

# Probiotics in animal nutrition and health

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## Abstract

The use of probiotics for farm animals has increased considerably over the last 15 years. Probiotics are defined as live microorganisms which can confer a health benefit for the host when administered in appropriate and regular quantities. Once ingested, the probiotic microorganisms can modulate the balance and activities of the gastrointestinal microbiota, whose role is fundamental to gut homeostasis. It has been demonstrated that numerous factors, such as dietary and management constraints, can strongly affect the structure and activities of the gut microbial communities, leading to impaired health and performance in livestock animals. In this review, the most important benefits of yeast and bacterial probiotics upon the gastrointestinal microbial ecosystem in ruminants and monogastric animals (equines, pigs, poultry, fish) reported in the recent scientific literature are described, as well as their implications in terms of animal nutrition and health. Additional knowledge on the possible mechanisms of action is also provided.

Keywords: probiotics, live microorganisms, livestock, microbial interactions, gastrointestinal tract

## 1. Introduction

The gastro-intestinal tract (GIT) of domestic animals harbours dense and complex microbial communities, which can be composed of bacteria, protozoa, fungi, archaea, and viruses. Considerable research has been devoted during the last 30 years to characterisation of digestive ecosystems in terms of microbial composition and functional diversity, which has led to a better understanding of the major contribution of the gut microbiota to animal nutrition and health. Amongst the beneficial effects, GIT microbial communities are involved in digestion and fermentation of plant polymers which is of particular importance in herbivorous animals. The indigenous gut microbiota is also responsible for the synthesis of vitamins; bioconversion of toxic compounds to non toxic residues; stimulation of the immune system; maintenance of gut peristalsis and intestinal mucosal integrity, and plays a barrier role against colonisation by pathogens.

Numerous environmental factors are able to affect the composition and functions of gut microbiota in livestock

animals. Indeed, feeding practices, composition of animal diets, farm management and productivity constraints are parameters which can influence the microbial balance in the GIT and consequently affect feed efficiency, digestive welfare and health of the animals. An abrupt shift from forage-based to high readily-fermentable diet has been shown, for example, to induce important modifications of the ruminal microbial communities, leading to an increased risk of ruminal acidosis. Weaning represents also a critical period during which the still immature gut microbiota has to face an abrupt change in diet, which leads to an increase in the susceptibility of the young animals to pathogen colonisation.

In this context, the possibility to use feed supplements to achieve better animal health, welfare and productivity through manipulation of the GIT microbial ecosystem has gained considerable attention in the last 25 years. Growthpromoting antimicrobials, such as ionophore antibiotics, have been widely distributed and are still used in some countries. However, due to increasing safety concerns about the risk of releasing antibiotic resistance in the environment, and the persistence of chemical residues in animal products, other strategies based on supplementation of more 'natural' products such as probiotics, have been developed to improve herd health and productivity. Increasing amounts of scientific data are supporting the view that these products, which are defined as a source of live (viable) naturally occurring microorganisms, can beneficially affect the balance of GIT microbiota and that they have a real benefit in animal nutrition and health.

### 2. Probiotics for ruminants and monogastric herbivores

The most significant effects of probiotics have been reported when they have been included in the diet of animals during particularly stressful periods for the gut microbiota and the animal: at weaning; at the beginning of the lactation period; and after a dietary shift from high forage to high readily fermentable carbohydrates (Table 1).

In adult ruminants, probiotics have mostly been selected to target the rumen compartment, which is the main site of feed digestion. The rumen microbial ecosystem consists of a wide diversity of strictly anaerobic bacteria, ciliate protozoa, anaerobic fungi, and archaea which are responsible for degradation and fermentation of 70-75% of the dietary compounds. The most common marketed products for ruminants are live yeast (Saccharomyces cerevisiae) preparations. In dairy ruminants, live yeasts have been shown to improve performance, the most consistent effects being an increase in dry matter intake and milk production (Jouany, 2006; Sniffen et al., 2004; Stella et al., 2007). Also, in beef cattle, growth parameters (average daily gain, final weight, intake, feed to gain ratio) have been reported to be improved by daily live yeast supplementation (Lesmeister et al., 2004). Some of these benefits have been related to greater total culturable ruminal bacterial population densities (Newbold et al., 1995, 1996), stimulated growth and fibre-degrading activities of cellulolytic microorganisms (Chaucheyras-Durand and Fonty, 2001; Mosoni et al., 2007) leading to increased fibre digestibility (Guedes et al., 2008; Marden et al., 2008) (Figure 1).

