

## Review Article

## Beneficial Implementation of Probiotics in Farm Animals and Poultry Husbandry

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### Abstract

Probiotics have become highly realized as additives for livestock because of their beneficial impacts on health and well-being. The current article aims to provide knowledge about the potential probiotic microbial strains referred to their relation to animal health. Lactic acid bacteria are the most regularly used microorganisms as probiotics, "besides other microbiota including certain fungi" which can be obtained from human, animal, plant, and environment. Not all microbiota be a good probiotic but there are main selection criteria for any potential probiotic microorganism. Nowadays, probiotics are used on large scale as growth promoters and productivity enhancers in both poultry and farm animals. Finally the promising probiotic must achieve the safety status for humans or animals and lack of antibiotic resistance genes transfer.

**Keywords:** Probiotics- Mechanism of Probiotic- Probiotics selection criteria

### Introduction

Probiotics are nonpathogenic beneficial microorganisms. They have microbiological, and metabolic effects. They are very important for the health of man and animal. Lactic acid bacteria and *bifidobacteria* strains are mostly used as probiotics. In clinical and veterinary practices, they consider as prophylaxes and are used for therapeutic purposes. They are safe, good chance of replacing antibiotics in animal husbandry, and viable alternative to antibiotics for increasing performance in livestock, besides nutrient digestibility [1]. It was reported that about 4.4 and 4.8 billion dollars in 2020 and 2021 respectively profits from probiotic global market, expected to reach 7.3 billion dollars in 2026 at a CAGR 8.8% in the animal feed market; farm and pets animals [2]. Therefore, probiotics are very important not only for health and the disease in man and animal but also, for economic purposes. Traditional applications of probiotics have been mostly presented through the fermentation process usually directed to digestive processes. More than 1000 years ago, old Carthusian silos and ancient evidence from Egyptian drawings show that farmers used silage for their animals during winter times as it was an excellent way to preserve summer crops [3].

#### 1. What is a Probiotic?

The term probiotic is meaning 'for life'. It is a composite of the Latin preposition *pro* ("for") and the Greek adjective (biotic) from the noun *bios* ("life") [4] although they have sundry different meanings over the years. Probiotics are nowadays defined as "Live microorganisms that when administered in adequate amounts confer a health benefit on the host" [5].

#### 2. Classification of Probiotics:

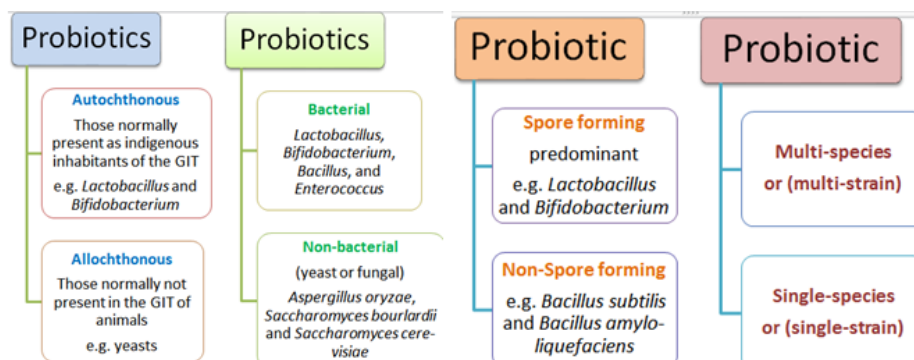


Figure (1): classification pattern of probiotics; [1, 6-8].

### 2.1. Lactic acid bacteria (LAB):

- Genus *Lactobacillus*: *Lactobacillus acidophilus*, *L. brevis*, *L. bulgaricus*, *L. casei*, *L. cellobiosus*, *L. curvatus*, *L. delbrueckii* spp. *bulgaris*, *L. fermentum*, *L. gallinarum*, *L. helveticus*, *L. johnsonii*, *L. lactis*, *L. paracasei*, *L. plantarum*, *L. reuteri*, *L. rhamnosus*;
- Genus: *Streptococcus* spp.: *Streptococcus salivaris*, *Streptococcus thermophiles*;
- Genus: *Lactococcus*: *L. lactis cremoris*;
- Genus: *Leuconostoc*: *Lc. mesenteroides*
- Genus: *Pediococcus*: *P. pentosaceus*, *P. acidilactici*.

### 2.2. Bifidobacteria:

- Genus: *Bifidobacterium*: *B. adolescentis*, *B. animalis*, *B. bifidum*, *B. breve*, *B. essensis*, *B. infantis*, *B. laterosporum*, *B. thermophilum*, *B. longum*.

