Role of Probiotics and Prebiotics in Poultry

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Abstract

Probiotics and prebiotics play a very important role in poultry by increasing the growth, health, and production of poultry birds. Probiotics are live microorganisms that have a good effect on host health while prebiotics are indigestible feed ingredients that increase beneficial bacteria in the gut. These have been commonly used as an alternative to antibiotics because of increasing antibiotic resistance. They also improve the immune system, growth performance, and nutrient absorption, decrease illness, reduce pathogen load, and stimulate beneficial bacteria in poultry birds. Lactobacillus spp., Enterococcus spp., Bacillus spp., and *Saccharomyces cerevisiae* (yeast) are commonly used probiotics in poultry. While, Mannan-oligosaccharides, fructo-oligosaccharides, galacto-oligosaccharides, beta-glucans, and inulin are commonly used as prebiotics in poultry. Probiotics work by two types of mechanisms; Competitive exclusion and immune system regulation. This chapter defines prebiotics, probiotics, and their criteria for selection. It also explains the mechanism of action of probiotics and the role of prebiotics and probiotics in poultry. Moreover, it also describes the limitations of their usage.

Keywords: Limitations, Lactobacillus species, Mode of action, Oligosaccharides, Probiotics and Prebiotics, Selection criteria

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Introduction

The poultry industry has become an important part of human activities because it provides food and a source of income in different countries, globally. With the increase in human population, demand for safe food has become increased (Reuben et al., 2021). According to the FAO report, the global intake of poultry meat and egg will rise by 52 and 39% respectively, in 2050 (Krysiak et al., 2021; Susanti et al., 2021). So, this increased demand requires safe products. Previously, antibiotics have been used for disease treatment and as a growth promoter (Biswas et al., 2021; Krysiak et al., 2021). As antibiotic resistance has increased, the demand for synthetic products become less and attention has been directed toward safe, non-toxic, antibiotic-free, economical, and natural alternatives like probiotics and prebiotics (Reuben et al., 2021).

Probiotics are defined as live beneficial microbes that give a good effect on the host's health when ingested by the digestive pathway (Wang et al., 2023). While prebiotics are indigestible food ingredients that enhance good bacteria (lactobacillus, bifidobacteria) which further improves gut and host health (Ahlawat et al., 2021). Different kinds of microorganisms can be used as probiotics like lactobacillus spp., Bifidobacterium spp., Enterococcus spp., *Streptococcus thermophilus*, and *Escherichia coli* (Ranadheera et al., 2017; Kerry et al., 2018). While, Mannan-oligosaccharides (MOS), galacto-oligosaccharides (GOS), fructo-oligosaccharides (FOS), xylooligosaccharides (XOS), beta-glucans, and inulin are commonly used as prebiotics in poultry (Song et al., 2020; Morgan, 2023; Ezzat et al., 2024; Youssef et al., 2024).

Gut health is described as the equilibrium of bacteria within different parts of the gastrointestinal tract (Aruwa et al., 2021). Bird's highest efficiency showed that the bacteria are balanced in the gut while, imbalance occurs in stressed bird's gut when good bacteria (lactobacilli) are lowest in number or harmful bacteria are increased. This can lead to enteric (diarrhea), or subclinical diseases and decrease the production of birds such as growth, feed efficiency, etc. (Riva, 2020; Gill, 2023). Although the beneficial microbial gut flora is extremely stable, but dietary and environmental conditions can create instability in it. The conditions that can create instability are stress, extreme hygiene, and antibiotic treatment. In the wild, chickens are protected from infections by obtaining gut flora from their mother's feces (Gill, 2023). But commercially raised chickens are hatched in sterile incubators and lack those bacteria that are commonly detected in chicken guts (Rychlik, 2020). The features of gut microbiota may be altered by the impact of shell microbial contamination. Further, HCL secretion (at 18 days of incubation) also affects microflora choice. That's why, there is an urgent need of probiotics for poultry birds compared to other animals (Anwar et al., 2023; Gill, 2023).

Apart from poultry, prebiotics and probiotics have been also used in humans and other animals like large ruminants, small ruminants, and pigs. These may be used as a single or a combination. (Cangiano et al., 2020; Grosu & Caisîn, 2020; Zommiti et al., 2020; Zapata et al., 2021). A combination of prebiotics and probiotics is called a synbiotic (Dev et al., 2021). Nanoparticles have been used with probiotics and prebiotics that give better results in different fields (Mason 2021). This chapter is about the role of probiotics and prebiotics in poultry. It also explains the mechanism of action of probiotics. Criteria for selection and limitations of probiotics and prebiotics.

