

Review on application of probiotics in poultry farm

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Summary

Poultry farming, raising of birds domestically or commercially, primarily for meat and eggs but also for feathers. In poultry industry, probiotics applications have widely been shown to improve the barrier function of intestine and reduce pathogenic problems in gastrointestinal tract thus leading to the enhancement of immune response and replacement of sub-therapeutic antibiotics. Probiotics, the novel feed or food supplements are naturally occurring beneficial live microbes including bacteria, fungi and yeasts. Probiotics are proven to improve the health and wellbeing of birds. As a group of growth promoters, the addition of probiotics to the diet of poultry has been found to improve growth performance, feed conversion efficiency, immune responses and help in combating enteric pathogens. The addition of probiotics to poultry diet also improves egg production in layers. Three basic mechanisms of probiotics include competitive exclusion of pathogenic microbes, production of antibacterial substances and immune modulation of host. Generally, live a pathogenic microbial strain, singly or as multi-strain probiotics, belonging to genus *Lactobacillus*, *Streptococcus*, *Bacillus*, *Enterococcus*, *Pediococcus*, *Aspergillus* and *Saccharomyces* are used in poultry. The injudicious and prolific use of sub-therapeutic levels of antibiotics in animal feed to suppress or eliminate harmful organisms, and to improve growth and feed efficiency, have paved way for their presence as residues in animal by-products and have also given rise to drug resistant microorganisms. Because of public health concerns, nowadays, the use of probiotics as substitute for antibiotics in poultry production has become an area of great interest. The present article reviews the beneficial effects and potential applications of probiotics or direct fed microbial (DFM) on health and production performances in poultry.

Keywords: Immunity Intestine, Microorganisms and Poultry Probiotics

1. Introduction

Poultry are domesticated birds kept by humans for their eggs, their meat or their feathers. These birds are most typically members of the superorder Galloanserae, especially the order Galliformes. Poultry is usually taken by the rural farmer to mean chickens because they are reared more often than any other kind of poultry [01].

The world has over 23 billion poultry birds-about three per person on the planet, and about 5 times more than 50 years ago. They are kept and raised in a wide range of production systems, and provide mainly meat, eggs and manure for crop fertilization [02].

Feed is the largest cost in poultry production; small improvements in feed use efficiency have a significant economic impact. The improvement in performance and productivity of poultry due to the use of probiotics in feed has been attributed to increased feed intake and improved feed efficiency [03]. The steady increase in the cost of poultry feed ingredients and compounded feed is making less profit to poultry farmers. To lessen the cost of

feeding, several feed additives like synthetic hormones and antibiotics have been extensively used to promote poultry production in recent years. From the varying additives, like antibiotics have been widely used in livestock diets due to their therapeutic effects [04].

The poultry industry uses antibiotic feed additives and synthetic hormones extensively for disease prevention, growth and enhancing poultry production, but due to development of antibiotic resistant bacterial strains and residual effects of these feed additives in eggs and meat, they lead to various health risks to consumers. Therefore, the use of probiotics feed additives for better and intact production in livestock and poultry is compulsory [05, 06]. Probiotics are a dietary supplement that increase the population of the micro flora, which are needed in the intestinal tract in order to process food properly, strengthen the immune system and help chickens to digest their food more efficiently and helping remain healthy and gain weight faster in chickens [07]. The primary purpose of the gastrointestinal tract (GIT) is to digest and engage in absorption of nutrients in order to meet metabolic demands for maintenance, normal growth and

development, but it also acts as a vital barrier preventing the entry of several potentially harmful pathogens from the external environment. It is well known that a well-balanced gut micro flora is crucial for animal health and performance [08].