### 2.3. Propionibacteria:

- Genus: *Propionibacterium*: *P. acidipropionici*, *P. jensenii*, *P. freudenreichii*, *P. thoenii*.

### 2.4. Enterobacteria:

- Genus: *Enterococcus*: *E. faecalis*, *E. faecium*.

### 2.5. Sporulated bacteria:

- Genus: *Bacillus*: *B. alcolophilus*, *B. cereus*, *B. clausii*, *B. coagulans*, *B. subtilis*.

### 2.6. Other bacteria:

- Genus: *Escherichia*: *E. coli*;
- Genus: *Sporolactobacillus*: *S. inulinus*.

### 2.7. Yeasts: Genus: *Saccharomyces*: *S. cerevisiae* (*boulardii*).

## 3. How to select a good probiotic?

The good probiotic has certain features; viable cells, non-toxic, non-pathogenic, a beneficial effect on the host animal, surviving and metabolizing in the gut environment condition at low pH and presence of bile, adherence to intestinal cells lines, stabilization of the intestinal microflora, production of certain preparations (i.e., bacteriocins), colonization and multiplication faster, generic specificity of probiotics and viable for long periods under certain conditions [9].

## 4. Efficacy of Probiotics on farm animals:

### 4.1. Polygastric Animals

The most widely distributed animals of mammals are ruminants, which moreover about 150 wild and domestic species, while cattle, sheep, goats, and water buffaloes are very important due to economic interest [10]. There are microorganisms in the rumen microbiome which composed mainly of bacterial species moreover, flagellated and ciliated protozoa, fungi, methanogenic archaea and bacteriophages [11] and present in the rumen with the level of  $10^{10}$  (bacteria),  $10^8$  (protozoa),  $10^7$  (archaea), and  $10^3$  (fungal spores) CFU / ml in rumen fluid [12].

The fermentation process, in the ruminants and farm animals, is essential for health and productivity. It is depended on the rumen microorganisms which are killed or decreased due to the use of the systematic antibiotics “used as growth promoters in animal husbandry”. Not only had the use of

antibiotics affected the rumen metabolism but also, releasing multidrug-resistant microorganisms and antibiotic residues in animal products [13].

Over the world; in the European Union, probiotics are widely used in animal husbandry and livestock production. Also, China, USA, and Australia use probiotics instead of antibiotics [14-19]. Digestive disorders and GIT pathogens can be treated via using probiotics [20], and control of pH in the rumen [21], enhanced animal performance, digestibility of fiber, and increased efficiency of feed conversion [22], production of huge milk yield and improved composition of milk and colostrum [23-25], treatment of mastitis [26, 27] and mitigation of methane [28, 29] stimulation of the immune system [30].

The pre-ruminant life of calves, lambs, and kids goat is critical life as they are more susceptible to enteric diseases so the administration of probiotics orally, directly, or in the feed is very essential to save them [31, 32]. *Lactobacillus*, *Bifidobacterium*, *Streptococcus*, *Enterococcus* species, *Escherichia coli*, *Bacillus*, fibrolytic *Prevotella* species and lactic acid bacteria, (*Propionibacterium* and *Megasphaera elsdenii*) had been reported to use them as probiotics [33, 34]. In buffaloes and reindeers, cellulolytic *Ruminococcus* species are used [35, 36] *Saccharomyces cerevisiae* and *Aspergillus oryzae* are used as yeasts and fungi probiotics [37].

Table (1): some applications of probiotics in ruminants.

Effects	Probiotics	References
Control of disease of Feedlot cattle, beef steer, Lithuanian Black-and-White, calve	<i>Propionibacterium spp.</i> , <i>Enterococcus faecium</i> , <i>Lactobacillus acidophilus</i>	[38]
Inhibition of <i>Staphylococcus aureus</i> invasion in bovine mammary epithelial cells	<i>Lactobacillus casei</i> CIRM-BIA 667, <i>Lactobacillus casei</i> BL23, and <i>Lactobacillus casei</i> CIRM-BIA 1542	[39]
Nutrition digestion of Calves(Holstein–Friesians, Friesian–Jersey)	<i>Bacillus amyloliquefaciens</i>	[40]
Yield improvement of newly born ruminants	<i>Bacillus licheniformis</i> , <i>Bacillus subtilis</i> , <i>Prevotella bryantii</i>	[41]
Increase in growth performance, improving weight gain, feed conversion efficiency in weaning calves	<i>Bifidobacterium</i> , <i>Lactobacillus</i> , and <i>Enterococcus</i>	[42]

