The Journal of Animal & Plant Sciences, 25(1): 2015, Page: 53-59
ISSN: 1018-7081

EFFECTS OF DIETARY SUPPLEMENTATION OF YEAST (*SACCHAROMYCES CEREVISIAE*) CULTURE ON GROWTH PERFORMANCE, BLOOD PARAMETERS, NUTRIENT DIGESTIBILITY AND FECAL FLORA OF DAIRY HEIFERS

S. Ghazanfar[* 1,3], M. I. Anjum[1], A. Azim[1] and I. Ahmed[2]

[1]Animal Nutrition Program, Animal Sciences Institute, National Agricultural Research Centre, Park Road, Islamabad-45500, Pakistan
[2] National Institute for Genomics & Advanced Biotechnology, National Agricultural Research Centre, Park Road, Islamabad-45500, Pakistan
[3]Department of Microbiology, Quaid-i-Azam University, Islamabad 45320, Pakistan
[*]Corresponding author: shakira_akmal@yahoo.com

ABSTRACT

In a 120 day feeding trial, 8 cattle heifers (87±5 kg and 6 to 7 months) were randomly divided into two equal groups of four animals each (control and supplemented). During the trial, heifers in both the groups were offered National Research Council (NRC) recommended diet with or without yeast supplementation. The diet was formulated by adapting the small dairy breeds heifers' nutrients requirements for growth rate of 0.6 kg/day considering body weight of 100 kg. The heifers in the supplemented group fed with *Saccharomyces cerevisiae;* Yea-Sac[1026] (Alltech Inc., Nicholasville, KY), 5 g/animal/day. The effects of *Saccharomyces cerevisiae* on growth performance, blood parameters, nutrient digestibility, fecal coliform and Lactobacillus were studied. Average dry matter intake (DMI) was not different among both groups; whereas average daily weight gain was higher ($p<0.05$) in supplemented compared with control group.The digestibility of dry matter, organic matter, crude protein, neuter detergent fibre and acid detergent fibre was higher ($p<0.05$) in yeast-supplemented group compared with control group. Yeast-supplementation increased ($p<0.05$) the eosinophils and hemoglobin levels and erythrocytes and leukocytes counts.The average fecal population of Lactobacillus was greater ($p<0.05$) with yeast-supplemented than in control group. It is therefore concluded that incorporation of *Saccharomyces cerevisiae* in the NRC recommended diet improved growth and health performance of dairy cattle heifers.

Key words: *Sahiwal* heifers, *Saccharomyces cerevisiae*, Growth rate, Digestibility, Coliform, Lactobacillus.

INTRODUCTION

Feed additives can improves gut health of the animals, which results in increased digestion rate and better growth performance (Frizzo *et al*., 2010; Kawakami *et al.*, 2010; Frizzo *et al.*, 2011). Many microbial species have been approved as feed additives; among them the fungi/yeast culture (e.g. *Saccharomyces cerevisiae*) has been found to exert a positive effect on the ruminant's production. Mode of action of yeast depends on the rumen microbial population. Yeast cells contain different vitamins, enzymes and some unidentified cofactors that may improve the microbial activity and growth rate in rumen (Dawson *et al.*, 1992). Many researchers reported that yeast culture improved feed intake (Robinson and Erasmus, 2009; Ayad *et al*., 2013); feed conversion efficiency, growth rate (Lascano *et al.*, 2009) and nutrient digestibility (Wohlt *et al.*, 1991) in cost effective manners (Hutjens, 2003). Yeast also has positive effects on blood hematology resulting in improvement in health status of animals (Agazzi *et al.*, 2014). Lascano *et al* (2012) and Lesmeister *et al* (2004) reported that yeast supplementation increased the hemicelluloses degradability and some important nutrient digestibility. The addition of yeast culture has many positive effects in the absorption of some minerals (Cole *et al*., 1992) and improves the metabolic heath of animals (Dolezal *et al.*, 2011). Many heifers can get digestive disturbance during their growing period which leads to imbalance bacterial flora (Bayatkouhsar *et al.*, 2013). Supplementation of microbial feed additive as a tool to maintain the microbial balance of intestine, prevents diarrhea (Abu *et al.*, 1996; Galvao *et al.*, 2005; Timmerman *et al*., 2005), and improved fecal bacterial flora of the ruminants (Kawakami, 2010). It is also effective in improving resistance to colonization with pathogen and thus it results to improve health of animals (Jatkauskas and Vrotniakiene 2010). Since it is difficult to obtain intestinal samples for microbial analyses, enumeration of fecal microbial flora has been used as an indirect method of determining bacteria inhabiting in the intestinal tract (Gilliland *et al.*, 1980). The fecal flora is assumed to represent only the luminal flora and not that associated with mucosal epithelial surfaces and is assumed to vary with different types of diets (Ellinger *et al.,* 1980). Few research reports are available regarding microbial characterization, diversity and other prospective applications of cattle fecal material (Yokoyama *et al*., 2007). In Pakistan most dairy heifers are reared on conventional feeding system resulting in poor growth rate (Anjum *et al.*, 2012). No information on use of probiotic is available in Pakistan and there is strong need of research work that can highlight the effect of probiotic on the better growth rate. Keeping in view, the present study was undertaken to analyze the effect of yeast culture on growth performance, nutrient digestibility, blood parameters and numbers of fecal coliforms and lactobacillus of dairy cattle heifers.