To maintain the balance of intestinal micro flora in animals and chicken, it is important to prevent diseases by controlling the overgrowth of pathogenic bacteria by supplementation of fermented foods containing probiotic for improved health. Probiotics are used to improve the health of birds and subsequently result in better production [09]. The major advantage of probiotics is that different from antibiotics, they leave no residues in meat [10]. The use of probiotics has been primarily to establish normal intestinal flora with broad target of prevention or minimizing the disturbances caused by enteric pathogens [11-13]. Feeding of probiotics have been reported to have beneficial impacts on the commercial animals by enhancing weight gain, increasing feed conversion efficiency, increasing egg production, lowering the incidence of disease as well as lowering mortality rates. These living organisms may be bacteria, fungi or yeasts. They are isolated from the gut of a healthy adult animal and poultry typically of the same species to which the probiotics will be given [14]. Most commonly used probiotics bacteria in poultry are *Lactobacillus*, *Bifidobacterium*, *Leuconostococcus*, *Enterococcus*, *Lactococcus*, *Bacillus*, *Saccharomyces*, *Aspergillus* and *Pedi coccus* species [15]. In Ethiopia, the knowledge and application of probiotics in poultry production is yet limited. Therefore, the main

Objective: of this seminar paper is to provide general information about the major application of probiotics in poultry production.

2. Literature Review

2.1. Overview of probiotics

2.1.1. Definition of probiotics: The term probiotic is derived from Latin (pro) and Greek (bios) meaning literally for life [16]. Probiotics are viable bacterial or fungal cultures which are able to enhance the balance of intestinal flora and exercise valuable effects on the individual in which it has been administered. Probiotics are group of dietary products that can be incorporated in animal and poultry rations to raise performance or reduce pathogenic bacteria. It is a beneficial microorganism or combination of such microorganisms, which United Nations (FAO would quickly establish in the gut to suppress colonization and growth of harmful bacteria [17]. The joint Food and Agriculture Organization of the) and World Health Organization (WHO) Working Group defined probiotics as live micro-organisms which when administered in adequate amounts confer a health benefit on the host [18]. This definition is widely accepted and adopted by the International Scientific Association for Probiotics [19].

2.1.1. History of probiotics: The characteristics, actions, effects and importance of probiotics have merited a renewed popularity in contemporary culture, but probiotics are ancient history. Thousands of years ago, a Roman naturalist named Pliny the elder recommended drinking of fermented milk to treat intestinal problems. Fermented foods are also mentioned in the Bible and the sacred books of Hinduism. Climates in the Middle East and Asia favored the souring of milk products, which

were recommended for intestinal illness. This represented the therapeutic use of probiotics, even before the bacteria contained within them were recognized. Many of the same soured milk products are still being consumed today in 1906 Elie Metchnikoff, Russian scientist, at Institute Pasteur in Paris suggested concept of probiotic in *Bacillus* bacteria. In 1907 he first observed the beneficial role of certain bacteria and suggested that it would be possible to modify the gut flora and replace harmful microbes with useful microbes and in 1908 he has received the Nobel Prize in medicine for his work demonstrating that harmful microbes can be replaced by beneficial microbes to treat intestinal illnesses [20]. Metchnikoff is generally considered to be the “father of probiotics [21].

2.1.2. Types of probiotics: Different kinds of probiotics have been used so far in the diets or otherwise of poultry for stimulating production and/or feed utilization efficiency. Examples include: RE-3, RE-3 Plus, P3, Prima Lac, Maz, Poultry Star, Lacto-Sacc etc. The difference in these probiotics comes with the strain of bacteria that was used, dosage, mode of application, time of application etc. Probiotic products may contain different genera, different species, or even different strains of the same species, and not all products should be expected to work the same. Therefore, claims of efficacy should be target specific and should be made only for products that have been found efficacious in carefully designed studies [22]. Probiotics can also be categorized in to probiotic drugs, probiotic foods and direct feed microbial. They are either single or multiple live microbial cultures which promote health benefit to the hosts. They are nonpathogenic and nontoxic in nature, when administered through the digestive route, are favorable to the hosts health [23].

2.1.3. Timing and route: of administrations There are various different methods for administering probiotic preparations to the host animal. It can be given as a powder, capsule, liquid suspension and spray. The amount and interval between doses may vary. Probiotics may be given only once or periodically at daily or weekly intervals [24]. The way of administration and timing are main factors affecting the effectiveness of probiotic supplementation administration via the feed, compared to administration in the drinking water, result in a higher increase of average daily gain; besides the supplementation of probiotics during early life is of great importance to the host because harmful bacteria can modulate expression of genes in intestinal epithelial cells, so this can create a favorable habitat.