In Egypt, some trials were performed to explore the beneficial effects of probiotics additives on ruminant performance, productivity and health; Hassan et al., (2020) [43] investigated the mixing of two different forms (powder or liquid) of *Ruminococcus flavefaciens* with concentrate feed that presented to group of Barki lambs for 75 days. They mentioned that the addition of examined probiotics resulted in a significant increase in nearly all of the dry matter digestibility coefficients, average daily gain and nutrient digestibility compared to the control group. Another study probed the influence of *Saccharomyces cerevisiae* dietary yeast supplement fed by examined primiparous and multiparous Egyptian buffaloes in early to mid-lactation. The study's results revealed increase in milk production in early lactation and a more persistent milk production during mid-lactation. Feed conversion and energy and nitrogen conversion efficiency increased with the use of yeast supplementation [44].

#### 4.2. Monogastric Animals

Pigs, poultry and horses are monogastric which have simple stomach. The bacteria, archaea, methanomicrobia, and thermococci have been identified as microbiota of pigs [45, 46]. There are 13 phyla of bacteria were estimated in the poultry GIT with firmicutes, bacteroidetes, and proteobacteria being the more 900 in chicken and in 500 turkey. *Lactobacillus*, *Ruminococcus*, *Bacteroides*, *Clostridium*, methanogenic archaea and fungi are most probiotics in chicken and turkeys [47, 48]. The GIT of horses is improved by the presence of beneficial bacteria, protozoa, fungi, and archaea [49, 50].

In monogastric animals, the most probiotics are used *Lactobacillus*, *Enterococcus*, *Pediococcus*, *Bacillus* (bacteria) and *Saccharomyces boulardii* and *S. cerevisiae* (yeasts) that targeting the cecum and the colon. The probiotics in monogastric animals are beneficial in increasing body weight, reducing diarrhea risk, improving feed efficiency, and digestibility of diet [51]. In swine, there was increased diet intake and weight performance in the period of pregnancy and lactation [52, 53]. Probiotics have been provided supportive care to piglets during their suckling and weaning life, Kim *et al.* (2014b) [54] illustrated that probiotics are more effective than antibiotics on the growth performance of pigs. Reduction of *Salmonella* infection [55], and protect against other enteric pathogens in post weaning piglets [56].

On the other hand, the concentration of ammonia in the excreta of poultry can be reduced by using probiotics like *Enterococcus faecium* and *Bacillus subtilis* [8], reduction of *Salmonella* and *Campylobacter* colonization [55, 57, 58], prevention or treatment *E. coli* infection in broilers [59]

Regarding poultry industry in Egypt, studies have recently directed to the impact of probiotics in poultry and poultry meat production; it was found that probiotic cell-free supernatants of four probiotic strains, *Lactobacillus rhamnosus*, *Lactobacillus fermentum*, *Pediococcus acidilactici* and *Lactobacillus delbrueckii subsp. lactis* could significantly inhibit the growth of *Clostridium perfringens* in poultry meat during storage as well as their antioxidant effect [60].

#### 5. Antibiotic resistance associated with probiotics:

The genes transformation is one of the serious risks combined with probiotics; they have antibiotic resistance genes that may be transferred to certain pathogenic microorganisms, leading to emergence of antibiotic resistance microorganisms [61]. Therefore, it is very important criteria should be considered at choosing of probiotics; *Lactobacillus*, *Bacillus* and *Enterococcus* are of greater risks, as they carry transferable genes, while *Bifidobacteria* are considered less risky as these bacteria are non-transferable [63].

However, even the antibiotic resistance genes present, the probiotic strains may not be dangerous if these genes are intrinsic; in chromosomes and not transferable. Nevertheless, precautions should be taken in used probiotics to prevent antibiotic resistance genes transfer [63].

#### 6. Probiotics action mechanisms:

Major action mechanisms of probiotic include competitive exclusion of pathogenic microorganisms, improvement of the epithelial barrier of GIT, increased mucosal adhesion of intestine, and inhibition of pathogen adhesion, releasing of anti-microorganism molecules, and immune system modulation as shown in figure (2) and table (2) [64, 65].

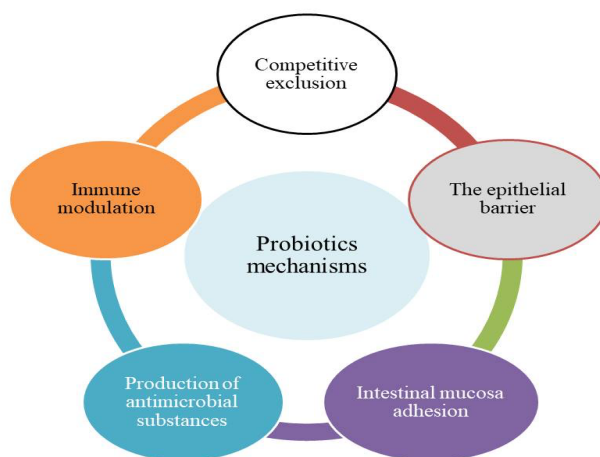


Fig. (2): probiotics action mechanisms.