There is also an increasing amount of evidence that live yeast stabilises ruminal pH and decreases the risk of acidosis (Chaucheyras-Durand *et al.*, 2008; Marden *et al.*, 2008).

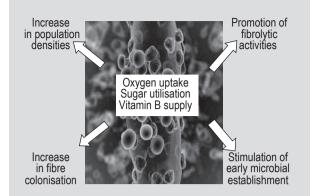


Figure 1. Main effects and mechanisms of action of live yeast probiotics on ruminal fibre-degrading communities.

Ruminal acidosis continues to be a common digestive disorder in high producing dairy or beef cattle, and the severity of acidosis can vary from acute forms due to lactic acid overload to subacute forms due to the accumulation of volatile fatty acids (Nagaraja and Titgemeyer, 2007). Acidosis is not only responsible for a decrease in animal performance, but is also often related to health issues such as laminitis, bloat, or liver abscess (Nocek, 1997; Enemark, 2008). In vitro studies have reported that live veasts could influence the balance of lactate-metabolising bacteria, by limiting lactate production by Streptococcus bovis and favouring lactate uptake by Megasphaera elsdenii or Selenomonas ruminantium (Chaucheyras et al., 1996; Nisbet and Martin, 1991; Rossi et al., 2004). Brossard et al. (2006) reported that one strain of S. cerevisiae could prevent pH decrease by stimulating certain populations of ciliate protozoa, which rapidly engulf starch and thereby effectively compete with amylolytic, lactate-producing bacteria. Regarding bacterial probiotics, lactate-producing bacteria (Enterococcus, Lactobacillus), which would sustain a constant level of lactic acid, thus allowing the lactateutilising species to flourish (Nocek et al., 2002; Nocek and Kautz, 2006) may represent a possible means to limit acidosis in high-concentrate fed animals. M. elsdenii or *Propionibacterium* spp., which utilise lactate as an energy source, could be administered as direct-fed microbials to avoid ruminal lactate engorgement (Klieve et al., 2003; Stein et al., 2006).

#### Table 1. Main targets for probiotics' use in ruminants.

Young ruminants	Dairy cattle	Beef cattle
Promoting optimal maturation of the rumen microbiota Increasing digestive safety at weaning Reducing risk of pathogen colonisation	Increasing milk yield and quality Increasing feed efficiency Promoting health (limit acidosis)	Promoting weight gain Increasing feed efficiency Promoting health (reduce acidosis) Limiting shedding of human pathogens

A growing interest for using probiotics is to reduce digestive carriage by adult ruminants of human pathogens, such as *Escherichia coli* O157 or *Salmonella*. Certain strains of *Lactobacillus acidophilus* have shown to decrease numbers of *E. coli* O157 in feedlot cattle faeces (Tabe *et al.*, 2008; Younts-Dahl *et al.*, 2004, 2005) or *in vitro* in sheep faecal suspensions (Chaucheyras-Durand *et al.*, 2006) and also appear to reduce shedding of *Salmonella enterica* (Stephens *et al.*, 2007). Distribution of probiotics on farms would represent a very practical strategy to limit pathogen release in the environment and thereby the risk of foodborne infections in humans.

Reducing the environmental impact of livestock, for example by mitigating methane excretion by ruminants, which is estimated to represent 3-5% of the global warming power, is also an increasing concern and probiotics may represent an interesting ecological tool to achieve this goal (Martin et al., 2006). For example, hydrogen utilisation and acetate production by a ruminal acetogen bacterial isolate have been shown to be improved in vitro by the addition of a yeast strain, even in the presence of methanogens (Chaucheyras et al., 1995). Other yeast strains have been screened in Rusitec for their capacity to reduce methanogenesis (Newbold and Rode, 2006). The recent isolation of high hydrogen-utilising bacterial species from diverse gut environments could also offer the possibility to increase the ruminal contribution of alternative reductive acetogenesis (Klieve and Joblin, 2007). An increase in the proportion of hydrogen utilised for acetate production instead of methane production would also be interesting from an energetic point of view for the animal, as acetate is a source of energy for the ruminant, whereas the eructated methane represents a loss of 2-12% of gross energy intake (Martin et al., 2006).

In young pre-ruminants, bacterial probiotics such as lactic acid bacteria (*Lactobacillus* spp., *Bifidobacterium* spp.,

*Enterococcus* spp., *Propionibacterium* spp.) or *Bacillus* spores generally target the small intestine, as the rumen is not yet developed, and they represent an interesting means to stabilise the gut microbiota and limit the risk of pathogen colonisation. However, live yeast distributed from the first days after birth have been reported to favour microbial colonisation and the set-up of fermentative capacities in the rumen (Chaucheyras-Durand and Fonty, 2002). Improved weight gain and rumen development in young calves have been reported with several products (Abu-Tarbousch *et al.*, 1996; Adams *et al.*, 2008; Galvao *et al.*, 2005).

