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Review

Role of probiotics in nutrition and health of small ruminants

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Abstract

Small ruminants represent an important economic source in small farm systems and agriculture. Feed is the main component of livestock farming, which has gained special attention to improve animal performance. Many studies have been done to improve feed utilisation through addition of feed additives. For a long period, antibiotics have been widely used as growth promoters in livestock diets. Due to their ban in many countries, search for alternative feed additives has been intensified. Probiotics are one of these alternatives recognised to be safe to the animals. Use of probiotics in small ruminant nutrition has been confirmed to improve animal health, productivity and immunity. Probiotics improved growth performance through enhancing of rumen microbial ecosystem, nutrient digestibility and feed conversion rate. Moreover, probiotics have been reported to stabilise rumen pH, increase volatile fatty acids production and to stimulate lactic acid utilising protozoa, resulting in a highly efficient rumen function. Furthermore, use of probiotics has been found to increase milk production and can reduce incidence of neonatal diarrhea and mortality. However, actual mechanisms through which probiotics exert these functions are not known. Since research on application of probiotics in small ruminants is scarce, the present review attempts to discuss the potential roles of this class of feed additives on productive performance and health status of these animals.

Key words: digestibility, goat, immunity, performance, probiotics, ruminal ecosystem, sheep

Introduction

For many years, nutritionists have been interested in manipulating the microbial ecosystem of the rumen for improving feed utilisation, therefore animal production and health, as well as, in more recent years, safety and quality of food products from ruminants. These goals can be achieved by facilitating desirable fermentation, minimising ruminal disorders and excluding pathogens. Antibiotics, probiotics and prebiotics have been studied with the objective to manipulate the microbial ecosystem and fermentation characteristics in the rumen and the intestinal tract of livestock animals (Seo et al. 2010).

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Antibiotic growth promoters have been widely used in the past, as feed additives for livestock to improve feed consumption, to increase production and to prevent infections (Morrill et al. 1977). However, use of antimicrobial drugs in livestock has various disadvantages, e.g., development of bacterial antibiotic resistance in animals and humans (Fey et al. 2000, Budino et al. 2005), transfer of antibiotic residues into the food chain (Chen et al. 2005) and elimination of various, potentially beneficial, microorganisms (Spika et al. 1987). As the result of consumer demand, antibiotics have been banned as feed additives. Vancomycin-resistant enterococci were the trigger to ban antibiotics as feed additives in Sweden back in 1986 (Kuehn et al. 2005). Then, some further antibiotics have been banned in the European Union in January 1997 (Commission Directive 97/6 EC) and most of the remaining ones in December 1998 (Commission regulations 2788/98 and 2821/98). Hence, a search for suitable alternatives with similar beneficial effects has been intensified. Among possible alternatives are prebiotics, probiotics and symbiotics.

Nowadays, probiotics are widely used as feed additives in livestock animals and have been defined as non-pathogenic microorganisms. Objective of their use is to improve production performance and disease prevention through maintenance of a healthy gastrointestinal environment and improved intestinal function (Chaucheyras-Durand et al. 2008, Mountzouris et al. 2009). Further, probiotics enhance rumen microbial ecosystem (Sandine 1979, Musa et al. 2009), nutrient digestibility (Krehbiel et al. 2003, Abd El-Ghani 2004), nutrient absorption and feed conversion rate (Antunovic et al. 2006, Whitley et al. 2009) leading to better production performance of animals in which they are administered. It is also considered that probiotics can have an antagonistic action to pathogenic microorganisms for adhesion sites and nutritional growth factors (Rolfe 2000, Guillot 2003), can reduce incidence risk of intestinal infections (Casas and Dobrogosz 2000) and can restore gut microflora in cases of diarrhea (Musa et al. 2009). Probiotics have also been found to enhance host immunity through stimulation of immunoglobulins, macrophages, natural killer cells and cytokine production. However, exact mechanisms by means of which probiotics exert their beneficial roles have not been fully elucidated (Koop-Hoolihan 2001).

Objective of the present review paper is to focus mainly in the role of probiotics in nutrition and health of small ruminants. The article describes the potential effects of probiotics supplementation on rumen microbial ecosystem and nutrient digestion, growth performance, carcass characteristics, blood metabolites, intestinal microflora, and on animal defensive abilities.

Definitions and types of probiotics

The concept of probiotics probably evolved from a theory first proposed by Nobel Prize-winner Russian scientist Metchnikoff. In 1907, he hypothesised in his book entitled "The prolongation of life" that the long lasting life of Bulgarian peasants was the result of consumption of fermented milk products. The term "probiotics" was first mentioned by Lilly and Stillwell (1965) to describe substances secreted by a microorganism that promote the growth of another microorganism(s).