MATERIALS AND METHODS

Experimental layout: The study was conducted at Animal Nutrition Program, National Agricultural Research Centre, Islamabad, Pakistan. Eight dairy heifers of *Sahiwal* breed at 6 ± 1 months of age with 87 ± 10 kg body weight were used in this experiment and were divided into two equal groups, i.e., four heifers on control diet without yeast addition (control group) and other four heifers fed control diet with yeast addition (yeast supplemented group).

Diet preparation: Experimental diets were prepared at Feed Technology Unit, Animal Nutrition Program, National Agricultural Research Centre (NARC), Islamabad, Pakistan to meet the NRC (2001) small dairy-breeds heifer's nutrient requirements (Table1) During this study heifers in both groups were offered same diet to meet the 100 % metabolizable energy (ME) of NRC requirement of Jersey heifer. The heifers of supplemented group received yeast culture (*Saccharomyces cerevisiae*, Yea-Sac[1026], Alltech Inc., Nicholasville, KY) of 5 g/animal/day.

Growth performance: Feed intake was recorded on daily basis by weighing feed offered to and refused by the heifers using an electroic scale after feed and water resrtction for 16 hours (Anjum *et al*., 2014). Feed efficiency was calculated as amount of kilograms of feed intake per kilogram of weight gain. This experiment lasted for 120 days.

Digestibility Trial: During last two weeks of experiment digestibility trial was conducted. Three heifers of almost similar body weight from each group were placed in individual digestibility pens (measuring 4ft×9 ft) having facility to collect feces and urine. For 5 consecutive days, feces were weighed and mixed daily, and a representative sample (2%) was taken, stored at −20°C, and subsequently thawed, dried at 55°C for 48 h, and ground through a 1-mm screen Wiley mill for chemical analysis. For proximate analysis, methods describe by AOAC, (1990); for neutral detergent fibre (NDF) and acid detergent fibre (ADF) analysis, methods describes by Van Soest *et al*. (1991).

Blood parameters: Blood was taken after 120 days of the experiment. Blood samples were collected from the jugular vein of all the control and yeast-supplemented herifers. Blood samples were put into 5 ml heparinised venoject® EDTA (K^3) tubes and mixed thoroughly for red blood cell, white blood cell, haemoglobin, haematocrit, poly lymphocyte, monocyte and eosinophils determination. These hematological parameters were estimated by using Beckman Coulter® AcT Diff™ Haematology Analyzer at National Veterinary Laboratories, Islamabad, Pakistan by using method of Benjamin (1985).

Fecal bacterial counts: Fecal grab samples were collected from 8 heifers at 0, 30, 60, 90 and 120 days of the experiment direct from rectum in sterile plastics bags. Fecal sample 1 gm was mixed in trypticase salt solution (TSS) and allowed to vortex for 15-20 minutes at 40Hz and then shifted in reagent bottle in sterile conditions. Man, Ragosa and Sharpe agar (MRS) (Oxoid, Basingstoke, UK) was used for lactobacillus and macconkey agar (oxiod) media was used for coliform isolation. The macconkey agar and MRS agar plates were incubated aerobically at 37 ^{o}C for 48 h. Colony counts were done manually with numbers over 300 designated as too numerous to count (TNTC).

