Journal of Animal and Feed Sciences, 29, 2020, 115–124 https://doi.org/10.22358/jafs/124040/2020
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Performance and health of young rosé veal calves supplemented with yeast (*Saccharomyces cerevisiae*) and a postbiotic from *Lactobacillus acidophilus*

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KEY WORDS: probiotic, *Saccharomyces cerevisiae*, *Lactobacillus acidophilus*, calves, performance, health

Received: 20 November 2019
Revised: 22 April 2020
Accepted: 8 June 2020

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ABSTRACT. The aim of the study was to investigate the effects of a product 'ZooLac Bovimix', containing yeast (*Saccharomyces cerevisiae*) and a postbiotic product from *Lactobacillus acidophilus*, on the performance and health of young rosé veal calves. In total 120, mainly Holsteins, bull calves arrived at a Danish rosé veal production over a period of three months. On arrival, the calves were either allocated to a control diet (CON) (25.3 ± 1.3 days and 55.1 ± 1.3 kg) or a diet with the yeast/*lactobaccilus* (PRO) (25.3 ± 1.0 days and 55.3 ± 0.8 kg) for six weeks. PRO calves received the yeast/*lactobacillus* product in the milk replacer and the concentrate. The product constituted 0.09% of dry matter (DM) in the concentrate and 1.5% of DM in the milk replacer. All calves were fed milk replacer for four weeks and had access to concentrates for all six weeks. Manure was sampled twice, around days 14–17 and 28–31, and blood was sampled on days 3 and 28–35. The growth performance was significantly higher for PRO calves, while the product did not affect the number of veterinary treatments and DM content in the manure. Serum metabolites were similar between the treatments at the first sampling. However, the concentrations of total protein and immunoglobulin G were significantly higher in the serum from PRO calves at the second sampling. Thus, supplementation of the yeast/*lactobacillus* product in the diet of young rosé veal calves did not affect the overall health but had a positive effect on growth performance.

Introduction

The bull calf faces several challenges, e.g., new environment and pen mates after it is shipped from the dairy herd to the veal calf production at 2 to 6 weeks of age. Commingling increases the infection level in the herd and stress level of the calf (O'Connor et al., 2005). Stress triggers the secretion of adrenal corticosteroids, which may suppress immune function (Seymour et al., 1995). Additionally, the young calf is especially vulnerable to diseases in the period right after shipping as the adaptive immune system has not yet acquired an immunological memory. Thus, the calf is mainly protected by the innate immune system and the passively transferred immunity from colostrum (Galvao et al., 2005), but blood immunoglobulin G (IgG) levels usually decline during the first weeks of life (Roodposhti and Najafgholi, 2012). Both prior and around the time of weaning, enteric diseases and subsequent problems dehydration are major health issues affecting these calves (Magalhaes et al., 2008). Treatments of enteric diseases are often related to increased use of antibiotics, which can constitute a threat to human health as it increases the risk of developing multiple antibiotic-resistant bacteria (Baynes et al., 2016).

Feed additives, such as probiotics and postbiotics, are preventive strategies that potentially can lower the use of antibiotics in animal production (Signorini et al., 2012). Probiotics are live non-pathogenic microorganisms that have the ability to improve the microbial balance in the gastrointestinal tract (GIT) of the host. In general, they act through molecular and cellular mechanisms by disturbing the adhesion of pathogens, enhancing innate immunity, decreasing pathogen-induced inflammation, promoting intestinal epithelial cell survival, barrier function, and protective responses (Williams, 2010). Yeast products of *Saccharomyces cerevisiae* (SC) origin are a type of probiotics commonly included in the diets of production animals to enhance the health and performance (Jensen et al., 2008). Despite the well-described effects of yeast in more adult cattle on rumen digestion with roughage-rich diets, it was recently suggested that yeast products also have beneficial effects on the performance of young calves fed milk and concentrate (Alugongo et al., 2017a). Several products based on different strains of SC are available on the market. Based on the content of active ingredient, the products can be classified as live SC or as SC fermentation products (Robinson and Erasmus, 2009; Poppy et al., 2012). However, the two types of products might not have significant differences in their mode of action in the GIT (Lynch and Martin, 2002).