Subsequently, Parker (1974) proposed that probiotics were organisms and substances which contributed to intestinal microbial balance. Fuller (1989) then defined probiotics as a live microbial feed supplement including *Lactobacillus* species, *Bifidobacterium* species, *Streptococcus* species, yeasts and molds (Table 1), which beneficially affected host animals by improving their microbial balance. He also mentioned that probiotics were bio-preparations containing living cells or metabolites of stabilised autochthonous microorganisms which might optimise colonisation and composition of gut microflora in animals and humans and might have a supporting effect on digestive processes and immunity of hosts. Probiotics have been defined as non-pathogenic microorganisms, which, when ingested, exert a positive influence on the host health or physiology (Dunne et. al. 1999). They can restore and maintain balance of the desirable microorganisms in times of stress or disease and enhance growth of young animals (Simon et al. 2001, Antunovic et. al. 2005).

Probiotics are viable microorganisms and, when administered in sufficient numbers, can modify the microflora of the digestive tract of the host (Rook and Burnet 2005) in a way resulting in improved health and production. Several microbial species, mainly bacteria (lactic acid and non-lactic acid bacteria), yeasts (dairy strains) or fungi are considered as probiotics (Tripathi et al. 2008).

Strains for potential use were characterised as normal inhabitants of the target species. They have the ability to adhere and colonise epithelial cells of the gut (Musa et al. 2009). Moreover, probiotic strains are not hydrolyzed or absorbed in the upper part of the gastrointestinal tract (GIT). These organisms are genetically stable, able to produce antimicrobial substances and to modify the colonic flora in favour of a healthier composition and hence induce luminal or systemic effect that is beneficial to the host health (Kaur et al. 2002, Parvez et al. 2006).

Table 1. Microorganisms used as probiotics (Fooks and Gibson 2002, Lodemann et al. 2006, Seo et al. 2010).

<table>
<tr><th>Genus</th><th>Species</th><th>Genus</th><th>Species</th></tr>
<tr><td rowspan="3">Lactobacillus</td><td rowspan="3">L. acidophilus
L. casei
L. rhamnosus
L. reuteri
L. plantarum
L. fermentum
L. brevis
L. helveticus
L. delbruckei
L. gallinarum
L. salivarius</td><td>Pediococcus</td><td>P. pentosaceus</td></tr>
<tr><td>Bacillus</td><td>B. subtilis
B. cereus
B. toyoi
B. natto
B. mesentericus
B. licheniformis</td></tr>
<tr><td>Bifidobacterium</td><td>B. bifidum
B. pseudolongum
B. breve
B. thermophilum</td></tr>
<tr><td>Lactococcus</td><td>L. lactis</td><td rowspan="2">Saccharomyces</td><td>S. cerevisiae</td></tr>
<tr><td>Enterococcus</td><td>E. faecium</td><td>S. boulardii</td></tr>
<tr><td>Streptococcus</td><td>S. thermophilus</td><td>Escherichia</td><td>E. coli (avirulent strains)</td></tr>
<tr><td>Aspergillus</td><td>A.oryzae
A. niger</td><td></td><td></td></tr>
</table>

Probable modes of action of probiotics

Although the probiotics concept has been recognised for many years, their precise mode of action has not been fully elucidated. Principal microorganisms used as probiotics for ruminants are bacteria and yeasts. Their mode of action can be distinguished as detailed below.

Yeast probiotics

Various modes of action have been proposed to explain effects that yeast cultures may have on rumen fermentation and ruminant production. Feeding of yeast stabilises rumen pH, increases total volatile fatty acids (VFAs) and reduces ammonia concentration (Erasmus et al. 1992, Newbold et al. 1996, Abd El-Tawab 2007, Bakr et al. 2015). Increased bacterial population is central to the action of the yeast in improving ruminant productivity (Wallace and Newbold 1992). Yeasts may stimulate growth and enzymatic activity of cellulolytic bacteria, as well as improve microbial protein synthesis and fibre digestibility (Yoon and Stern 1996, Bomba et al. 2002). Yeast supplementation reduces the redox potential that creates better conditions for growth of strict anaerobic microorganisms, produces specific factors, e.g., vitamin B_{12} or branched chain fatty acids, that way stimulating synthesis of microbial biomass in the rumen (Chademanaet al. 1990, Jouany 2006, Chaucheyras-Durand et al. 2008). Moreover, yeast supplementation reduces rumen acidosis, stimulates growth and activity of lactic acid-utilising rumen bacterium *Selenomonas ruminantium* (Nisbet and Martin 1990, 1991). Above activities of yeast lead to stimulation of rumen fermentation and contribute to improved digestibility and feed utilisation.