Criteria of Selection for Probiotics and Prebiotics

Probiotic bacteria should contain some necessary properties: normally present in the gut, and it has the intestinal adherence ability to get over potential barriers like lower stomach pH, the existence of intestinal bile acids, and competition with other intestinal microbes (Dixon et al., 2022). Potential probiotics should increase host nutrition and immune response (Zommiti et al., 2020; Raheem et al., 2021). Probiotic has to be technologically compatible with industrial operations and stable under typical storage conditions (Wang & Zhong, 2024).

There are many species that are utilized in probiotic formations (Wang et al., 2023) such as *Lactobacillus plantarum*, *Lactobacillus bulgaricus*, *Lactobacillus salivarius*, *Lactobacillus casei*, *Lactobacillus lactis*, *Lactobacillus acidophilus*, *Lactobacillus helveticus*, *Enterococcus faecium*, *Enterococcus faecalis, Streptococcus thermophilus*, *Escherichia coli.*, and *Bifidobacterium spp.* as shown in Figure 1. From these, *L. bulgaricus* and *Strep. thermophilus*, are yogurt starter microbes. *Saccharomyces cerevisiae* species are also included in probiotics (Ranadheera et al., 2017; Kerry et al., 2018).

Prebiotics must contain some characteristics such as a particular substance that increases the functionality of some good bacteria, can beneficially change microbiota, is not dissolved or digested in the upper portion of the gastrointestinal tract, and has the ability to introduce some beneficial effects (systemic or luminal) on the host (Ferreira-Lazarte et al., 2021; Bełdowska et al., 2024; Richards et al., 2024; Youssef et al., 2024).

Nondigestible carbohydrates have to be consumed by endogenous colonial bacteria and do not digest in the upper part of the gastrointestinal tract (Yaqoob et al., 2021). Common prebiotics used in the poultry industry is nondigestible oligosaccharides such as GOS, MOS, XOS, FOS, inulin, and non-starch polysaccharides e.g., β-glucan (Song et al., 2020; Morgan, 2023; Ezzat et al., 2024; Youssef et al., 2024). These prebiotics can be extracted from plants and are produced from microorganisms (Anwar et al., 2023; Teague et al., 2023).