2.1.4. Features of ideal probiotics and their mechanism of action: Probiotic bacteria contain a minimum of 30 x 10⁹ Colony Forming Units. They have high survival ability and multiply fast in the conditions within the poultry gut. In addition to being non-pathogenic to animals, micro-organisms used as probiotics are selected on the basis of their survival in the gastro-intestinal environment and ability to withstand low pH and high concentrations of bile acids. In addition, the chosen strain should tolerate the manufacturing, transportation, storage and application processes, maintaining its viability and desirable characteristics [25]. The capacity of potential probiotic micro-organisms to with stand the gastro-intestinal environment can be tested in vitro by challenging with low pH [26]. Another desirable characteristic is the ability to adhere to the intestinal

epithelium, enabling the probiotic strain(s) to colonize the intestine. In addition, ability to grow rapidly on inexpensive media is a requisite for economically viable production [27].

Probiotics are free of intermingle antibiotic resistant genes, sufficiently stable while manufacturing, processing and storage and do not undergo recombination with pathogenic strains. These live microorganisms can preferably be compatible with feed additives and have good sensory properties. Probiotics contain gram positive bacteria and must have a short generation time. They must attach to intestinal epithelium, have the ability to rapidly colonize the intestine edge of pathogenic microbes and constantly present gut micro flora [28, 29].

Probiotic supplementations are used to preserve a healthy microbial balance within the intestine to promote gut integrity and prevent enteric disease. This is accomplished through: establishing and maintaining healthy gut micro flora, improving digestion and utilization of nutrients, competitive exclusion of harmful bacteria/pathogens, decreases pH, neutralization of toxins, competition for nutrients with pathogens, reduction in ammonia production and stimulation of the immune system [11-13, 16, 28-30].

Enhancement of the epithelial barrier, increased adhesion to intestinal mucosa, production of antimicrobial substances and modulation of immune system are other mechanisms of action by. A front line of defense against the adverse effect of pathogens is provided by probiotics showing its antimicrobial effect. For example, lactic acid producing probiotics show antimicrobial effects by reducing the pH of the gut [31, 32].

2.2. Role of probiotics in poultry production: The bird intestine is shorter, and the gastric juice has lower pH as compared to that to the mammalian Gastro intestinal tract. Populations of microorganisms colonizing the chicken GIT should reveal higher acid tolerance in shorter periods of time [33]. Probiotic bacteria are either anaerobic or facultative anaerobic. A large number of the applied microorganisms are deactivated during the passage through gastrointestinal tract due to high acidity and bile salt effect. Thus, large numbers of cells are required for each dose [34].

The crop, proventriculus and gizzard have very few anaerobic bacteria due to the presence of oxygen and hydrochloric acid [35]. The small intestine contains a large number of facultative anaerobes such as *Lactobacillus*, *Streptococci* and anaerobes like *Bacteroides* and *Bifidobacterium specius*. The most heavily colonized regions of the Gastro Intestinal tract are the colon and cecum with colonization of 10¹⁰ to 10¹³ CFU/ml [36]. Probiotics colonize three different parts within the GIT; enterocyte, caecal and colonic epithelium; the most colonized region is colon and caecum [37].

The diversity/composition of the microbial flora of poultry gut depends on several factors including species, breed, geographic location, diet composition, age of the chicken and the specific section of the Gastrointestinal tract such as small intestine, ileum, caeca [38]. The effect of probiotic depends on physiological state of the bird type, concentration of probiotic strain, persistence

of intestine, ability to survive during feed processing of Gastro Intestinal Tract and compatibility with natural microbial of the intestine [39].

2.3. Production performance and health benefit of probiotics:

The importance of probiotic supplementation in poultry production are: enhancing gut health by increasing a desired equilibrium in its microbial population in digestive enzymes and nutrient absorption, inhibits growth of pathogens, reduces mortality and improves growth rate [40]. Probiotics maintain a proper balance of useful bacterial population in the intestine of animals and birds, which is important for their efficient feed conversion, growth and productivity [41]. The intestinal tracts of newly hatched chickens have no harmful microorganism. Through feeding, microbes slowly colonize the Gastrointestinal forming a stable microbial association [42]. As harmful microbes are established they could cause localized microbial infections, intestinal putrefaction and toxic production [43].