Bacterial probiotics

Lactobacilli and bifidobacteria are the two genera most frequently used as bacterial probiotics. Various possible mechanisms of action have been considered. Some bacterial probiotic strains can competitively exclude pathogenic bacteria through colonisation and adhesion to gut mucosa. This competition could be for receptors (Guillot 2003) or for nutrients (Bomba et al. 2002, Tripathi and Karim 2010), inhibiting colonisation by harmful pathogens (Abu- Tarboush et al. 1996).

Bacterial probiotics antagonise pathogen growth through production of a variety of inhibitory substances for both Gram-positive and Gram-negative pathogenic bacteria. Potentially inhibitory agents may include organic acids, hydrogen peroxides and bacteriocins (Bomba et al. 2002, Marinho et al. 2007, Schierack et al. 2009). Moreover, many lactobacilli produce antibiotic metabolites (acidophillin, acidolin, lactobacillin, and lactocidin), which have an inhibitory activity against Salmonella, *Shigella*, *Staphylococcus*, *Proteus*, *Klebsiella*, *Pseudomonas*, *Bacillus* and *Vibrio* spp., as well as against enteropathogenic *Escherichia coli* (Mikolajcik and Hamadan 1975).

Probiotic bacteria can exert an immunemodulatory effect through stimulation of the immune system and regeneration of intestinal mucosa (Isolauri et al.

2001, Vondruskova et al. 2010). Probiotics can improve immunoglobulin production (Perdigon et al. 1995) and enhance the activity of macrophages and natural killer cells (Matsuzaki and Chin 2000). They can also regulate anti- and pro-inflammatory cytokine production (Shu et al. 2001, Roselli et al. 2005).

Effect of probiotics on rumen

Rumen pH

Effects of probiotic supplementation of small ruminants have not been clearly defined. Some researchers have found no effect in rumen pH of small ruminants (Doreau et al. 1998, Tripathi et al. 2008), whilst others have recorded an increase in rumen pH (Radev 1999, Abd El-Ghani 2004). In contrast, Kowalik et al (2011) and Tripathi and Karim (2011) have reported a reduction in rumen pH after supplementation with *Saccharomyces uvarum* (ATCC9080; SU) or an equal mixture of *Kluyveromyces marximanus* (NRRL3234; KM), *Saccharomyces cerevisiae* (NCDC42; SC) and *S. uvarum* (ATCC9080; SU) to growing lambs. Other researchers have indicated that the dietary feeding of probiotics stabilised rumen pH (Chaucheyras-Durand and Fonty 2006) leading to efficient rumen functioning, hence preventing risk of sub-acute ruminal acidosis (Lettat et al. 2012).

Various mechanisms have been identified to explain effects of probiotics on rumen pH regulation. Probiotics may compete with *Streptococcus bovis* and/or lactobacilli for glucose utilisation, thus reduced amounts of lactic acid would be produced (Chaucheyras et al. 1996). On the other hand, probiotics may release malate and small peptides, which in turn may stimulate L-lactate use by *Megasphaera elsdenii* and *Selenomonas ruminantium* (Nagaraja 2012). Further, probiotics can modify protozoa concentrations in the rumen (Galip 2006) which regulate lactic acid concentrations, as ruminal protozoa compete with *S. bovis* for glucose uptake and can metabolise lactic acid (Nagaraja 2012). Additionally, rumen protozoa can ferment starch at a slower rate than amylolytic bacteria (Mendoza et al. 1993).

Rumen volatile fatty acids

Effects of probiotics on ruminal volatile fatty acids (VFAs) are still not fully clarified. Some authors found that feeding of probiotics to small ruminants increased VFA production (Sadiek and Bohm 2001, Abd El-Ghani 2004). Increase of total VFAs concentration in yeast supplemented animals may be attributed to decreased methane production and consequent energy loss saving, thus additional energy would be employed for VFA (Williams and Newbold 1990). However, other studies recorded a significant reduction in ruminal VFA formation in growing lambs or adult goats given probiotic supplemented diets (Kowalik et al. 2011, Tripathi and Karim 2011). Nevertheless, some researchers have found no effect of probiotic feed additives in total VFA concentrations in the rumen (Galip 2006, Tripathi et al. 2008, Soren et al. 2013).