Statistical analysis: Data collected were analyzed with a linear model using student t-test (Steel *et al*., 1997). Data are given as means plus minus standard error of the means.

RESULTS AND DISCUSSION

Growth performance: The results of the DMI, average daily gain, FCR and economic efficiency are given in the Table 2. The average daily weight gain (kg/d) of the heifers was higher ($P < 0.05$) in supplemented group than control group. The higher growth rate might be due to increased microbial protein flow escaping from the rumen and an enhanced supply of different amino acids entering the small intestine. Similar results were reported by Kumar and Ramana (2008) who observed higher daily weight gain in animals when fed using the diets supplemented with yeast culture. Heifers of both groups almost consumed similar DMI. The FCR improved ($P>0.05$) in supplemented group and was reflected in the increased ($P<0.05$) average daily gain in supplemented group. These results are in agreement with the findings of Kumar and Ramana (2008), who reported improved FCR with yeast culture in the diets. The results of skeletal measurements are shown in the Table 2. Our result showed that skeletal measurements were not changed ($P>0.05$) by addition of yeast culture. These results are in agreement with the findings of Zanton and Heinrichs (2007), and Lascano *et al.* (2009), who found no change in the structural measurements in the dairy heifers fed diet with yeast culture supplementation.

Digestibility trail: The results of nutrients digestibilities are presented in Table 3. The results revealed that

Saccharomyces cerevisiae supplementation increased ($P<0.05$) dry matter (DM), organic matter (OM), crude protein (CP), NDF and AFD digestibility and is comparable to the previous studies (Lascano *et al.*, 2012; Lascano and Heinrichs, 2009). That improved nutrient digestibility might be due to increased cellulose degrading microbial biomass population inside rumen. NDF, ADF and cellulose digestibility were also affected ($P<0.05$) by yeast supplementation in these studies. The increased digestibility can be due to stable rumen pH and removal of oxygen from the rumen in the yeast supplemented group. That stable rumen pH provides better environment for growth of rumen microbes, especially cellulose degrading bacteria and fungi. At the same time the anaerobic condition inside rumen also helped in better growth of fibrolytic microbial biomass. Consequently, these microbial species helped in better fibre digestion. The stable pH also enhanced microbial protein synthesis in the rumen. Our data support the findings of Lascano *et al.* (2012) who reported that NDF and ADF digestibility was improved in yeast supplemented group. In contract to our result, few researchers also reported no effect of yeast on the nutrient digestibility (Tripathi and Karim 2010). The variation in nutrient digestibility may be due to the nature and quality of diet fed to animals (Desnoyers *et al.,* 2009). Moreover, yeast can utilize part of free sugar in the rumen and limit a fermentation shift due to rapid degradation of these compounds.

Blood parameters: The values for hematological parameters are presented in Table 4. All values were within the normal physiological range, which is a sign of good health. Supplementation with yeast culture increased ($p<0.05$) the eosinophils and hemoglobin levels. Counts for erythrocytes and leukocytes were also higher ($p<0.05$) in the supplemented groups than heifers in control group. These results are in agreement with the finding of Agazzi *et al.* (2014) and Heinrichs *et al.* (2003) who reported that hematological parameters was affected by probiotic addition.