Microbial infections occur in intestine of chicken resulting in weight loss, death and poor meat quality. Effective probiotics can reduce harmful microbes from the GIT of chickens [44]. Colonization of probiotic *Lactobacillus* strains has been demonstrated to have a preventive function against *Salmonella enterica* serovar Enteritis infection in chicken. Proposed mechanisms of pathogen inhibition by the probiotic microorganisms include competition for nutrients, production of antimicrobial conditions and compounds (volatile fatty acids, low pH and bacteriocins), competition for binding sites on the intestinal epithelium and stimulation of the immune system [45].

Antimicrobial activity of probiotics against Gram-positive and Gram-negative bacteria varies, and this has to do with production of bacteriocins. Some inhibit taxonomically related Gram-positive bacteria, whilst others are active against a much wider range of Gram-positive and Gram-negative bacteria as well as yeasts and molds [46]. Probiotics increased egg production and egg quality and decrease egg contaminations [47, 48]. Probiotic was also increased egg shell weight, shell thickness and serum calcium [49]. A combined mixed culture of *Lactobacillus acidophilus*, *Lactobacillus casei*, *Bifidobacterium thermophiles* and *Enterococcus faecium* enhanced egg size and lowered feed cost in laying hens [50]. *Bifidobacterium thermophiles* and *Enterococcus faecium* improved egg production and quality [47]. Heat-stressed laying hens, administration of multi strain or probiotic *Bacillus licheniformis* increased egg production [51, 50].

The highest production and egg weight in layers can be achieved with Liquid Probiotics Mixed Culture (LPMC) containing two types of microorganisms: *Lactobacillus* and *Bacillus* species. Probiotics supplementation improve meat quality of broilers and intramuscular lipid content involved in determining meat quality particularly nutrition, tenderness, odor, tastes and flavor characteristics [52]. Improvements in performance and carcass characteristics with the use of probiotics may occur due to increase in consumption and digestibility of the diet [04]. The probiotics could increase digestive enzymes activity and promote beneficial intestinal micro biota, improving intestinal health [53]. Many researchers stated that there is a greater tendency

to higher ratio of unsaturated fatty acids to saturated fatty acids in pectoral and thigh meat of broilers fed with probiotics diet containing *Bacillus*, *Lactobacillus*, *Streptococcus*, *Clostridium* and *Saccharomyces*. Their results concluded that the fat in meat was converted into favorable fat in the presence of probiotics, which contributed to smoother meat texture in chicken [54]. Probiotics like *Clostridium butyricum* added in diet of broiler have sensory attributes of the meat appearance, texture, juiciness and acceptability [55].

Probiotics may potentially stimulate growth through increased Short chain fatty acid production in poultry and through selective regulation of insulin signaling in different tissues. Short chain fatty acids like acetate, propionate and butyrate are used as energy source in tissues. Particularly in chickens, butyrate has shown beneficial effects by selectively partitioning the nutrients away from liver and adipose tissues towards muscles through up regulation of insulin receptors [56]. Short chain fatty acid production due to probiotics helps to promote intestinal health and integrity by directly stimulating epithelial cell proliferation, acts as the epigenetic regulators of the gene expression of multiple genes that help in growth and overall health of poultry [57]. Another mechanism by which probiotics may stimulate growth is by regulating the immune system. When immune system is regulated, it suppresses the negative effects of chronic immune activation. When immune system is activated, there is diversion of nutrients from production process towards immune response [58]. On the other hand, there is direct effect on epithelial barrier thus producing better growth [59].

2.4. Probiotic role on control of the diseases and immune response: Probiotic supplementation in feed is considered to be the potential for controlling necrotic enteritis and *Eimeria acervulina* and *Eimeria tenella* infection with effective reduction of oocyte [42]. With the concept of competitive exclusion, probiotics may have effects against common pathogens and the origin of food-borne illnesses such as *Salmonella* species, pathogenic strains of *Escherichia coli*, *Campylobacter jejuni*, *Clostridium perfringens*, and *Listeria monocytogenes* [60]. Probiotic administration has been found to enhance antibody responses as well as leukocytes count in birds reared under hot temperature [50, 61, 62].