Some probiotics are able to secrete soluble factors that are termed postbiotics. They can be obtained from cell-free culture supernatants of several probiotics. One of the most studied types of postbiotics is from *Lactobacilli* strains (Cicenia et al., 2014). The product 'ZooLac' (ChemVet, Silkeborg, Denmark) is a postbiotic product available on the market. The product contains killed, but intact, whole lactic acid bacterial bodies, lactic acids and lactic acid salts. The product is suggested to work as a biofilm in the intestine and thereby prevent adhesion of pathogens.

The aim of the current study was to investigate the effects of a yeast/postbiotic product, containing live yeast (*Saccharomyces cerevisiae*) and a postbiotic product from *Lactobacillus acidophilus*, on the performance and health of young rosé veal calves after six weeks of supplementation during the weaning-transition phase.

It was hypothesized that the supplementation of the yeast/*lactobacillus* product would improve the growth performance and health of these young rosé veal calves.

Material and methods

Animal, housing and diets

A production trial was conducted at a Danish rosé veal calf production in Southern Denmark. All procedures involving animals were conducted in accordance with the guidelines of the Danish Ministry of Justice with respect to animal experimentation and care of animals under study (The Danish Ministry of Justice, 2014, LBK no. 474). The Danish Animal Experiments Inspectorate under the Danish Veterinary and Food Administration was consulted for guidance on required permissions and approved the project activities in writing without requiring further formal application and approval process.

On arrival at the veal calf production from the dairy herds, 120 bull calves were randomly allocated to either a control diet (CON; 60 calves) or a diet with a product containing probiotic SC and postbiotic from *Lactobacillus acidophilus* in the milk replacer (MR) and concentrate (PRO; 60 calves). The calves arrived in five blocks of 24 calves from November 2018 to January 2019. Before departure from the dairy herds, the calves were weighed. As the production trial was carried out in a real production setup, it was not possible to influence the breed of the calves. Thus, 110 of the 120 calves were purebred Holstein and 10 were Holstein × Beef crossbreds. The calves originated from 21 different dairy herds. On arrival at the veal calf production, the calves were sorted based on their body weight (BW) before departure, breed and dairy herd of origin. Hence, the calves in each pen in the hutch, which contained two pens, had a similar average BW, proportion of crossbreds and calves per dairy herd. The average age and BW on arrival were 25.3 ± 1.3 days and 55.1 ±1.3 kg for the CON group and 25.3 ± 1.0 days and 55.3 ± 0.8 kg for the PRO group.

It was not possible to obtain information regarding vaccination protocols in the dairy herds or birth weight and colostrum feeding of the calves purchased for the production trial. However, as the calves were sorted by the herd of origin, the effects of these factors were taken into account.

The straw-bedded hutches contained two pens with six calves per pen, separated by an open wall. The pens were provided with one trough for concentrate and six bawls where the calves were offered MR and water. Roughage was offered in a shared hay-rack between the two pens in the hutches.

It was not possible to change the design of the hutches in the veal calf production so the treatment groups did not share the hay-rack. The day following arrival, the calves were weighed again. The initial weight was calculated as a mean of the BW on arrival and BW the following day.

The calves were fed 6 l of MR per day (60% skimmed milk powder, 135 g MR/l) for approximately four weeks and concentrate and roughage for the whole six-week period. Besides skimmed milk powder, the MR was based on whey powder, vegetable oil (Palm/Coco 60/40), and hydrolysed wheat protein concentrate. The calves were fed MR twice a day, around 8:00 and 16:00. The crude protein (CP) contents were 23.7 and 22.9% and the contents of fat were 18.2 and 17.9% for CON and PRO diets, respectively. The energy content of the MR was similar between the treatments (11.1 vs 11.0 MJ NE/kg DM for CON and PRO diets, respectively).