In horses, whose targeted digestive compartment is the caecum-colon, probiotic distribution appears particularly relevant in case of stress (e.g. transportation) or during distribution of high concentrate diet (Table 2). Live yeasts have been demonstrated to elicit an increase in fibre digestibility in the colon and modulate the balance of hindgut bacterial communities, leading to a decreased risk of lactic acidosis (Jouany *et al.*, 2008; Médina *et al.*, 2002).

## 3. Probiotics for pigs and poultry

The most common probiotics for monogastric animals are yeasts (*Saccharomyces boulardii*), and bacteria (*Lactobacillus* spp., *Enterococcus* spp., *Pediococcus* spp., *Bacillus* spp.) targeting the hindgut (caecum, colon) which harbours an abundant and very diverse microbial population mainly composed of bacteria and archaea.

In gestating sows, the administration of probiotics has shown beneficial effects on feed intake and average live weight (Böhmer *et al.*, 2006) with at the same time a greater size and vitality of the litter (Hong et al., 2005; Taras *et al.*, 2005, 2006) (Table 3). Oral administration of probiotics immediately after caesarean section delivery has been shown to alter the initial mucosa-associated colonisation pattern of preterm formula-fed piglets, and

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Gestating mares	Foals	Racing horses
Increase diet digestibility Improve milk quantity and quality	Promote growth Limit diarrhoea	Avoid hindgut disorders (acidosis, colic) and increase digestibility of diet Limit stress (transportation, race, etc.)

#### Table 3. Main applications for probiotics' use in pigs.

Gestating sow	Lactating sow and piglets	Fattening pigs
Improve diet digestibility Limit constipation Decrease stress	Improve colostrum quality, milk quality and quantity Increase litter size and vitality Increase piglet weight Reduce risk of diarrhoea	Improve feed efficiency Improve meat quality Reduce risk of diarrhoea

thereby reduced the bacterial-dependent mucosal atrophy and GIT dysfunction preceding necrotising colitis (Siggers *et al.*, 2008), which is the most serious gastrointestinal disease afflicting preterm piglet neonates (Sangild *et al.*, 2006). In addition, from birth to post-weaning piglets are very sensitive to gut colonisation by pathogenic bacteria (*E. coli, Clostridium difficile, Clostridium perfringens, Salmonella, Listeria*), parasites (*Isospora, Cryptosporidium*) or viruses (*Coronavirus, Rotavirus*), which are responsible for growth reduction and diarrhoea. Probiotics are therefore recommended during this period and numerous studies have demonstrated the efficacy of such products (Casey *et al.*, 2007; Konstantinov *et al.*, 2008; Lallès *et al.*, 2007; Taras *et al.*, 2007).

Performance benefits have also been reported after weaning, as for example with *S. boulardii* (Bontempo *et al.*, 2006). In this study, the yeast probiotic promoted a 'healthy' intestine by encouraging an early restoration of the intestinal mucosal thinning generally occurring at weaning, and would possibly improve local resistance to infection. Similar findings have been reported with *Pediococcus acidilactici*-based probiotic supplementation (Di Giancamillo *et al.*, 2008). The benefits for intestinal IgA secretion and reduction of translocation of enterotoxinogenic *E. coli* have also been observed with *S. boulardii* or *P. acidilactici* given to piglets (Lessard *et al.*, 2009). Similar findings on modulation of IgA development, together with a decreased ileal prevalence of ETEC, have been reported with a strain of *Lactobacillus sobrius* (Konstantinov *et al.*, 2008).

In fattening pigs, growth performance has been shown to be improved in the presence of probiotics: some of them (lactic acid bacteria) also have beneficial effects on the microbiological and nutritional quality of liquid feed (Moran *et al.*, 2006; Van Winsen *et al.*, 2001).

In poultry, benefits of probiotic supplementation (live yeast or bacteria) are reported in broilers' performance and health, with evidence of increased resistance of chickens to *Salmonella, E. coli* or *C. perfringens* infections (Banjeree and Pradhan, 2006; Higgins *et al.*, 2007, 2008; La Ragione *et al.*, 2003, 2004). Probiotics can increase feed efficiency and productivity of laying hens (Kurtoglu *et al.*, 2004; Yörük *et al.*, 2004), and an improvement in egg quality (decreased yolk cholesterol level, improved shell thickness, egg weight) has also been reported (Kurtoglu *et al.*, 2004; Xu *et al.*, 2006).