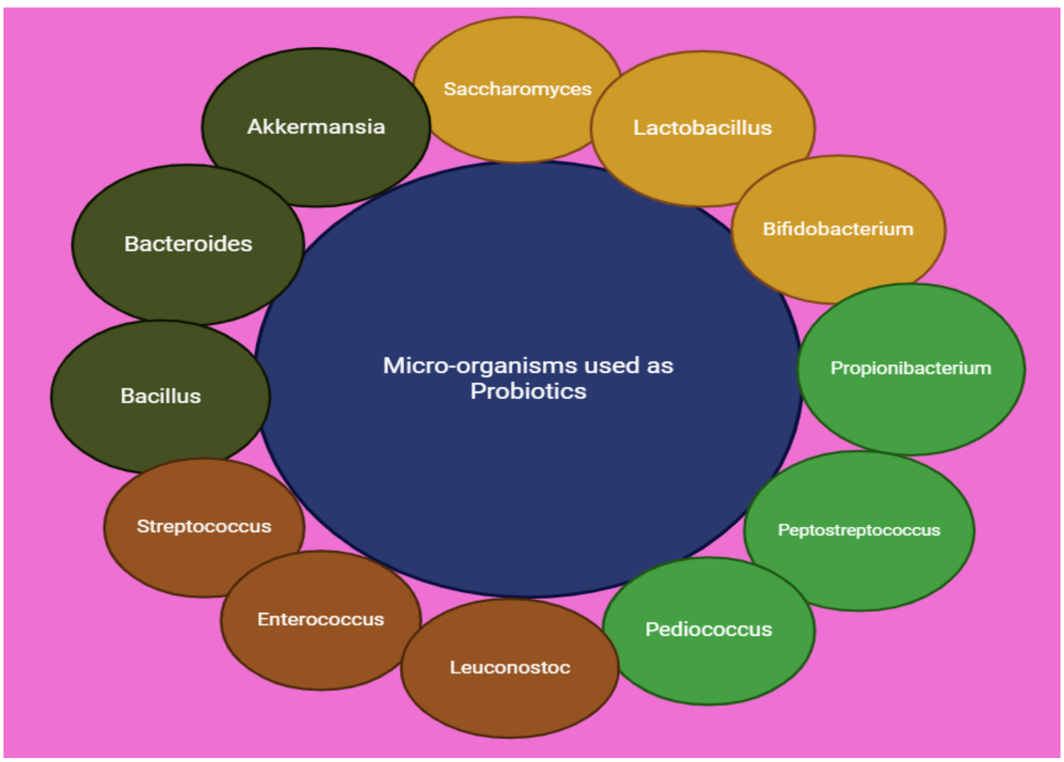

Fig. 1: Micro-organisms used as probiotics

Mechanism of Action of Probiotics

Competitive exclusion and immune system regulation are two types of mechanisms of action of probiotics (Mehmood et al., 2023). Competitive exclusion of microbes by probiotics involves: the generation of inhibitory substances (defensins, mucins, bacteriocins, etc.), inhibiting microbial adhesion, rivalry for nutrition, toxin bioavailability, and modulating host immunity (Hernandez-Patlan et al., 2020).

Various kinds of inhibitory compounds (antimicrobial peptides or AMPs) are generated by probiotics that plays an important part in lessening microbes' invasion. These peptides include bacteriocins (ribosomal synthesized AMP), diacetyl (arginine-binding protein of G^- bacteria), ethanol, hydrogen peroxide, and organic acids (Liao & Nyachoti, 2017). Probiotics block pathogenic bacteria attachment with the binding site of intestinal epithelium by using competitive inhibition (Fusco et al., 2023). It stimulates the mucus immune system and promotes the synthesis of mucins (glycosylated glycoproteins) and defensins that increase the intestinal barrier. Mucins are the main macromolecular components of mucus that are generated by epithelial cells of the intestine and block the attachment and colonization of harmful bacteria. The probable exclusion of harmful bacteria is a result of the particular link between the intraepithelial lymphocytes (IEL) and the surface proteins of probiotic bacteria. Mucin also alters immunity (Fusco et al., 2023). Defensins (membrane-disrupting peptides) are minute cationic peptides that cause mortality or stop the spread of bacteria by causing direct membrane damage or blocking bacterial cell wall formation (Kagan et al., 1990; Fusco et al., 2023). The electrostatic connection of anionic phospholipid groups of the membrane of epithelium has the ability to disrupt and kill harmful bacteria by producing pores in the cell membrane (Kagan et al., 1990). Moreover, it takes part in toxin neutralization produced by harmful bacteria (Ansari et al., 2024). Adherence of IEL to probiotics not only results in competitive depletion of vital nutrients but also shrinks the pathogen-binding area of the GIT. As a result of it, the expansion and colonization of harmful bacteria is decreased (Fusco et al., 2023). Probiotics (lactobacillus) decrease the gut cells' absorption of toxic compounds. The decreased level of toxins in the gut is due to the beneficial effect of lactic acid bacteria-based probiotics (Liao & Nyachoti, 2017). Mycotoxins are dangerous chemicals for animals and lactic acid bacteria act as a natural barrier against these chemicals (Tsai et al., 2012; Peng et al., 2018). Some strains also eliminate the harmful effects of aflatoxins in animals and humans (Abbès et al., 2016; Li et al., 2017).

Probiotics have been identified for their immunomodulatory properties (Mehmood et al., 2023). The innate immune system blocks pathogens and infection spread by increasing IEL cells. Probiotics also enhance the epithelial barrier by increasing mucus and yield of antimicrobial peptides (Fusco et al., 2023). Active immunity initiates with the association of pattern recognition receptor (PRR) of antigen-presenting cells (like dendritic cells) which liberate T and B cells (produce antibodies) (Lebeer et al., 2010; Iwasaki & Medzhitov, 2015) that shows the efficacy of probiotic candidate e.g, LAB (Fusco et al., 2023).

Innate immunity has IEL and mucus, which serve as a primary line of defense. Probiotic strains cause alteration in host immunity by the elevation of AMPs and mucus, protecting epithelial cells from harmful bacteria, changing cytokines, and enhancing the immune system (Fusco et al., 2023). Probiotics have the capacity to decrease inflammation of the intestine by decreasing the TLR activation and blocking the NF-κB in intestinal absorptive cells (Joo et al.,2011).