### 6.1. Competitive exclusion:

The exact mechanism of action of probiotics associated with "competitive exclusion" or "bacterial antagonism" which refers to the physical colonization blocking of pathogen and it depends on microbe-to-microbe interaction by competition for available epithelial adhesion sites and nutrients figure (3) [10].

Primary and secondary metabolites; organic acid, volatile fatty acid (VFA), and lactic acid lower the gut pH that is detrimental to the survival of pathogenic bacteria such as *Salmonella* and *E. coli* [66].

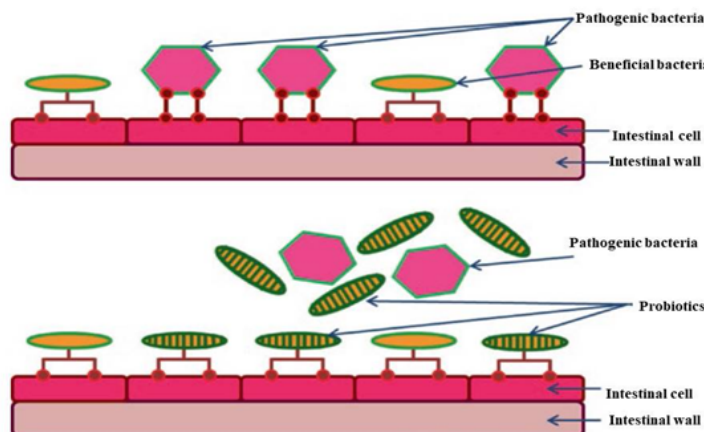


Fig. (3): competition between probiotics and pathogenic bacteria for binding sites on the intestinal epithelium.

### 6.2. The epithelial barrier:

There is permanent contact between intestinal epithelium cells besides their contents and the dynamic, variable enteric bacteria. The major defense mechanism which protects the host from the microorganisms is the intestinal barrier and its integrity [67]. The production of mucous, antimicrobial molecules, IgA secretion, and the adhesion complex of the epithelial junction are defense mechanisms for the epithelial barrier [68].

### 6.3. Intestinal mucosa adhesion:

The intestinal mucosa adhesion plays an important role for preventing pathogen colonization and pathogens antagonism. It is considered one of the most selection criteria of proper probiotics. Lactic acid bacteria (LABs) display different surface determinants that involved in interaction with intestinal epithelial cells (IECs) and mucous membrane. Mucin is a glycoprotein that secreted from IECs, and represented as the main component of mucous. It contains immunoglobulins, free proteins, salts, lipids that prevent pathogenic bacteria adhesion [69].

Table (2): summary of different mechanism of action of probiotics in the GIT [70].

Target	Mode of Action
1. Promote epithelial cell homeostasis	<ul style="list-style-type: none"> <li>• Enhance barrier function</li> <li>• Promote cytoprotective responses</li> <li>• Improve cell survival and increase mucin production.</li> </ul>
2. Neuromodulatory effects	<ul style="list-style-type: none"> <li>• Induce receptors on epithelial cells</li> <li>• Reduce visceral hypersensitivity and stress response</li> </ul>
3. Block effects of pathogenic bacteria	<ul style="list-style-type: none"> <li>• Reduce pathogen binding</li> <li>• Decrease luminal pH</li> <li>• Produce antibacterial bacteriocins</li> </ul>
4. Nutritional benefits	<ul style="list-style-type: none"> <li>• Assist in the breakdown of un-digestible foods to produce usable nutrients</li> </ul>

**6.4. Production of antimicrobial substances:**

Probiotics can produce various molecules that inhibit pathogenic bacteria such as hydrogen peroxide, bacteriocins and organic acids [71, 72]. There are three different types of protective cultures; antagonistic bacteriocinogenic protective cultures acting by releasing one or more bacteriocins or bacteriocin-like substances, figure (4), non-bacteriocinogenic protective cultures table (3) and the last one is protective cultures producing antifungal molecules figure (5) [10, 73- 76].