Fecal bacterial counts: The average faecal lactobacilli and coliform count of dairy heifers fed on live yeast as determined at 0, 30, 60, 90 and 120 days of the experiment are given in Table-5. There was a variation in count of coliform and lactobacillus in faeces at 0 day in both supplemented and control groups. Because heifers were obtained from different locations, variation may be due to feeding behavior and different environmental condition. Environmental circumstances and host factors usually influence the multifarious composition of the gastro-intestinal micro-flora (Vlkova *et al.*, 2006). Once heifers were fed on uniform feeding scheme and gave same management, variation was reduced (Table 4). There were significant ($p<0.05$) effects of yeast supplementation on the numbers of lactobacillus in heifers' fecal samples in 60, 90 and 120 days. Yeast supplementation increased ($p<0.05$) the numbers of fecal lactobacillus with passage of time. Considering the overall growth impact from day 0 up till day 120, the CFU/g values demonstrated an increased lactobacillus growth trend in both groups. The increased growth was higher ($p<0.05$) in supplemented group than control group suggested that the yeast supplementation has a capability to improve gut microbial flora and reduce the diarrhea. Same results were reported by Kawakami *et al.* (2010).The coliform were similar and no ($p>0.05$) difference was seen in both groups during 60, 90 and 120 days of the experiment. Coliform count was higher ($p<0.05$) in control group than supplemented group at day 30. That might be due to the digestive problem in control group during these days. Two heifers of control group got diarrhea which may leads to have a significance difference ($p<0.05$) in both group during 30 days of experiment. With passage of time the diarrheal condition gone and the number of fecal coliform bacteria decreased in supplemented group than control group. Our results indicated that the number of coliform in supplemented and control group were higher on day 0 compared with day 120. The number of lactobacilli in feces is a widely used index for estimation of intestinal flora (Fuller, 1989). Lactobacillus as a beneficial bacterial species normally associated with balance normal in the gut flora of heifers. Increase in lactobacilli concentration can show a normal occurrence in the development of intestinal flora of heifers. In present studies, the results showed that yeast had positive effect on lactobacilli population with respect to age. The present results are agreed with Agazzi *et al.* (2014); Bayatkouhsar *et al*. (2013); Ayad *et al*. 2013 and Kawakami *et al.* (2010) who reported that fecal population of lactobacilli become greater with probiotic supplementation. However, the result of previous reports was controversial about the effect of probiotics on the number of fecal coliform of ruminants. Some researchers reported that feeding probiotics significantly decreased fecal coliform, as reported by Agarwal *et al.* (2002) but Ellinger *et al.* (1980) reported no effect of probiotic on coliform. Many other studies perversely indicated that coliform rage could increased when some disorder occur (Ellinger *et al.*, 1980; Gilliland *et al.*, 1980; Jenny *et al.,* 1991). During these studies diarrheal condition were not seen throughout the experimental period and may account for the absence of significant changes in coliform numbers. The number of coliform is higher than that of lactobacilli in the animal's suffering from diarrhea, but lower in healthy animals (Signorini, 2012), which suggested that coliform numbers L: C ratio could be used as indicator for estimation of intestinal microbial flora associated with diarrhea. The L: C ratio hypothesis also confirm in these studies. The improved gut health condition in our study was confirmed by lower incidence of diarrhea in supplemented group as compared to control group. Similar result were reported by Kawakami *et al.* (2010) who reported that feeding yeast and lactic acid bacteria lower the incidence of diarrhea by improving fecal flora.

Table 1. Ingredients and nutrient composition and cost of total mix ration (TMR).

Item	Control diet
Chemical composition (% DM)	
Crude protein	15.23
Neutral detergent fibre	27.88
Acid detergent fibre	18.04
Calcium	0.69
Total phosphorous	0.57
ME (Mcal/kg)	2.45
Ingredients (%)	
Maize oil cake	17.00
Cottonseed meal	13.00
Sunflower meal	1.00
Canola meal	6.00
Rice polish	6.00
Wheat bran	7.00
Corn gluten feed	4.00
Corn grains	11.00
Vegetable oil	2.00
Wheat straw	24.00
Cane molasses	6.00
Urea	0.50
Dicalcium phosphate	1.00
Limestone power	0.50
Sodium chloride	0.50
Minerals premix	0.50

Table 2. Effect of dietary yeast supplementation on growth characteristics and structural measurements in dairy cattle heifers

Parameters	Treatments		p-value
	Control	Yeast-supplemented	
Growth characteristics			
Average initial body weight, kg	88.53±5.22	87.56±5.52	0.421
Average final body weight, kg	160.84±2.10	172.38±3.39	0.072
Average daily body weight gain, kg	0.60±0.04	0.72±0.02	0.037
Average daily DM intake, kg	3.72±0.03	3.81±0.06	0.416
Feed conversion ratio (kg feed/kg gain)	6.19±0.49	5.41±0.52	0.056
Structural measurements (cm)			
Body height			
Initial	41.84±1.28	42.27±1.12	0.700
Final	84.13±1.60	78.34±1.14	0.107
Body length			
Initial	39.42±0.92	40.22±1.20	0.500
Final	79.36±1.83	78.53±2.12	0.750
Heart girth			
Initial	41.53±1.29	46.23±1.59	0.106
Final	86.38 ±1.56	89.13±.0.62	0.259
Top line			
Initial	54.51±1.31	51.98±0.50	0.345
Final	90.56±0.304	89.12±3.50	0.299