Role of Probiotics and Prebiotics in Poultry

Probiotics at different concentrations affect poultry birds and give various benefits (Boodhoo et al., 2023). Lactic acid bacteria-prepared probiotics are used to cure illnesses caused by harmful bacteria (Mehmood et al., 2023). At a rate of 108 CFU/g, *Lactobacillus casei* enhanced the spread of intraepithelial lymphocytes via the chemokine signaling cascade and also influenced the mucosal immune system in chicks by upregulating the synthesis of cytokines (Tian et al., 2021). Moreover, Wang et al. (2023) investigated that 0.1 and 0.5% *Lactobacillus plantarum* raised average daily 11.88 and 9.78% weight of the yellow-feather broilers, at 1-3 weeks of age, respectively. While at the age of 3-6 weeks, *L. plantarum* at 0.1 and 0.5% concentration, increased 6.40 and 4.29% weight, respectively. IgA and IgG were also slightly increased at 1-3 weeks. Moreover, *L. plantarum* also causes hindrance in the multiplication of harmful bacteria by producing lactic acid because lactic acid decreases intestinal pH. Further, at the age of 1-21 days, *Clostridium perfringens* infected broilers were fed with *L. lactis* NZ9000 (pre-treatment) and concluded that *L. lactis* decreases the necrotic enteritis lesions. It may be because of the lessening inflammation mechanism, changes in the intestinal immune cells, and altering intestinal microbiota. In the same way, different probiotics have beneficial effects in the poultry industry (Boodhoo et al., 2023) as shown in Table 1.

Table 1: Role of probiotics in poultry

Role of probiotics	Action	References
Enhancement of gut microbiota	Compete with pathogens to establish the balance of beneficial gut flora. Decrease harmful bacteria like *Clostridium perfringens, Salmonella*, *Eimeria, and E. coli*	(Mohsin et al., 2022; Wang et al., 2023; Boodhoo et al., 2023; Mirsalami & Mirsalami 2024)
Enhance digestive efficacy	Assist in the decomposition of feed components, boosting nutrient absorption. Increase the synthesis of digestive enzymes.	(Jha et al., 2020)
Boost immunity	Increase local gut immunity by activating mucosal immune systems Boost systemic immunity via altering immune cells.	(Boodhoo et al., 2023; Wang et al., 2023)
Stress reduction	Aid poultry cope with stressors such as heat, disease, and transportation.	(Aydin & Hatipoglu, 2024)
Enhance growth performance	Improve body weight and feed conversion ratio (FCR)	(Wang et al., 2023)
Pathogen control	Generate antimicrobial substances such as bacteriocins and organic acids. Drop gut pH, produces an unfavorable environment for pathogens.	(Xiang et al., 2022; Wang et al., 2023)

Prebiotics exert their beneficial effects at different dose rates in poultry birds (Teague et al., 2023). Like, Ahmad et al. (2023) investigated that β-galacto-oligosaccharides (0.2, 0.2, and 0.5%) as a dietary supplement partially enhances meat quality by lowering pH, boosting antioxidant activity, and decreasing cooking loss. *Saccharomyces cerevisiae* derived β-glucans at 50 and 100mg/kg of feed against *Salmonella typhimurium* raised gut health by lessening destruction to the intestinal mucosal barrier. Moreover, yeast β-glucans are potential immunomodulators and can be used as an alternative treatment against *Salmonella typhimurium* (Anwar et al., 2023). Probiotics enhance pathogen antagonism by competitive exclusion in the gastrointestinal tract; thus, they selectively increase the development and proliferation of beneficial microbes (Solis-Cruz et al., 2019). One of the major causes for AGP replacement with prebiotics is the enhancement of birds' growth, sustainability, and non-toxic effects (Adhikari & Kim, 2017). At last, prebiotics exert beneficial effects on poultry as shown in Table 2.

Table 2: Role of prebiotics in poultry

Role of prebiotics	Action	References
Selective Growth of Beneficial Bacteria	Feed beneficial microbes like *Lactobacillus* and *Bifidobacterium*	(Richards et al., 2024)
Gut Health Improvement	Reduce colonization of harmful microbes by supporting a healthy gut microbiota Promote the growth of intestinal villi which increase nutrient absorption	(Teague et al., 2023)
Immune System Modulation	Increase immunological responses by encouraging beneficial microbes to produce immunomodulatory compounds	(Bełdowska et al., 2024)
Reduction of Harmful Bacteria	Bind to pathogens to stop them from binding to the gut lining (e.g., prebiotic binding to *Salmonella* and *E. coli*).	(Fadl et al., 2020)
Improved Nutrient Utilization	Boost feed efficiency by improving gut health and nutrient absorption	(Youssef et al., 2024)
Environmental Stress Resistance	Help poultry cope with challenges like diet changes, heat stress, and diseases.	(Richards et al., 2024)