Rumen protozoa

The influence of probiotics on rumen protozoa varies according to the type of the probiotics and the protozoan species in the rumen. It has been found that dietary supplementation of rams with *S. cerevisiae* (YS), sodium bicarbonate (BC) or their combination (YS+BC) did not lead to changes of the proportions of the various protozoa (Galip 2006) although there was a tendency for *Epidinium* spp. to increase in yeast culture treatments. However, *Diplodinium* spp. populations tented to be smaller before feeding in animals given YS, BC or BC+YS, than in unsupplemented controls. Similar findings have been reported by Mathieu et al. (1996), supplementation with *S. cerevisiae* had no significant effects in ruminal protozoan population of sheep. Moreover, Arakaki et al. (2000) reported a smaller proportion of *Entodinium* spp. and an increase in proportion of *Dasytrichia* spp. in steers given yeast culture. However, Brossard et al. (2006) reported improved growth of protozoa population in the rumen of sheep supplemented with live yeasts (Levucell® SC, *S. cerevisiae* CNCM I-1077).

The same results were obtained by Kowalik et al. (2011), who observed that feeding of live yeast or their metabolites to adult female goats caused eight-fold reduction in the number of protozoa of the genus *Isotricha*. Nevertheless, yeast metabolite supplementation increased the total number of protozoa and the number of *Diplodinium* spp. from 2.5 $\times 10^4$ to 5.8 $\times 10^4$, while feeding of live yeast resulted in significant reduction in total protozoan populations, but an increase in populations of *Diplodinium* spp. It has also been reported that individual yeast cultures increased, but mixed yeast culture reduced total ciliate protozoa (Tripathi and Karim 2011), which would be involved in fibre utilisation, hence contribute to improved digestibility of fibre in sheep/goats supplemented with yeast culture (Kamel et al. 2004, Kritas et al. 2006). This effect is, however, likely to be small. Furthermore, rumen protozoa are known to represent more than 90% of rumen fibrolysis activity (Tripathi and Karim 2011).Increased cellular activities of proteases, α-amylase, β-glucosidase and xylase supported the fact that probiotic supplementation stimulated establishment and increase bacterial cells numbers in rumen (Newbold et al. 1995).

Rumen digestion

Probiotics can improve nutrient digestibility (Krehbiel et al. 2003, Abd El-Ghani 2004), degradation of fibres (El-Waziry and Ibrahim 2007) and ruminal digestion (Kamel et al. 2004). This may be attributed to enhancing growth and/or cellulolytic activity of rumen bacteria (Williams 19) and preventing ruminal acidosis by balancing the VFA ratios in the rumen (Arcos-Garcia et al. 2000). Haddad and Goussous (2005) have reported that supplementation with yeast culture (Diamond V®YC) in the diets of Awassi lambs had resulted in increased digestibility of dry matter (DM), organic matter (OM), crude protein (CP) and neutral detergent fiber (NDF) (676, 683, 653 and 561 g kg^{-1}, respectively) compared to controls (632, 645, 589 and 521 g kg^{-1}, respectively). In contrast, Titi et al. (2008) have reported that feeding yeast culture at a dose of 12.6 kg per tonne of dry matter ("XP" Yeast Culture, Diamond "V") had no effect on digestibility of DM, CP and NDF, but also found that digestibility of OM and acid detergent fiber (ADF) increased in lambs and kids after supplementation with yeast culture. Finally, Whitley et al. (2009) have reported improved apparent digestibility of DM, CP, NDF and ADF in meat given a diet with probiotics.

Favourable responses of ruminal digestion to yeast feeding in ruminants include increased DM intake (Erasmus et al. 2005) and improved organic matter degradation (Kamel et al. 2004). It may be possible that stimulation of rumen bacterial populations improved fibre and organic matter degradation, therefore, yeast supplementation improved availability of energy for microbial growth and the larger organic matter degradation allows increased DM intake. However, Soren et al. (2013) have reported that feeding *S. cerevisiae* or a combination of *S. cerevisiae* and *Lactobacillus sporogenes* to lambs had no effect on dry matter intake and digestibility of DM, OM and NDF. Supplementation with *S. cerevisiae* or a combination of *S. cerevisiae* and *L. sporogenes* improved CP digestibility by 18% and 14%, respectively. The digestibility of ADF was also significantly increased in supplemented animals than in controls. Significant improvement in the ADF digestibility in supplemented animals might be due to stimulated growth of cellulolytic bacteria (Chaucheyras-Durand et al. 2008, Francia Di et al. 2008), which might have contributed to increased rumen fermentation activity. Fermentation activities of bacteria, especially of cellulolytic strains, have been reported to increase by probiotic supplementation leading to improvement in fibre digestibility (El-Waziry and Ibrahim 2007). Similarly, Mousa et al. (2012) reported significant improvement in the digestibility of DM, CP and fibre in lambs supplemented with dried live yeast in diets containing a 60:40 concentrates: roughages. Nevertheless, other studies (Tripathi et al. 2008, Tripathi and Karim 2011) have reported no improvement in the digestibility of DM, OM, CP, NDF and ADF in lambs before and after weaning, when supplemented with different yeast probiotics in diet containing a high amount of concentrate. The same findings were reported in lactating goats and bucks (Abd El-Ghani 2004). However, Fayed (2001) have reported that digestibility coefficients of all nutrients of goats fed yeast culture were higher than in control animals. In a recent publication, Ghoneemand Mahmoud (2014) have reported that digestibility of most nutrients and nutritive value of feeds improved after supplementation of *S. cerevisiae* var. *ellipsoideus* Doxal strain's Thepax®, (1 × 1010 CFU g^{-1}) to lambs. Conflicting results concerning feeding of dried yeast on nutrient's digestibility between the various studies might have resulted from variations in feeding systems, animal species, age of animals, frequency of feeding, dose of yeast and strains employed, physiological state of the experimental animals, environmental conditions, ration composition and plane of nutrition (Mousa et al. 2012).