Calves allocated to CON group were fed MR without any additives, while the MR fed to the calves allocated to PRO group was supplemented with 2 g of the yeast/*lactobacillus* 'ZooLac Bovimix Milk' (ChemVet; Silkeborg, Denmark) (7.7 MJ NE/kg DM, 2.83% CP) per 135 g of MR. 'ZooLac Bovimix Milk' contained 55% of 'Actisaf Powder' (living SC cells: Lesaffre proprietary strain: NCYC Sc 47/CNCM I-4407) and 45% of 'ZooLac' (ChemVet; Silkeborg, Denmark). The colony forming unit (CFU) of SC in the MR was $7.88 \pm 0.18 \times 10^9$ CFU per kg (mean ± SEM). 'ZooLac' is thermostabilized lactic acid bacteria, lactic acids and lactic acid salts. The product is produced by fermentation of vinasse by a special strain of *Lactobacillus acidophilus* which after fermentation is thermostabilized by thermal treatments, followed by lyophilisation. The bacterial bodies still are appearing intact, even though they are not alive. The number of thermostabilized *Lactobacillus acidophilus* was 3.1×10^7 per kg MR.

When the smallest calves in the group of six calves weighed approximately 75 kg, the group was gradually weaned by removing the afternoon MR feeding for one week. Instead, the calves were supplied with glucose-water in the weaning week. After the week, the calves were completely weaned.

The ingredients and chemical composition of the concentrates are shown in Table 1. The PRO concentrate contained the yeast/*lactobacillus* 'ZooLac Bovimix' (ChemVet; Silkeborg, Denmark). The difference between 'ZooLac Bovimix' and 'ZooLac Bovimix Milk' is that 'ZooLac Bovimix' contains the yeast product 'Actisaf HR$^+$', which is more thermostable in comparison to 'Actisaf Powder'. This is needed due to the heat associated with the pelleting process. The CFU of SC in the pelleted PRO concentrate was $2.38 \pm 0.13 \times 10^9$ CFU per kg (mean ± SEM). The PRO concentrate also contained 0.05% 'ZooLac'. The ratio between SC and 'ZooLac' in the concentrate was the same as in the MR.

Table 1. Ingredient composition and nutrient content of control concentrate (CON) and concentrate supplemented with a product containing yeast (*Saccharomyces cerevisiae*) and postbiotic from *Lactobacillus acidophilus* (PRO)

Indices	CON	PRO
Ingredients, %		
barley	27.00	27.07
wheat	13.30	13.53
sugar beet	12.92	13.48
maize grain	12.00	12.00
soybean meals	9.54	9.02
rapeseed cakes	6.67	9.00
sunflower meals	6.00	6.00
distillers' grains	4.13	5.00
rapeseed meals	2.33	–
limestone	2.17	1.86
fat	1.00	1.00
molasses	1.00	0.84
wheat bran	0.87	–
sodium chloride	0.68	0.67
vitamin-mineral pre-mix	0.26	0.26
monocalcium phosphate	0.13	0.16
Saccharomyces cerevisiae	–	0.06
('Actisaf HR+') postbiotic from *Lactobacillus acidophilus* ('ZooLac')	–	0.05
Analysed nutrients		
crude protein, %	18.4	17.6
crude fat, %	4.8	4.6
neutral detergent fibre, %	5.9	5.8
ash, %	5.6	5.5
net energy, MJ/kg DM	7.2	7.4

The PRO concentrate was planned to be similar to the CON concentrate in all ingredients. However, small differences in the amounts of added feedstuffs occurred unintentionally (but were monitored in the recipe) between the two batches when prepared at the commercial feed mill. The differences in rapeseed products and wheat bran are the only unfortunate component differences between the two concentrates. Overall, these small differences only changed the contents of protein, fat and energy by 3 to 4% between the two concentrates. All calves were offered hay *ad libitum*. The net energy content per kg of hay was 6.1 MJ NE/kg DM.