Limitations of Prebiotics and Probiotics

While using probiotics, a proper dose is necessary because overdose causes more harmful effects than probiotic inadequacy. According to one study, high probiotic dose causes semen quality destruction in roosters flocks. Probiotics plus *Lactobacillus* species can produce infertility in a head of roosters due to the direct impact of *Lactobacillus* species on semen quality (Haines et al., 2015; Kiess et al., 2016). For the storage of probiotics, the best method of storage should be selected. Hydrator variety affects the bacterial fatty acids and protein formations. In one study, plastic bags along with different dryers are used for probiotic storage. From Silicon granules, NaOH, and LiOH, only NaOH showed the best shape maintenance results. To obtain proper efficiency of probiotics, effective storage along with the advancement of packaging technology is very important to maintain the best product characteristics (Dianawati et al., 2013). According to some studies, probiotics showed no effect on chickens (Hidayat et al., 2016). Like, in one research, the weight of the primary parts of the carcass (heart, spleen, neck, thighs, liver, breast, and back) is not increased. Moreover, changes in pH are also not seen (Hidayat et al., 2016). Behrouz et al. (2012) said many risk factors are involved in the examination of probiotic efficacy, and their real impact on poultry birds' body and their production. It highlights the dependence of efficacy on the dose and variety of microorganisms and the parameters of administration of these dietary supplements.

Administration of probiotics appears a reasonable procedure because of fewer side effects in its administration. Despite great potential and combination, it becomes difficult to create ideal strains under optimal circumstances. The effectiveness of the delivered dosage is influenced by a variety of interspecies combinations and tactics (Hofacre et al., 1998; Nisbet, 2002). Probiotic effectiveness alters with the changes in different factors like dosage, day, condition, kind of gut microorganism, and probiotic strain (Boodhoo et al., 2023; Wang et al., 2023). Feed thickness and structure also influence probiotic exposure (Mikulski et al., 2020). The number of animals impacts the efficacy of the probiotic bacteria particularly, a large number of animals due to difficulty in the maintenance of hygienic conditions (Blajman et al., 2014). For the formation and efficacy evaluation of probiotics, it is necessary to examine the relation between gut microflora and the kind of probiotic microorganism in the dietary supplement (Wang et al., 2023). Genetic variation which is linked to the biological processes of bacteria with particular microbial operations in GIT, is necessary. Industrial strains can be preserved and particular relationships between bacterial populations may be identified by monitoring them (Bortoluzzi et al., 2019).

No doubt, probiotics showed good efficiency in many studies, but the type of microorganism to be applied must be identified. Therefore, the risk assessment must be conducted for different probiotic strains. Although major types of probiotics are safe, but they can have negative characteristics like virulence parameters, transferable antimicrobial resistance, and toxin production ability (Donohue, 2006; Lee et al., 2017). There are some guidelines that can be used for the assessment of probiotic security, identification of strain, evaluation of tests to check the potential of probiotic types, identification of risk factors, in vivo tests for the evaluation of target host health (FAO/WHO Joint report, 2002; Kim et al., 2018).

Galactooligosaccharides and sodium butyrate, *in ovo*, have a negative effect on broiler production and do not affect carcass composition (Dunisławska et al., 2024). Galactooligosaccharides do not mitigate the harmful effect of heat stress on chicken' performance (Tavaniello et al., 2022). Mannanoligosaccharide is more costly than other groups. Moreover, it does not improve performance, lipid composition, and gut bacterial ecology in broilers (Kamran et al., 2021). Dietary supplementation of Mannanoligosaccharides has no effect on feed intake in layers (Salami et al., 2022). Prebiotics have been used as alternative growth promoters but, in one study mannanoligosaccharide has no difference in results than the other antibiotic growth promoter groups (zinc bacitracin, furazolidone, enramycin). Therefore, to obtain the best results, researches should be conducted to evaluate the exact dosage and mode of action of prebiotics in poultry nutrition (El-Shall et al., 2019; Kamran et al., 2021; Abd El-Aziz et al., 2022).

Conclusion

The use of probiotics and prebiotics gives multiple health, nutritional, and production benefits. They are mostly used as alternatives to AGPs and other chemical compounds in poultry. These are eco-friendly, economical, and decrease disease treatment expenditure. Marketing of probiotics has increased by almost 44.2 billion US$ recently, all over the world. It is expected that it will increase by 74.3 billion US$ by 2025 with a 7.7% growth rate. But prebiotics and probiotics have some limitations e.g., infertility, no effect on birds' growth, and toxicity. In large numbers of animals, hygienic conditions cannot be maintained which changes their efficacy. Therefore, there is need to conduct more researches on probiotics and prebiotics to examine the exact dose rate, efficacy, mode of action, effect on the host, and kind and toxicity of microorganisms.

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