## 4. Probiotics in aquaculture

When looking at probiotics intended for an aquatic usage, it is important to take into account the intricate relationship that an aquatic organism has with its direct environment, compared to terrestrial animals (Kesarcodi-Watson *et al.*, 2008). Gram-negative facultative anaerobic bacteria are dominant in fish and shellfish digestive tract, but the intestinal microbiota of aquatic animals may change very rapidly with the intrusion of microbes coming from water and food (Gatesoupe, 1999). This is probably the reason why a large number of probiotics developed in aquaculture are bacteria directly originating from the aquatic environment. However, more 'traditional' bacterial or yeast species marketed for animal nutrition (Lactobacillus, Pediococcus, Bacillus, and S. cerevisiae) are also used. They can target fish eggs and larvae, fish juveniles and adults, crustaceans, bivalve molluscs and also live food such as rotifers, artemia, or unicellular algae (Verschuere et al., 2000). Growth-promoting effects, through better feed utilisation and digestion, as well as biological control of pathogen colonisation are the most important expected benefits of probiotic applications. Disease outbreaks caused by Vibrio spp. or Aeromonas spp. have been recognised as a significant constraint on aquaculture production (Verschuere et al., 2000), particularly in the shrimp subsector, where vibriosis is currently one of the main diseases identified (Castex et al., 2008). Whereas in vitro, antagonism to pathogens has been clearly demonstrated for a wide range of probiotic strains (Gatesoupe, 1999), in vivo evidence of efficacy is still very scarce. A recent study shows that under pond conditions, the distribution of a *P. acidilactici*-based probiotic could be an effective treatment for limiting prevalence and load of Vibrio nigripulchritudo strains in haemolymph of marine shrimps (Castex et al., 2008). Some probiotics have been shown to protect rainbow trout against skin infections caused by Aeromonas bestiarum and a eukaryotic pathogen, Ichthyophthirius multifiliis (Pieters et al., 2008).

# 5. Modes of action

Several mechanisms have been proposed to explain the effects of probiotics and it is likely that the positive results reported in the different animal studies are due to a combination of some, if not all, of these. The metabolic activities of the probiotic strains and survivability throughout the gut appear to be of great importance for an optimal efficacy (Chaucheyras-Durand et al., 2008). Effects are also greatly dependent on the strain used (Newbold et al., 1995). In monogastrics, the production of organic acids (lactic or acetic acid) by bacterial probiotics can help decrease the gut pH, create more favourable ecological conditions for the resident microbiota and decrease the risk of pathogen colonisation (Servin, 2004). The release of antimicrobial peptides, such as bacteriocins, which inhibit the growth of pathogenic bacteria, or production of enzymes able to hydrolyse bacterial toxins (Buts, 2004) have been demonstrated. Some strains can competitively exclude pathogenic bacteria through their higher affinity for nutrients or adhesion sites (La Ragione et al., 2003, 2004). Some probiotics produce nutrients and growth factors which are stimulatory to beneficial microorganisms of the gut microbiota. In addition to interacting and stimulating other microorganisms, probiotics also interact with the host, by influencing the immune response (Delcenserie *et al.*, 2008), or producing components able to positively affect mucosa development or the metabolism of the host's intestinal cells (Johnson-Henry *et al.*, 2008). Some probiotics can also metabolise or aid in the detoxification of certain inhibitory compounds such as amines or nitrates or scavenge for oxygen (Chaucheyras-Durand *et al.*, 2008; Marden *et al.*, 2008), which is of great importance in gut anaerobic ecosystems. Most of these mechanisms have also been proposed to explain the effects of probiotics in the human gut, where benefits both in terms of nutrition and health have been demonstrated.

# 6. Conclusions

Probiotic microorganisms, which benefit from a 'natural image, can expect a promising future in animal nutrition. Controlled research studies demonstrate that they can positively balance gastrointestinal microbiota, and thereby improve animal production and health. However, care must be taken in the way that the probiotic candidate-strains are selected. Better knowledge of the structure and activities of the gut microbiota, functional interactions between gut microbes and interrelationships between microbes and host cells represent a fundamental aspect of future probiotic research. In this context new 'omic' technologies will be very helpful to better characterise and understand the effects of probiotics on the balance of the gastrointestinal microbiota. It will be possible to select more powerful or targeted strains on a scientific basis and follow their behaviour in the host animal. Thanks to these techniques, which are complimentary to anaerobic culture methods and gnotobiotic animal or cellular models, probiotic research has had, and will also certainly have in the future, a very important place in the improvement of animal health and nutrition

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