Table 3. Effect of dietary yeast supplementation on nutrient digestibility (Means ± SEM) in dairy cattle heifers

Parameters	Treatments		p-value
	Control	Yeast-supplemented	
Dry matter	55.52±1.10	60.25±0.82	0.041
Organic matter	60.31±1.22	64.48±.36	0.047
Crude protein	57.87±0.71	61.02±0.65	0.049
Crude fibre	54.28±1.40	57.51±1.08	0.043
Neutral detergent fibre	58.63±0.46	61.79±0.59	0.036
Acid detergent fibre	53.08±0.43	55.45±0.48	0.017

Table 4. Effect of dietary yeast supplementation on blood parameters (Means ± SEM) in dairy cattle heifers

Parameters	Treatments		p-value
	Control	Yeast-supplemented	
Haematological values			
Erytrocytic count, x10 6/µl	8.35±0.67	11.54±1.33	0.043
Total leukocytic count, x10 3/ml	8.45±0.54	10.37±0.85	0.048
Packed cell volume, %	28.99±1.53	30.84±2.55	0.517
Hemoglobin, g/dl	9.76±0.55	13.26±1.20	0.045
Lymphocytes, %	49.70±2.09	52.81±2.98	0.397
Monocytes, %	4.37±0.30	4.70±2.21	0.396
Eosinophils, %	4.85±0.46	6.94±0.75	0.017

Table 5. Effect of dietary yeast supplementation on bacteria counts (Log10 cfu/g ± SD) in fecal samples of dairy cattle heifers

Days of age	Treatments		p-value
	Control	Yeast-supplemented	
Lactobacilli			
0	4.22±0.94	4.61±0.86	0.607
30	4.63±0.38	5.44±0.13	0.178
60	4.86±0.43	6.31±0.14	0.039
90	5.28±0.30	6.47±0.10	0.030
120	5.53±0.49	6.98±0.28	0.042
Coliforms			
0	3.06±0.32	3.03±0.27	0.959
30	4.11±0.27	3.17±0.25	0.001
60	3.81±0.25	3.14±0.19	0.215
90	2.59±0.32	2.56±0.19	0.881
120	2.79±0.30	3.05±0.52	0.521

Conclusion: Thus, it can be concluded that dietary addition of yeast (*Saccharomyces cerevisiae* (Yea-Sacc $^{1026\ TM}$ strain, Alltech inc., Nicholasville, KY) at the rate of 5g/animal/day improved ADG, FCR, better nutrient digestibility, immunity and health in growing dairy cattle heifers without any adverse effects. However further research with more numbers of heifers would be needed for better understanding the effect of yeast supplementation in high energy diet.

REFERENCES

Abu, T., H. M. Al-Saiady, and A. H. Keir (1996). Evaluation of diet containing lactobacilli on performance, fecal coliform, and lactobacilli of young dairy calves. Anim. Feed Sci. Technol. 57(1): 39-49.

Anjum, M. I., A. Azim, M. A Jabbar, and I. H. Mirza (2012), Age and weight at puberty in Nilli Ravi buffalo heifers reared on three dietary energy restriction periods followed by compensatory growth. Pakistan Vet.J. 32 (3): 367-371

Anjum, M. I., I. H. Mirza, I. Begum, and S. Javaid (2014), Effcets of varying levels of calcium in rations for buffalo (*Bubalus Bubalus*) calves on growth rate and nutrient digestibility. J. Anim. Plant Sci. 24 (1): 24-29.

Agarwal, N., D. N. Kamra, L. C. Chaudhary, I. Agarwal, A. Sahoo, and N. N. Pathak (2002), Microbial status and rumen enzymes profile of crossbred claves fed on different microbial feed additives. Letters in Applied Microbiology 23(5): 329-336.