Effect of probiotics on greenhouse gas emissions

Yeast cultures based on *S. cerevisiae* are widely used in ruminant diets. Feeding of such probiotic products is widely associated with increases in livestock production, enhanced ruminal capture of ammonia into microbial protein, improving dietary N usage and reducing emissions (Chaucheyras-Durand et al. 2008). Use of yeast and other live microorganisms to specifically decrease methane emissions has been suggested (Newbold and Rode 2006); however, to date, overall effects appear to be rather small and inconsistent (Beauchemin et al. 2008). More experimental approaches based on addition of acetogens (Lopez et al. 1999), methane oxidising organisms (Valdes et al. 1996), bacteriocins and bacteriophages (McAllister and Newbold 2008) have been postulated but, while potentially promising, are some years away from commercial exploitation.

Effect of probiotics on nutrient flow to the small intestine

Yeast culture may affect the contributions of microbial protein synthesis to the nutrients profile of digesta supply to the small intestine. Williams et al. (1990) found that apparent absorption of dry matter (DM) and non-ammonia nitrogen (NAN) between duodenum and terminal ileum increased by 35 and 23%, respectively, when SC was supplemented to the diet of sheep. Further, *S. cerevisiae* tended to increase flow of DM and NAN at the duodenum, but flow at the terminal ileum was unchanged. These findings suggested that this increased flow and absorption of NAN probably represented an increase in flow of useful microbial protein to the small intestine. Yeast culture can influence the amino acid profile of the bacterial protein flowing out of the rumen, presumably by selective stimulation of growth of certain species of anaerobic bacteria (Dawson et al. 1990, Erasmus et al. 1992, Yoon and Stern 1995).

Probiotic supplementation improves the microbial activities in rumen resulting in enhanced ammonia capture to synthesise microbial protein (Erasmus et al. 1992) and have profound influence in lambs (Jouany et al. 1998a). Probiotics have been reported to enhance N-retention (Paryad and Rashidi 2009, Khalid et al. 2011) by enhancing microbial peptidolytic and proteolytic activities in the rumen (Cole et al. 1992) and post-ruminal amino acid flow (Erasmus et al. 1992, Enjalbert et al. 1999). This increase in post-ruminal amino acid flow has also been reported by other researchers (Putnam et al. 1997, Doreau and Jouany 1998). However, Hernandez et al. (2009) reported no effect of probiotic supplementation in N-retention, N-intake and faecal and urinary N in lambs fed mature orchard grass. Jouany et al. (1998b) also reported no change in urinary N excretion in response to probiotic supplementation.

Ruminal liquid and particulate outflow rates have been measured with or without fungal supplementation (Wiedmeier et al. 1987, Harrison et al. 1988, Caton et al. 1993). Data suggest that ruminal liquid outflow rate increases with fungal culture supplementation although the magnitude of response is low and unlikely to be significant with the small number of animals used in each experiment.

Effect of probiotics on growth performance of small ruminants

Studies on performance responses of sheep and goats supplemented with yeast or yeast cultures have been variable. Growth rate and efficiency of bodyweight gain were found to be similar or reduced in some studies (Agarwal et al. 2002, Erasmus et al. 2005, Kawas et al. 2007b, Tripathi et al. 2008, Tripathi and Karim 2010), while others researchers reported improved weight gain, feed consumption and feed efficiency of gain after yeast supplementation (Lesmeister et al. 2004, Stella et al. 2007). A positive effect of probiotic supplementation on nutrient intake, bodyweight gain and feed conversion rate (FCR) in small ruminants has been recorded by many researchers (Antunovic et al. 2006, Whitley et al. 2009). It has, in general, been reported that impact of probiotics in performance of animals may vary, as supplementation can increase feed intake (Abd El-Ghani 2004, Antunovic et al. 2005, Desnoyers et al. 2009), FCR (Khalid et al. 2011) or bodyweight gain (Jang et al. 2009, Hussein 2014). Haddad and Goussous (2005) found that supplementation with yeast culture of diets of Awassi lambs had resulted in increased bodyweight gain compared to controls (266 *versus* 212 g daily). Similarly, Anandan et al. (1999) found increased bodyweight gain in kids given a probiotic supplement (curds) compared to controls (4.37 *versus* 3.15 kg and 44.6 *versus* 32.1 g daily). In contrast, Titi et al. (2008) have reported that yeast supplementation had no effect on growth rate or DM intake in lambs and kids, these authors have explained a lack of beneficial effect of yeast supplementation by the high protein diet content. Moreover, Kawas et al. (2007b) mentioned that addition of yeast improved bodyweight gain in lambs fed low protein diets with no favourable effects on those fed high protein diets.