Probiotics act in the lower or upper intestinal tract in several mechanisms to improve productivity; including competition with pathogens to exclude them from luminal cell adhesion or to limit their nutrients in the environment, stimulation of immunity, production of antimicrobial substances for antimicrobial activity, increasing the barrier integrity, and regulation of cytokine production to reduce inflammation.

The immune modifying effects in poultry happens as the live microbial move along the wall of the intestine and multiply to a limited extension and the dead organisms are absorbed and stimulate the immune system [63]. Continuous use of probiotics has a noticeable effect on the immune system. Stimulation of humoral and cell mediated immunity through enhanced production of natural interferon/cytokines, increased macrophage, lymphocyte, natural killer cell activity, up regulated oxidative burst in heterophils and immunoglobulin (IgG, IgM

and IgA) [10-12].

Production of cytokines leads to the overall immune modulation in the chicken. LAB has shown the modulating effects on the immune system of both layer- and meat-type chickens. The ability of *Lactobacillus* to modulate chicken cytokines, toll-like receptors and chemokine gene expression has been demonstrated [64]. Increase in the antibody secretion due to increase in B-lymphocytes (humoral immunity) is a potential mechanism by *Lactobacillus* in boosting the immunity in broiler chicks [65]. The increase in the population of white blood cells may be attributed to the presence of LAB in the diet stimulating the production of lymphocytes, particularly the B cells that are responsible for forming antibodies that provide humeral immunity. Enhancement of gut barrier function through modulation of the cytoskeleton and epithelial tight junctions in the intestinal mucosa is one of the mechanisms of probiotics in preventing pathogens [16].

Probiotics favorably alter the balance of intestinal micro flora by inhibiting the growth of harmful bacteria, promoting good digestion, and increasing resistance to infection. Probiotics promote a balance of intestinal flora that produces organic compounds such as lactic acid, hydrogen peroxidase, and acetic acid. These products increase the acidity of the intestine, which inhibits the reproduction of harmful bacteria. Probiotic bacteria also produce bacteriocins which are natural antibiotics that kill undesirable microorganisms [66]. Increased Supplementation of probiotics in layers increases cellularity of Payer's patches in the ileum; it is an indication of stimulation of the mucosal immune system, which secretes IgA in response to antigenic stimuli. Probiotic treated birds have significantly more serum antibody IgM than birds that are not treated with probiotics. Probiotics have been reported to be useful to improve the immune system of birds subjected to heat stress [67].

Probiotics produce a gut stabilizing effect and immune regulation, particularly through balanced control of pro-inflammatory and anti-inflammatory cytokines. The use of probiotics increases the amount of IgA found in the lumen, the numbers of IgA, IgM and IgG producing cells as well as the number of T cells in the cecal tonsils [68]. Oral administration of probiotics increases natural antibodies against different antigens in both the gut and the serum.

Probiotics can enhance the resistance of birds and partially protect against the negative growth effects associated with pathogenic microbes and also considerably lowers the pathogenic microbial load in the intestine, lowering the risk of their spreading in the housing through fecal contamination [27, 28]. Feeding probiotics could improve antibody titers against viral diseases like Newcastle disease and Infectious bursal disease. By increasing the immune status, it is possible to prevent enteric infections and help reduce the losses due to secondary infections in birds observed commonly during viral diseases or immunosuppressive conditions. Further, probiotic stimulation of the immune system exhibited higher production of immunoglobulin, stimulation of macrophages and lymphocytes activity and also by augmentation of the production of γ -interferon [69, 70].

2.5. Influence of probiotics on intestinal development: The early post-hatch period is a critical stage for poultry growth and health as the new hatchling switches its nutrient source from the yolk to carbohydrate- and protein-based diet [71]. In order to accommodate the rapid transition of nutrient source, the digestive organs of newly hatched poulters undergo both anatomical and physiological changes and are the most rapidly developing organs during the early post-hatch period. The rapidly developed intestinal tract provides an ideal niche for microbial colonization. In the meantime, gut microbiome also plays an important role in intestinal development [72].