Agazzi, A., E. Tirloni, S. Stella, S. Maroccolo, B. Ripamonti, C. Bersani, J. M. Caputo, V. Dellorto, N. Rota, and G. Savoini (2014), Effects of species-specific probiotic addition to milk replacer on calf health and performance during the first month of life. Ann. Anim. Sci.14(1): 101-115

AOAC. (1990), Official Methods of Analysis, 15th ed., Association of Official Analytical Chemist, Washington, D.C, USA.

Ayad, M. A., B. Benallou, M. S. Saim, M. A. Smadi, and T. Meziane (2013), Impact of Feeding Yeast Culture on Milk Yield, Milk Components, and Blood Components in Algerian Dairy Herds. J. Veterinar. Sci. Technol. 4(135): 2

Bayatkouhsar, J., A. M. Tahmasebi, A. A. Naserian, R. R. Mokarram, and R. Valizadeh (2013), Effects of supplementation of lactic acid bacteria on growth performance, blood metabolites and fecal coliform and lactobacilli of young dairy calves. Anim. Feed Sci. Technol. 186(1): 1-11.

Benjamin, M. M. (1985), Outline of veterinary clinical pathology. 3rd edition. The Lowa State Univ. press. Ames. USA.

Cole, N. A., C. W. Purdy, and D. P. Hutcheson, (1992), Influences of yeast culture on feeder calves and lambs. J. Anim. Sci. 70: 1682-1690.

Dawson, K. A. (1992), Current and future role of yeast culture in animal production: A review of research over the past six years. In: Lyons TP (Ed.): Proceedings of Alltech's 8th annual symposium, Nicholasville, Kentucky. 1-23

Desnoyers, M., S. G. Reverdin, G. Bertin, C. D. Ponter, and D. Sauvant (2009), Meta-analysis of the influence of *Saccharomyces cerevisiae* supplementation on ruminal parameters and milk production of ruminants. J. Dairy Sci. 92(4): 1620-1632.

Dolezal, P., J. Dolezal, K. Szwedziak, J. Dvoracek, L. Zeman, M. Tukiendorf, and Z. Havlicek (2012), Use of Yeast Culture in the TMR of Dairy

Holstein Cows. Iranian J. Applied Anim. Sci. 2(1): 51-56.

Ellinger, D. K., L. D. Muller, and P. J. Glantz (1980), Influences of feeding fermented colostrum and Lactobacillus acidophilus on fecal flora of dairy calves. J. Dairy Sci. 63: 478-482.

Frizzo, L. S., L. P. Soto, M. V. Zbrun, E. Bertozzi, G. Sequeira, R. R. Armesto, and M. R. Rosmini (2010), Lactic acid bacteria to improve growth performance in youn.g calves fed milk replacer and spray-dried whey powder. Anim. Feed Sci. Technol. 157 (3): 159-167.

Frizzo, L. S., L. P. Soto, E. Bertozzi, M.V. Zbrun, M.L. Signorini, G.R. Sequeira, A. Rodriguez, and M. R. Rosmini (2011), Intestinal populations of Lactobacilli and coliforms after in vivo Salmonella Dublin challenge and their relationship with microbial translocation in calves supplemented with lactic acid bacteria and lactose. Anim. Feed Sci. Technol. 170 (1): 12-20.

Fuller, R. (1989), Probiotics in man and animals. J. Appl. Bacteriol. 66: 365–378.

Galvao, K. N., J. E. Santos, A. Coscioni, M. Villasenor, W. M. Sischo, and A. C. Berge (2005), Effect of feeding live yeast products to calves with failure of passive transfer on performance and patterns of antibiotic resistance in fecal Escherichia coli. Reprod. Nut. Dev. 45: 427-440

Gilliland, S. E., B. B. Bruce, L. J. Bush, and T. E. Staley (1980), Comparison of two strains of Lactobacillus acidophilus as dietary adjuncts for young calves. J. Dairy Sci. 63: 964.

Heinrichs, A. J., C.M. Jones, and B. S Heinrichs (2003), Effects of mannan oligosaccharide or antibiotics in neonatal diets on health and growth of dairy calves. J. Dairy Sci. 86(12): 4064-4069.

Hutjens, M. F. (2003). Economics of feed additives. Penn State Dairy Cattle Nutrition Workshop. dasweb.psu.edu/pdf/hutjens1.pdf.