Whitley et al. (2009) have found that growth performance of kids remained unaltered in cases of probiotic (dry yeast and lactic acid producing bacteria) supplementation, except in only one trial in which significant increase in bodyweight gain and improvement of FCR were observed in the supplemented animals. On the other hand, it was reported that supplementation of sheep diets with dry live *S. cerevisiae* had also conflicting results on performance data. This feed additive may contribute to increased growth and improvement of FCR, but it has no effect on feed intake (Haddad and Goussous 2005). Other researchers found that it increased growth and feed intake with no effect on FCR (Payandeh and Kafilzadeh 2007) or that it increased feed intake with no effect on growth and feed conversion (Khadem et al. 2007) or that it had no effect in any of growth, feed intake and feed conversion (Macedo et al. 2006, Kawas et al. 2007a, Titiet al. 2008). Soren et al. (2013) observed that feeding of *S. cerevisiae* or combination of *S. cerevisiae* and *L. sporogenes* to lambs also had no effect on bodyweight and daily weight gain.

A possible positive effect of probiotics on bodyweight gain of lambs or kids might be the effect of improved cellulolytic activity resulting in improved fibre degradation (Russell and Wilson 1996), increased microbial protein synthesis leading to more amino-acid supply post-ruminally (Erasmus et al. 1992, Chaucheyras-Durand et al. 2008). Further, improved bodyweight gain may also be related to increased consumption and improved efficiency of feed utilisation in the probiotic-supplemented animals (Antonovic et al. 2006, Musa et al. 2009, Papatsiros et al. 2011). Additionally, probiotics attach onto the intestinal mucosa and prevent adhesion of potential pathogens, leading to improved nutrient digestion that may enhance dry matter intake (Seo et al. 2010).

Effect of probiotics on blood metabolites

Published information on effects of probiotics on haematological and blood biochemical parametres of small ruminants is conflicting and controversial. With regard to protein metabolism, concentrations of blood urea nitrogen (BUN) and urea decreased in lambs given a probiotic-supplemented diet (Chiofalo et al. 2004, Antunovic et al. 2005, Antunovic et al. 2006, Dimova et al. 2013). Smaller concentrations of BUN in probiotic supplemented lambs might be due to improved nitrogen utilisation by ruminal bacteria (Bruno et al. 2009). Moreover, Chiofalo et al. (2004) have attributed the reduction of blood urea concentration in lactobacilli probiotic (a mixture of *Lactobacillus acidophilus*, *Lactobacillus salivarius*, *Lactobacillus reuteri*) supplemented kids to the improved nutritional status of supplemented animals that do not resort to the amino-acid de-amination (Riis 1983) in order to acquire energy. Other researchers have found no effect of probiotic supplementation on concentrations of BUN and urea in sheep and lambs (Galip 2006, Abas et al. 2007, Soren et al. 2013). With regard to other protein metabolites, it has been recorded that concentrations of total protein, albumin and globulin in probiotic supplemented lambs have not changed (Galip 2006, Abas et al. 2007, Dimova et al. 2013, Soren et al. 2013). Only Hussein (2014) has reported increased values of plasma total protein, albumin and globulin in lambs supplemented with probiotics (5 g and 10 g of probiotics per kg of diet; Biovet-YC + a concentrate feed mixture).

Probiotic supplementation can lead to decreased blood concentrations of glucose as the result of improvement in fibre digestion, which leads to increased acetic acid and reduction of propionic acid production in the rumen (Antunovic et al. 2005, Bruno et al. 2009). On the other hand, Sayed (2003) has reported a significant increase in glucose concentration in kids and lactating ewe after probiotic supplementation. Similar findings have been observed in lambs (Hussein 2014). An increase in serum glucose levels in supplemented animals may be attributed to gluconeogenesis, as after probiotic supplementation there is improvement in gluconeogenesis due to increased propionate production, which is the main precursor of glucose with a decisive influence on the glucose blood concentration in small ruminants (Huntington and Eisemann 1988). Nevertheless, some studies (Antunovic et al. 2006, Galip 2006, Ding et al. 2008) have found that blood concentrations of glucose have not changed in lambs given diets containing probiotics.

