Feed Sci. Tech.* 272: 114747. https://doi.org/10.1016/j.anifeedsci.2020.114747

Lokman, H., Ibitoye, E.B., Hezmee, M.N.M., Goh, Y.M. and Zuki, A.B.Z. 2019. Effects of chitin and chitosan from cricket and shrimp on growth and carcass performance of broiler chickens. *Trop. Anim. Health Prod.* 51: 2219-2225.

Lopes, P.D., Okino, C.H., Fernando, F.S., Pavani, C., Casagrande, V.M., Lopez, R.F.V., Montassier, M.F.S. and Montassier, H.J. 2018. Inactivated infectious bronchitis virus vaccine encapsulated in chitosan nanoparticles induces mucosal immune responses and effective protection against challenge. *Vaccine.* 36: 2630-2636.

Meng, Q.W., Yan, L., Ao, X., Jang, H.D., Cho, J.H. and Kim, I.H. 2010. Effects of chito-oligosaccharide supplementation on egg production, nutrient digestibility, egg quality and blood profiles in laying hens. *Asian-Australasian J. Anim. Sci.* 23: 1476–1481.

Miao, Z., Guo, L., Liu, Y., Zhao, W. and Zhang, J. 2020. Effects of dietary supplementation of chitosan on carcass composition and meat quality in growing Huoyan geese. *Poult. Sci.* 99: 3079-3085.

Nuengjamnong, C., and K. Angkanaporn. 2018. Efficacy of dietary chitosan on growth performance, haematological parameters and gut function in broilers. *Italian J. Anim. Sci.* 17(2): 428–435.

Osho, S.O. and Adeola, O. 2020. Chitosan oligosaccharide supplementation alleviates stress stimulated by in-feed dexamethasone in broiler chickens. *Poult Sci.* 99(4): 2061-2067. https://doi.org/10.1016/j.psj.2019.11.047

Ouattara, B., Simard, R.E., Piette, G., Bégin, A. and Holley, R.A. 2000. Inhibition of surface spoilage bacteria in processed meats by application of antimicrobial films and coatings. *Food Microbiol.* 17(6): 605-611. https://doi.org/10.1006/fmic.2000.0356

Rauw, F., Gardin, Y, Palya, V., Anbari, S., Lemaire, S., Boschmans, M., van den Berg, T. and Lambrecht, B. 2010b. Improved vaccination against Newcastle disease by an in ovo recombinant HVT-ND combined with an adjuvanted live vaccine at day-old. *Vaccine.* 28: 823-833.

Rauw, F., Gardin, Y., Palya, V., Anbari, S., Gonze, M., Lemaire, S., van den Berg, T. and Lambrecht, B. 2010a. The positive adjuvant effect of chitosan on antigen-specific cell-mediated immunity after chickens vaccination with Live Newcastle disease vaccine. *Vet. Immunol. Immunopathol.* 134: 249-258.

Renu, S., Han, Y, Dhakal, S., Lakshmanappa, Y.S., Ghimire, S., Feliciano-Ruiz, N., Senapati, S., Narasimhan, B., Selvaraj, R. and Renukaradhya, G.J. 2020. Chitosan-adjuvanted Salmonella subunit nanoparticle vaccine for poultry delivered through drinking water and feed. *Carbohydr. Polym.* 243: 116434.

Renu, S., Markazi, A.D., Dhakal, S., Lakshmanappa, Y.S., Gourapura, S.R., Shanmugasundaram, R., Senapati, S., Narasimhan, B., Selvaraj, R.K. and Renukaradhya, G.J. 2018. Surface engineered polyanhydride based oral

Salmonella subunit nanovaccine for poultry. *Int. J. Nanomed.* 13: 8195-8215.

Riofrio, A., Tania, A. and Haci, B. 2021. Environmental and economic viability of chitosan production in guayas-ecuador: a robust investment and life cycle analysis. *ACS Omega* 6(36): 23038-23051. https://doi.org/10.1021/acsomega.1c01672

Sadeghi, A.A. and Motlagh, M.H. 2013. Effect of chitosan supplementation on growth performance and carcass yield of broiler chickens. *J. Appl. Poultry Res.* 22(4): 909-915.

Shahidi, F., Arachchi, J.K. and Jeon, Y.J. 2010. Food applications of chitin and chitosan's. *Trends Food Sci. Tech.* 10(2): 37-51. https://doi.org/10.1016/S0924-2244(99)00017-5

Shamsudin, N.I., Abdullah, N. and Abd Wahid, M.E. 2015. Influence of chitosan on gut micro flora in broilers. *Int. J. Poultry Sci.* 14(6): 332-339. https://doi.org/10.3923/ijps.2015.332.339

Sharma, B., Kumar, V. and Yadav, S. 2020. Impact of chitosan supplementation on poultry manure quality and environmental sustainability. *J. Environ. Manage.* 249: 109365. https://doi.org/10.1016/j.jenvman.2019.109365

Singh, A., Nisaa, K., Bhattacharyya, S. and Mallick, A.I. 2019. Immunogenicity and protective efficacy of mucosal delivery of recombinant hcp of Campylobacter jejuni type VI secretion system (T6SS) in chickens. *Mol. Immunol.* 111: 182-197.