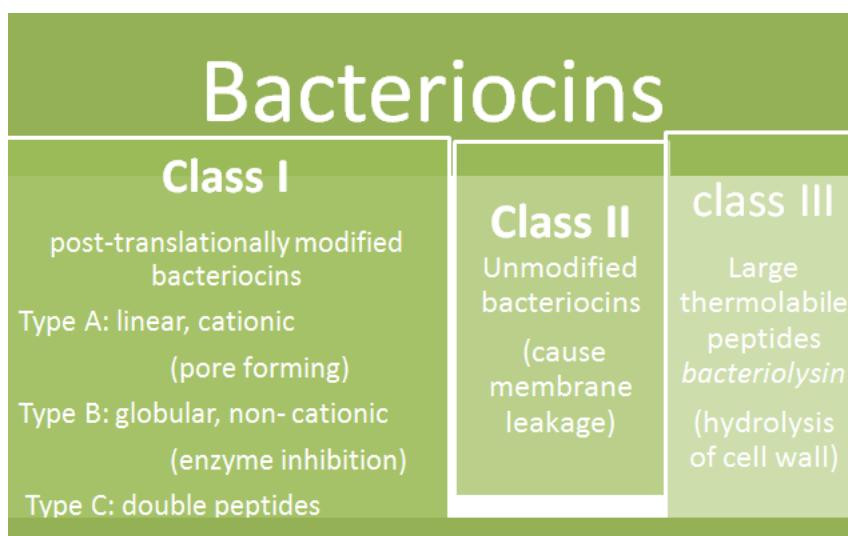


Figure (4): different bacteriocin classes.

Table (3): Non-bacteriocin antimicrobial compounds (Gioia and Biavati 2018).

Non-bacteriocin antimicrobial	Substances	Mode of action	Target
Organic acids	Lactic acid	Decrease the pH	Broad spectrum against non-acidophilic microorganisms
	Acetic acid	Undissociated hydrophobic form	

	Propionic acid (in traces amounts)	Disruption of the cytoplasmic membrane and interference with membrane potential	
	Carboxylic acids	Reduction in intracellular pH	
	Cinnamic acid derivatives		
	d-glucuronic acid		
	Salicylic acid		
	Benzoic acid		
	Hydroxybenzoic acids		
CO <sub>2</sub>	Heterofermentative LAB	Creates an anaerobic environment	Aerobic bacteria
Diacetyl	Produced during citrate fermentation by some strains		Gram-negative bacteria are more sensitive than Gram-positive bacteria
Hydrogen peroxide	Produced by flavoprotein oxidases in presence of oxygen	Oxidative damage of proteins Increase membrane permeability	Antimicrobial
Fatty acids	Long-chain hydroxylated fatty acids (C8–C12)	Partition of the lipid bilayers of fungal membranes resulting in loss of membrane integrity	Antibacterial and antifungal activity against a broad spectrum of yeasts and molds

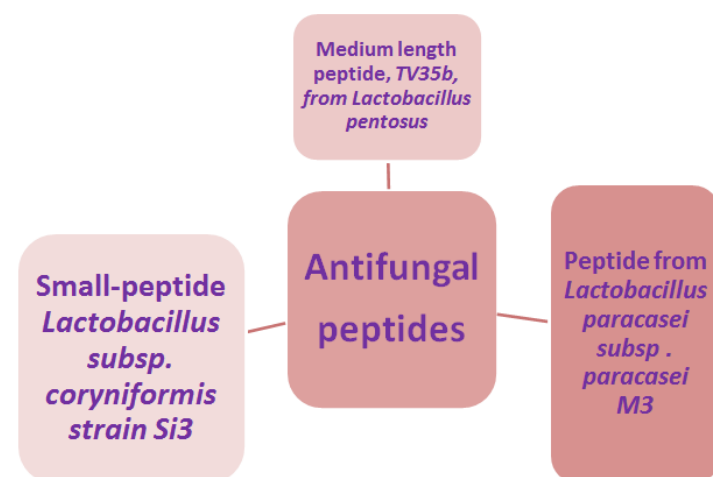


Figure (5): antifungal peptides produced by different probiotic strains.

### Conclusion and Future Prospects

Usage of probiotics leads to many health and production benefits in farm animals and poultry. Definitely, they will render as the best substitutional option for antibiotics in animal husbandry for obtaining of safe meat, milk and egg production. Many feedbacks of researchers deserve extra benefit of

probiotic usage, and directed to find the optimal dose and proper strain of probiotic microorganism for certain required action. Refinement in their form and delivery methods will also help in fulfilling maximum potential of probiotics. Further area of benefit can also be explored which can be achieved via their prospect use with more clinical trials and promising research data. Moreover, better understanding of their mechanisms of action will help in profiting extra advantages. Although significant amount of work is available declaring positive impact of using probiotics in veterinary field, still more research is required to come out with some standard protocol for their applications.

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