Many studies consider that probiotic supplementation may improve the lipid profile of animals. The concentrations of total lipids, non-esterified fatty acids (NEFAs), triglycerides and low density lipoproteins (LDL) were found to be decreased in probiotic-supplemented kids or lambs (Chiofalo et al. 2004, Abas et al. 2007, Baiomy 2011). This may be attributed to an imporved metabolic status and a positive energy balance associated with probiotic supplementation. Chiofalo et al. (2004) have reported a significant reduced concentration of NEFA (control 0.78 *versus* supplemented 0.40) and triglycerides and an increased one for high density lipoproteins (HDL) in growing kids supplemented with probiotics. Moreover, probiotic supplementation had no effect in blood cholesterol concentration in kids or lambs (Chiofalo et al. 2004, Galip 2006, Soren et al. 2013, Hussein 2014). However, Abas et al. (2007) have reported that supplementation of Kivircik male lambs with *Enterococcus faecium cernelle* 68 strain (Cylactin® LBC ME 10) did not lead to reduced cholesterol concentrations, in contrast with organic acid supplementation, which did. Reduction in cholesterol concentration may be attributed to inhibition of cholesterol synthesis or direct assimilation of cholesterol (Zacconi et al. 1992).

The effects of probiotic supplementation on blood concentrations of various enzymes have received attention. Variable results have been reported for enzymatic activities after probiotic supplementation. The activities of serum aspartate aminotransferase (AST) and alanine aminotransferase (ALT) have not been found to change, while activities of alkaline phosphatase (AP) and creatinine kinase (CK) were found increased in probiotic-supplemented kids (Chiofalo et al. 2004). Increase in AP activities has been attributed to higher osteoblastic activity, thus to a greater skeletal development (Benjamin 1984), while increased CK activities might be due to improved muscular development (Avallone et al. 1993). However, Soren et al. (2013) found that serum aspartate aminotransferase (AST) and alkaline phosphatase (AP) activities have not been affected by probiotic supplementation, although activity of ALT was increased in probiotic supplemented group. In contrast, Antunovic et al. (2005) reported that probiotic supplementation to growing lambs resulted in significantly smaller activities of ALT, AST and CK, while AP activity showed no significant difference.

Data regarding the effect of probiotic supplementation on bone metabolism are scarce. No significant differences were observed in the blood levels of calcium (Ca) and inorganic phosphorus (P) in probiotic supplemented kids (Chiofalo et al. 2004) or growing lambs (Abas et al. 2007). Similar findings have been obtained for blood creatinine concentrations as these were not affected by the probiotic supplementation in goats, lambs or rams (Galip 2006, Belewu et al. 2008).

Effect of probiotics on carcass characteristics

Published data regarding effects of probiotic supplementation on carcass characteristics of sheep and goats are inconsistent. Abdelrahman and Hunaiti (2008) have reported increased dressing percentage (DP) by lambs fed diets supplemented with yeast and methionine (cyc-methionine). Similar results were recorded by Belewu and Jimoh (2005) in probiotic-supplemented goats. However, no changes were observed in weights and proportions of carcass cuts in Awassi lambs or Shami goat kids in response to probiotic supplementation (Titi et al. 2008). Likewise, Whitley et al. (2009) reported that carcass weight and weights of fabricated cuts (shoulder, loin, leg, rack, shank and total parts), as well as carcass length, leg circumference, loin eye area and back fat thickness remained unaltered by probiotic supplementation in carcass of goats. Tripathi and Karim (2011) observed that pre-slaughter weight, empty live weight, hot carcass weight, dressing percentage, fore- and hind-quarter weight did not change by yeast culture supplementation to diets of growing lambs. Similarly, half-carcass cut weight (HCW) and carcass composition did not differ among control and yeast fed lambs. However, yeast culture-supplemented lambs had a trend of accelerated carcass composition (% of HCW) attributes of leg, neck and shoulder and breast and fork shank. Moreover, Soren et al. (2013) reported that pre-slaughter weight and hot carcass weight were similar in the control and probiotic supplemented lambs. The wholesale cuts (leg, loin, rack, neck, shoulder, breast, shank) were also similar among the groups with no difference. Similar results were also reported by Kawas et al. (2007b) in lambs fed finishing diet supplemented with either sodium bicarbonate or yeast. In their study, slaughter weight, hot carcass weight and dressed weight were not influenced by yeast supplementation.