After 20–23 days, eight calves from blocks 2, 4 and 5 were driven to facilities at Aarhus University where they were slaughtered to investigate the effects of the yeast/*lactobacillus* product on the

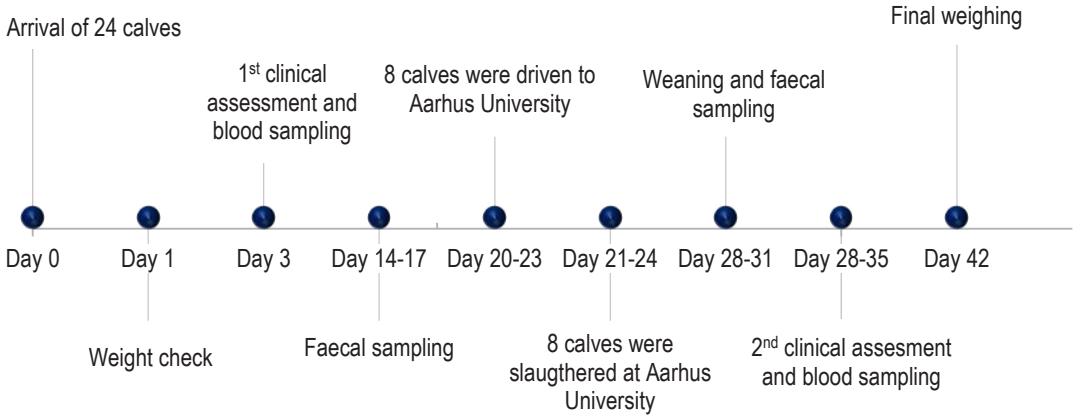

Figure 1. Timeline of the production trial for a block of 24 calves

GIT (Figure 1). The data from these calves and results from the slaughtering are not included in the current study. However, we measured CFU/g of digesta in the mid-small intestine and mid-colon by microbial enumeration by plating and found that the PRO group had a significantly higher number of live yeast cells in the GIT ($P < 0.001$). The logCFU/g of digesta in the mid-small intestine was 2.88 ± 0.14 and 5.56 ± 0.16 and in the mid-colon 3.6 ± 0.2 and 4.78 ± 0.3 for CON and PRO group, respectively.

Health

Antibiotics and electrolytes administered were recorded daily for each calf. During the production trial, two calves allocated to block 3 CON were put down within the first week of arrival due to morbidity. However, the calves already appeared sick on arrival from the dairy herds. Thus, the treatments in the production trial were most unlikely to have affected the health of these calves.

Performance measurements

After six weeks, records of final BW were obtained. The total growth of the calf was calculated by subtraction of the initial weight from the final BW. The average daily gain (ADG) of the calf was calculated by dividing the total growth with the number of days in the production trial.

The provision of concentrate for the whole period was measured per pen for all five blocks. Since roughage was provided in a shared hay-rack between the two pens in the hutch, measurements of the hay intake per pen were not possible to obtain. The concentrate intake of the calves slaughtered during the trial was estimated by using the concentrate intake of calves with similar age, breed and milk feeding level (Jensen, 2017) and subtracting it from the total amount of provided concentrate.

The feed conversion ratio (FCR) per pen was calculated by dividing the total feed intake in MJ NE per pen by the total growth of the actual number of calves in the pen. Next, the FCR was expressed per calf.

Faecal samplings

Twice during the production trial, manure was sampled from blocks 2, 4 and 5. The samples were collected around days 14–17 (n = 24 per block) and days 28–31 (n = 16 per block) (Figure 1). The faecal samples were collected from the rectum by using plastic gloves to stimulate the calf. All calves were individually handled. If the consistency of manure appeared thin, the manure was tested with a test-kit from 'Fassisi BoDia' (Fassisi; Goettingen, Germany). The test kit is a rapid test for *in vitro* diagnosis of antigens from *Escherichia coli* K99, rotavirus, coronavirus and *Cryptosporidium parvum* in the manure. The test-kits were used on 23 faecal samples but none of the tests showed positive results for the tested antigens. Afterward, the samples were stored at −20 °C until they were analysed.

The faecal samples were pooled within treatment, block and sub-block. The total number of pooled faecal samples was 24. The samples were analysed for DM after drying at 60 °C for 48 h.