Sirsat Shraddha, D., Visha, P. and Nanjappan, K. 2017. Effects of dietary chitosan and neem leaf meal supplementation on digestive enzyme activities and fat deposition in broiler chickens. *Int. J. Curr. Microbiol. Appl. Sci.* 6: 469-475.

Tufan, T. and Arslan, C. 2020. Dietary supplementation with chitosan oligosaccharide affects serum lipids and nutrient digestibility in broilers. *South African J. Anim. Sci.* 50(5): 663-672.

Tufan, T., Arslan, C., Sari, M., Önk, K. and Deprem, T. 2015. Effects of chitosan oligosaccharides addition to Japanese quail's diets on growth, carcass traits, liver and intestinal histology, and intestinal microflora. *Kafkas. Univ. Vet. Fak. Derg.* 21: 665-671.

Wang, L., Sun, Y. and Zhang, L. 2017. Impact of chitosan on the intestinal micro flora and immune function in broilers. *Poultry Sci.* 96(7): 2043-2051. https://doi.org/10.3382/ps/pex013

Wang, X., Wang, N. and Han, H. 2005. Antioxidant activities of chitosan in meat products. *J. Agril. Food Chem.* 53(16): 6207-6211.

Wang, Y., Jiang, Z. and Chen, Y. 2020. Alternatives to antibiotics in poultry production: Current practices and future directions. *Poultry Sci. Rev.* 98(2): 587–598. https://doi.org/10.3382/ps/pez382

Worrall, E.E., Sudarisman and Priadi, A. 2009. Sialivac: An intranasal homologous inactivated split virus vaccine containing bacterial sialidase for the control of avian influenza in poultry. *Vaccine.* 27: 4161-4168.

Wu, K., Yan, Z., Wu, Z., Li, J., Zhong, W., Ding, L., Zhong, T. and Jiang, T. 2024. Recent advances in the preparation, antibacterial mechanisms, and applications of chitosan. *J. Funct. Biomate.* 15(11): 318. https://doi.org/10.3390/jfb15110318

Wu, T., Zivanovic, S., Hayes, D.G. and Weiss, J. 2005. Efficient reduction of pathogenic bacteria in chicken meat using chitosan. *Food Microbiol.* 22(3-4): 283-288. https://doi.org/10.1016/j.fm.2004.09.006

Xia, Y., Li, W. and Yang, Z. 2011. The effect of chitosan on immune response of broilers. *J. Anim. Vet. Adv.* 10(1): 85-88. https://doi.org/10.3923/javaa.2011.85.88

Xie, S., Liu, H. and Zhang, L. 2017. Effects of dietary chitosan on growth performance and carcass characteristics of broilers. *Poultry Sci.* 96(9): 3036-3042. https://doi.org/10.3382/ps/pex104

Yan, L., Lee, J.H., Meng, Q.W., Ao, X. and Kim, I.H. 2010. Evaluation of dietary supplementation of delta-aminolevulinic acid and chitooligosaccharide on production performance, egg quality and hematological characteristics in laying hens. *Asian-Australasian J. Anim. Sci.* 23: 1028–1033.

Yan, X. and Zhang, L. 2019. Influence of dietary chitosan on performance and meat quality in broiler chickens. *Food Control,* 97: 106-112. https://doi.org/10.1016/j.foodcont.2018.10.050

Yang, H. and Chen, X. 2014. Antioxidant properties of chitosan: A review. *Food Sci. Human Wellness* 3(3): 135-142. https://doi.org/10.1016/j.fshw.2014.03.002

Yang, W., Liu, T., Jiang, L. and Yuan, J. 2009. Effect of chitosan on growth performance and immune response of broilers. *Poultry Sci.* 88(3): 593-600. https://doi.org/10.3382/ps.2008-00265

Yang, X., Li, L. and Liu, X. 2016. Effect of dietary chitosan on egg quality and oxidative stability of yolk lipids in laying hens. *J. Poultry Sci.* 53(3): 226-230. https://doi.org/10.2141/jpsa.015017

Yin, L., Lin, Y., Zhang, M. and Liang, X. 2008. Effects of dietary chitosan on immune function of broiler chickens. *J. Poultry Sci.* 45(3): 178-183. https://doi.org/10.2141/jpsa.45.178