Numerous studies have been carried out to investigate the effects of probiotic administration on the histomorphology of the intestine. Supplementation of dietary treatment with probiotic *Lactobacillus* species, influence the villi height and crypt depth in the small intestine of broiler chickens. Probiotics can enhance gut development and length of intestinal villi and decrease the depth of crypts in poultry. Increased cellular turnover requires a substantial amount of energy utilized towards growth. Longer villi and shallower crypts are related to decreased cell replacement and longer enterocyte lifespan [73]. The intestinal crypt in enterocytes undergo continuous proliferation in order to replace cells lost at the villus tip due to normal sloughing or inflammation from pathogens [74].

Intestinal morphology including duodenal and ileal villus height and crypt depth as well as villus height to crypt depth ratio are indicative of gut health in broilers. Increased villus height and villus height to crypt depth ratio are directly correlated with an increased epithelial turnover and longer villi are correlated with activation of cell mitosis [75, 76]. Whereas, shortening of villi and deeper crypts lead to poor nutrient absorption, increased secretion in gastrointestinal tract and reduced performance [77].

Probiotics may also reverse the impaired villus-crypt structure of heat-stressed birds by controlling the corticosterone level and the excessive release of proinflammatory agents that can cause intestinal tissue injuries and increase the permeability of the intestine [51, 78,79].

2.6. The effect of probiotic on feed intake and feed efficiency:

As feed is the largest cost in poultry production, small improvements in feed use efficiency have a significant economic impact. The improvement in performance and productivity of poultry due to the use of probiotics in feed has been attributed to increased feed intake and improved feed efficiency [04]. The apparent ileal digestibility (AID) of essential amino acids was improved in birds fed a maize-soybean-based diet supplemented with a low dose of a multi-strain commercial probiotic (Probiom) containing *Lactobacillus acidophilus*, *Bacillus subtilis* and *Collibacillus butyricum* [55].

Probiotics help in metabolism of minerals and synthesis of vitamins (Biotin, Vitamins-B1, B2, B12 and K), which are essential for proper growth and metabolism in chicken [11-13]. In addition, by directly protecting epithelial barriers, probiotics can enhance nutrient absorption, which also may result in enhanced growth [60]. Very importantly, there is evidence that probiotics may stimulate digestive enzyme activities, which may

increase nutrient availability in the gastrointestinal tract [81].

The use of Enterococci as probiotics in chickens prolonged feeding with *Eimeria faecium* improved egg laying intensity and feed conversion efficiency [48]. Rise in feed and water consumption is recorded in laying hens fed with LPMC containing two types of microorganisms: *Lactobacillus* and *Bacilli* species [53]. The investigators found that inclusion of probiotic (*Lactobacilli* and *Bacillus subtilis*) in diet enhance feed efficiency and growth performance in broilers. Broilers probiotic supplemented diet had better weight gain and feed efficiency when compared to the broilers feed without probiotic supplement [82-88].

3. Conclusion and Recommendation

Feed is the major constraint of the total cost for meat and egg production. It accounts for 70% of total cost poultry production. Feed composition is crucial to promote growth and maintain health of poultry. A well-balanced diet sufficient in nutrient and energy is also of significant importance to maintain gut in healthy state. In view of this, the concept of probiotics as feed additives has garnered much attention and support. Probiotics are live microorganisms, when administered in sufficient amounts; confer with a health benefit on the host. Use of probiotics is better for quality production of egg and meat than any other growth promoters. Significant work and studies have increasingly demonstrated that probiotics provide means to a balanced gut microbiota in poultry, maintaining health status in broilers, preserving gut condition and improving immune system as well as enhancing nutrient absorption, which are all crucial and needed to promote growth of broilers. Adding probiotics to the feed, the host receives a boost to establish a proper microbial population in its gut which is responsible for developing natural resistance in poultry to fight against various pathogens. With multiple utilities, these desirable microbes hold much promise to function as an effective agent to maintain growth, production and health in poultry operations. In Ethiopia, the usage of probiotics in poultry feed supplementation is very limited.

Based on the above conclusive remarks, the following points are forwarded as recommendation.

In Ethiopia, researches should focus on the advantage of supplementing poultry with probiotics.

Awareness creation to people and farmers on the usage of probiotics in poultry production should be exercised. To improve the poultry production and productivity, the government and private investors should participate in the establishment of probiotic feed processing.

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