Blood samplings

Blood samples were collected from blocks 4 and 5 on days 3 (n = 24 per block) and 28–31 (n = 16 per block) (Figure 1). The blood was sampled by puncture of the jugular vein using vacutainer serum tubes. The blood tubes were centrifuged at 3 500 *g* for 5 min in a centrifuge at 6 °C for separation of serum. The serum samples were stored at −20 °C until they were analysed.

The concentration of β-hydroxybutyrate (BHB), glucose, non-esterified fatty acids (NEFA), urea and

total protein (TP) were measured by a spectrophotometric assay following the manufacturer's guidelines (ADVIA 1800; Siemens Medical Solutions, Tarrytown, NY, USA). Concentrations of immunoglobulin G (IgG) and A (IgA) were determined by ELISA (Bethyl Laboratories Inc; Montgomery, TX, USA) following the manufacturer's guidelines.

Statistical analysis

The production trial included five blocks each consisting of 24 calves. Two hutches with calves shipped at the same date constituted a block, and each hutch acted as a sub-block in the experimental design. Thus, the five blocks were further divided into 10 sub-blocks. In the production trial, two calves were put down in block 3, sub-block 5, CON due to illness. Data from these calves were not included in the analysis.

The data was analysed by using a linear mixed model (R, software version 3.5.3, 2019; R Founation, Boston, MA, USA). The model included the fixed effect of treatment and block and the random effects of sub-block. A one-way ANOVA was used to test the effects of the yeast/*lactobacillus* product on the performance of the calves, serum blood metabolites and DM content in the manure, while the number of treatments was tested by using Fisher's exact test. Feed intake, faecal samples and FCR were tested on per pen basis while growth rates, serum levels, etc. were tested on animal basis. All data were examined to discard any possible outliers, which were defined as values more than mean ± 3 × standard deviation. Data were also tested for normality of the residuals by evaluating the QQ-plots constructed in R, and the means were tested for homogeneity of the variance by using Bartlett's test. Statistical significance was declared when $P \leq 0.05$ and statistical tendencies were declared when $0.05 < P \leq 0.10$. Results are presented as least squares means and standard error of the mean.

Results

Health

During the production trial, a total number of five and four calves received veterinary treatments from CON and PRO, respectively. Overall, 89% of the calves allocated to the CON group and 92% of the calves allocated to the PRO group did not receive any treatment for diseases during the six weeks. Neither did the number of treatments for diarrhoea, mycoplasma and pneumonia differ between CON and PRO groups (Table 2).

Table 2. Number of veterinary treatments for diseases in two groups of rosé veal calves either allocated to a control diet (CON) or a diet supplemented with a product containing yeast (*Saccharomyces cerevisiae*) and postbiotic from *Lactobacillus acidophilus* (PRO) for six weeks

Indices	CON	PRO	*P*-value
Total number of treatments[1]	5	4	0.847
Treatments for:			
diarrhoea[1]	1	1	0.999
mycoplasma[1]	2	0	0.456
pneumonia[1]	2	3	0.999

[1] 94 calves were included in the dataset: 46 calves received CON diet and 48 calves received PRO diet

Performance measurements

The BW was similar between CON and PRO groups at the beginning of the production trial (Table 3). However, after six weeks BW and ADG were significantly higher for the PRO group in comparison to the CON calves, while the energy intake was similar between the treatments. Higher growth performance combined with a similar intake of energy caused a significantly lower FCR for the calves allocated to PRO group.

Table 3. Body weight at the beginning and end of the production trial, average daily gain, energy intake and feed conversion ratio

Indices	CON	PRO	SEM	*P*-value
Initial body weight[1], kg	55.1	55.3	1.33	0.772
Final body weight[1], kg	93.1	96.1	2.26	0.049
Average daily gain (ADG)[1], g/day	885	948	26.3	0.036
Intake per day[2], MJ_{NE}	12.3	12.4	0.228	0.316
Feed conversion ratio[2], MJ_{NE}/kg ADG	13.9	13.1	0.347	0.045

SEM – standard error of mean;[1] 94 calves were included in the dataset; 46 calves received a control diet (CON) and 48 calves received a diet supplemented with a product containing yeast (*Saccharomyces cerevisiae*) and postbiotic from *Lactobacillus acidophilus* (PRO); [2] 20 sub-blocks were included in the dataset; 10 sub-blocks received CON diet and 10 sub-blocks received PRO diet. Feed intake was recorded and analysed per pen, but is expressed per calf, feed conversion ratio is also expressed per calf. The calves were kept in pens with 4–6 calves per pen.