Effect of probiotics on milk production and milk composition

Feeding of probiotics was found to improve milk production and milk composition in sheep (Kritas et al. 2006) and goats (Reklewska et al. 2000). Reklewska et al. (2000) found that goats provided daily with 2 g of Yea-Sacc1026 (YC) had a significantly increased milk yield. Their milk protein content increased in relation to the initial level more than in goats fed the standard diet. Similarly, Abd El-Ghani (2004) recorded that yeast culture (*S. cerevisiae*) supplementation to lactating Zaraibi goats had a positive effect on milk yield and contents of milk energy, protein, total solids and solids-non-fat. Milk yield was found to be up to 17.5% higher for goats fed yeast culture. The increase in milk yield after yeast supplementation may be attributed to an increase in DM intake (Robinson and Garrett 1999, Jouany 2006), an increased flow of microbial protein and amino-acids to the duodenum (Erasmus et al. 1992) and the fact that yeast supplementation may act as a source of vitamin B complex (Abdel-Khalek 2003, Helal and Abdel-Rahman 2010). These authors mentioned also that goats fed rations supplied with yeast culture also tended to have increased fat content in milk. The increase in milk fat content in supplemented animals may be attributed to an increment in total bacterial populations and cellulolytic microorganisms in rumen, which improve fibre digestibility and fermentation and consequently increase milk fat content (Wang 2001, Chaucheyras-Daurant et al. 2008). However, milk lactose content was found to decrease with increasing yeast supplementation. On the other hand, Kritas et al. (2006) observed a beneficial effect of *Bacillus licheniformis* and *Bacillus subtilis* (BioPlus 2B) administration in ewes' milk yield, as well as on fat and protein content of milk.

Likewise, Stella et al. (2007) reported that feeding daily 0.2 g of *S. cerevisiae* (Levucell SC20) to dairy goats led to increase in milk yield, whilst milk fat content was lower in the supplemented animals. In contrast, Giger-Reverdin et al. (1996) found a non-significant increase in milk production during early lactation in dairy goats supplemented with *S. cerevisiae*, when given a ration with 25% concentrate, increased fat content, but no effect in protein content were recorded. Finally, Hadjipanayiotou et al. (1997) reported no beneficial effects in milk production, as well as in fat or protein content after feeding yeast to lactating goats in a high concentrate diet.

Effect of probiotics on diarrhea and neonatal mortality

The health of organisms depends to a large degree upon the composition of the intestinal microflora. In this context, the ability of probiotics to modulate the gut microbiota through enhancement of beneficial microbes and reduction of potentially pathogenic bacteria are highly wanted, regardless of animal species (Maragkoudakis et al. 2010).

Probiotic supplementation has been found to reduce diarrhoea in lambs (Lema et al. 2001) and goat kids (Anandan et al. 1999). Probiotics were found to reduce significantly populations of β-haemolytic *E. coli* and O157 *E. coli* (Scharek et al. 2005), decrease the incidence of pre-weaning mortality (Taras et al. 2006) and diarrhoea (Alexopoulos et al. 2004, Scharek et al. 2007). Kritas et al. (2006) have not found evidence of beneficial effects of probiotic supplementation of lambs in reduction of mortality caused by enterotoxigenic strains of *E. coli*. Further, Apas et al. (2010), reported that oral administration of probiotics (*L. reuteri*, *Lactobacillus alimentarius*, *Enterococcus faecium* and *Bifidobacterium bifidum*) in goats has modified gut microbiota, it reduced *Enterobacteria*-like *Salmonella/Shigella* and increased lactic acid bacteria and *Bifidobacterium*. Additionally, probiotic administration was correlated with a 10-folddecrease of faecal putrescine (a neoplasia and bacterial disease marker) and a 60% reduction in mutagen faecal concentration, indicating a positive protective effect of the probiotic mixture. However, Stella et al. (2007) reported that live yeast supplementation to dairy goats did not affect faecal populations of clostridia, enterobacteria or coliforms, it decreased faecal *E. coli* counts and increased lactobacilli counts. The increased numbers of lactobacilli might have led to reduction in numbers of *E. coli*, intestinal pH control (Roa et al. 1997) and receptor competition (Chaucheyras-Durand and Fonty 2002), that way improving stability of the intestinal ecosystem.

Probiotics and immune modulation

Scarce studies are available regarding a possible immunomodulatory role of probiotics in small ruminants. Maragkoudakis et al. (2010) have reported that supplementation of dairy goats with *Lactobacillus plantarum* PCA 236 had no effects on blood IgG, IgM and IgA concentrations.

Conclusions

Probiotics appear as promising feed additives, they are of natural origin and generally regarded as safe for animals. Moreover, they may have the potential to improve production performance and health status of small ruminants. Their effects could be related to enhancing nutrient digestibility, stabilising ruminal ecosystem, stimulating the immune response and increasing milk production in lactating animals.

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