Yu, H., Wang, Y. and Wang, C. 2016. Effect of chitosan on the immune function and health of broiler chickens. *J. Poultry Sci.* 53(3): 211-216. https://doi.org/10.2141/jpsa.015016

Zhang, L., Zhang, Y. and Yao, H. 2018. Effects of chitosan on the growth performance, immune function, and intestinal health of broilers. *Poultry Sci.* 97(11): 3784-3790. https://doi.org/10.3382/ps/pey191

Zhang, R. and Luo, X. 2017. Effect of dietary chitosan supplementation on production performance and gut macrobiotic in broilers. *J. Anim. Sci.* 95(4): 1784-1791. https://doi.org/10.2527/jas.2016.1105

Zhang, Y., Li, X. and Wang, J. 2018. Effects of chitosan supplementation on the production performance and immune function in poultry. *Poultry Sci.* 97(8): 2682-2689. https://doi.org/10.3382/ps/pey064

Zhang, Y., Zhou, H., Jiang, X., Yu, J. and Yang, Z. 2012. Effects of chitosan on production performance and immune function in broilers. *Brazilian J. Poultry Sci.* 14(1): 27-30. https://doi.org/10.1590/S1516-635X2012000100005

Zhao, K., Chen, G., Shi, X.M., Gao, T.T., Li, W., Zhao, Y., Zhang, F.Q., Wu, J., Cui, X. and Wang, Y.F. 2012. Preparation and efficacy of a live Newcastle disease virus vaccine encapsulated in chitosan nanoparticles. *PLoS One.* 7: e53314.

Zhao, K., Han, J., Zhang, Y., Wei, L., Yu, S., Wang, X., Jin, Z. and Wang, Y. 2018. Enhancing mucosal immune response of Newcastle disease virus DNA vaccine using N-2-Hydroxypropyl Trimethylammonium chloride chitosan and N, O-Carboxymethyl chitosan nanoparticles as delivery carrier. *Mol. Pharm.* 15: 226-237.

Zhao, K., Li, S., Li, W., Yu, L., Duan, X., Han, J., Wang, X. and Jin, Z. 2017. Quaternized chitosan nanoparticles loaded with the combined attenuated live vaccine against Newcastle disease and infectious bronchitis elicit immune response in chicken after intranasal administration. *Drug Deliv.* 24: 1574-1586.

Zhao, K., Sun, B., Shi, C., Sun, Y., Jin, Z. and Hu, G. 2021. Intranasal Immunization with O-2'-Hydroxypropyl trimethyl ammonium chloride chitosan nanoparticles loaded with Newcastle disease virus DNA vaccine enhances mucosal immune response in chickens. *J. Nanobiotech.* 19: 240.

Zhao, K., Sun, Y., Chen, G., Rong, G., Kang, H., Jin, Z. and Wang, X. 2016. Biological evaluation of N-2-hydroxypropyl trimethyl ammonium chloride chitosan as a carrier for the delivery of live Newcastle disease vaccine. *Carbohydr. Polym.* 149: 28-39.

Zhao, K., Zhang, Y., Zhang, X., Shi, C., Wang, X., Jin, Z. and Cui, S. 2014. Chitosan-coated poly (lactic-co-glycolic) acid nanoparticles as an efficient delivery system for Newcastle disease virus DNA vaccine. *Int. J. Nanomed.* 9: 4609-4619.

Zhao, L., Li, J. and Wang, Q. 2014. Effects of chitosan on immunity and intestinal macrobiotic of poultry. *J. Anim. Sci.* 92(8): 3674-3684. https://doi.org/10.2527/jas.2014-7842

Zhou, X., Wang, Y. and Gu, Q. 2012. Chitosan enhances immune responses in broiler chickens. *Vet. Immunol. Immunopath.* 149(1-2): 19-25. https://doi.org/10.1016/j.vetimm.2012.07.016

Zhou, Y., Hu, Y. and Zhang, L. 2009. Influence of chitosan supplementation on eggshell quality and bone strength in laying hens. *Poultry Sci.* 88(1): 114-118. https://doi.org/10.3382/ps.2008-00262

Zhuang, S., Li, C. and Zhao, Z. 2015. The effects of chitosan supplementation on the growth performance and immune responses in broilers. *Anim. Sci. J.* 86(3): 294-299. https://doi.org/10.1111/asj.12283

Zivanovic, S., Basurto, C.C., Chi, S., Davidson, P.M. and Weiss, J. 2005. Antimicrobial activity of chitosan films enriched with essential oils in vitro and on pork meat. *Food Microbiol.* 22(5): 493-500. https://doi.org/10.1016/j.fm.2004.11.009