Jatkauskas, J., and V. Vrotniakiene (2010), Effects of probiotic dietary supplementation on diarrhoea patterns, faecal microbiota and performance of early weaned calves. Veterinarni. Medicina. 55(10): 494-503

Jenny, B. F., H. J. Vandijk, and J. A. Collins (1991), Performance and fecal flora of claves fed a Bcillus subsitis concentrate.J. Dairy Sci. 74: 1988-1973.

Kawakami, S. I., T. Yamada, N. Nakanishi, and Y. Cai (2010), Feeding of lactic acid bacteria and yeast on growth and diarrhea of Holstein calves. J. Anim. and Vet. Advances. 9 (7), 1112-1114.

Kumar, M. K., and D. B. V. Ramana (2008), Effect of supplementation of yeast culture to calves fed with complete diet. India Vet. J. 85: 667-669.

Lascano, G. J., A. J. Heinrichs, and J.M. Tricarico (2012), Substitution of starch by soluble fiber and Saccharomyces cerevisiae dose response on nutrient digestion and blood metabolites for precision-fed dairy heifers. J. Dairy Sci. 95(6): 3298-3309.

Lascano G. J., G. I. Zanton, M. F. Suarez-Mena, and A. J. Heinrichs (2009), Effect of limit feeding high- and low-concentrate diets with Saccharomyces cerevisiae on digestibility and on dairy heifer growth and first-lactation performance. J. Dairy Sci. 92(10): 5100-10

Lesmeister, K. E., A. J. Heinrichs, and M. T. Gabler (2004), Effects of supplemental yeast (Saccharomyces cerevisiae) culture on rumen development, growth characteristics, and blood parameters in neonatal dairy calves. J. Dairy Sci. 87: 1832-1839.

NRC. (2001), Nutrient Requirements of Dairy Cattle. 7th rev. ed. Acad. Sci. Natl.,Washington, DC.

Robinson, P. H., and L. J. Erasmus (2009), Effects of analyzable diet components on responses of lactating dairy cows to Saccharomyces cerevisiae based yeast products: A systematic review of the literature. Anim. Feed Sci. Technol. 149(3): 185-198.

Signorini, M. L., L. P. Soto, M. V. Zbrun, G. J. Sequeira, M. R. Rosmini, and L. S. Frizzo (2012), Impact of probiotic administration on the health and fecal microbiota of young calves: A meta-analysis of randomized controlled trials of lactic acid bacteria. Research in Veterin. Sci. 93(1): 250-258.

Steel, R. G. D., J. H. Torrie, and D. A. Dickey (1997), Principles and procedures of statistics. A biochemical approach 3rd. Ed. McGraw Hill Book Co. Inc. NY, USA

Timmerman, H. M., L. Mulder, H. Everts, D. C. Espen, W. E. Van Der, G. Klaassen, S. M. G. Rouwers, R. Harteminik, F. M. Rombouts, and A. C Beynen (2005), Health and growth of veal calves fed milk replacers with or without probiotics. J. Dairy Sci. 88: 2154–2165.

Tripathi, M. K., and S. A. Karim (2010), Effect of individual and mixed live yeast culture feeding on growth performance, nutrient utilization and microbial crude protein synthesis in lambs. Anim. Feed Sci. Technol. 155(2): 163-171.

Van Soest, P. J., J. D. Robertson, and B. A. Lewis (1991), Methods for dietary fiber, neutral detergent fiber and non-starch polysaccharide in relation to animal nutrition. J. Dairy Sci. 74: 3583-3597.

Vlkova, E., I. Trojanova, and V. Rada (2006), Distribution of bifidobacteria in the

gastrointestinal tract of calves. Folia Microbiologica. 51**:** 325-328.
Wohlt, J. E., A. D. Finkelstein, and C. H. Chung (1991), Yeast culture to improve intake, nutrient digestibility and performance by dairy cattle during early lactation. J. Dairy Sci. 74: 1395-1400
Yokoyama, H., M. Waki, A. Ogino, H. Ohmori, and Y. Tanaka (2007), Hydrogen fermentation properties of undiluted cow dung. J. Bioscience and Bioengineering. 104**:** 82-85
Zanton, G. I., and A. J. Heinrichs (2007), The effects of controlled feeding of a high-forage or high-concentrate ration on heifer growth and first-lactation milk production. J. Dairy Sci. 90 (7): 3388-3396.