Faeces

No treatment effect was found on the DM content at the faecal samplings (Figure 2).

Additionally, none of the 23 tested faecal samples turned out positive for antigens from *Escherichia coli* K99, rotavirus, coronavirus and *Cryptosporidium parvum*.

Blood metabolites

The treatments did not affect the serum levels of glucose, urea, BHB and NEFA at the first or the second sampling (Table 4). The levels of glucose and BHB increased for both treatments from the first

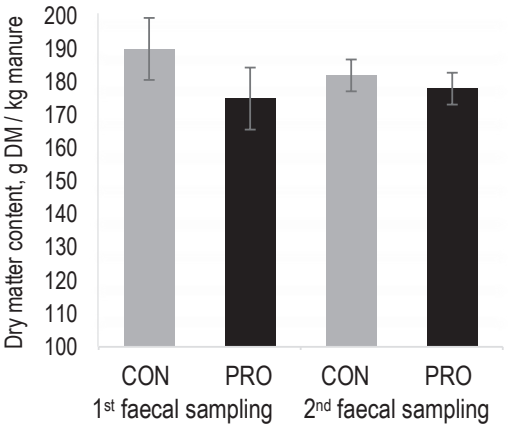

Figure 2. Content of dry matter (DM) in manure sampled from rosé veal calves around day 14–17 (1st sampling) and 28–31 (2nd sampling) after arrival at the veal calf production. The calves received a control diet (CON) or a diet supplemented with a product containing yeast (*Saccharomyces cerevisiae*) and postbiotic from *Lactobacillus acidophilus* (PRO) for six weeks.

Table 4. Level of glucose, urea, β-hydroxybutyrate, NEFA, TP, IgG and IgA in the blood serum of young calves three days and 28–35 days after arrival at the rosé veal calf production in calves fed a control diet (CON) and 48 calves fed a diet supplemented with a product containing yeast (*Saccharomyces cerevisiae*) and postbiotic from *Lactobacillus acidophilus* (PRO)

Indices	CON	PRO	SEM	*P*-value
1st blood sampling[1]				
glucose, mM	5.86	5.70	0.328	0.498
urea, mM	2.75	2.69	0.172	0.791
β-hydroxybutyrate, mM	0.34	0.35	0.025	0.592
NEFA, µM	169	169	21.7	0.957
TP, g/l	53.3	55.6	1.16	0.146
IgG, g/l	11.2	12.8	1.29	0.902
IgA, mg/l	48.7	55.4	3.70	0.197
2nd blood sampling[2]				
glucose, mM	6.78	6.75	0.228	0.651
urea, mM	2.74	2.59	0.214	0.578
β-hydroxybutyrate, mM	0.40	0.39	0.017	0.570
NEFA, µM	79.9	72.4	16.8	0.651
TP, g/l	55.6	59.1	1.033	0.013
IgG, g/l	11.6	15.1	1.20	0.028
IgA, mg/l	90.1	86.7	12.78	0.828

NEFA – non-esterified fatty acids, TP – total serum protein, IgG – immunoglobulin G, IgA – immunoglobulin A, SEM – standard error of mean, [1] 48 calves were included in the dataset; 24 calves received CON diet and 24 calves received PRO diet, [2] 32 calves were included in the dataset; 16 calves received CON diet and 14 calves received PRO diet. Two samples from PRO group were lost.

to second blood sampling while the level of NEFA decreased.

The concentrations of serum TP, IgA and IgG were similar between the treatments at the first sampling, while the PRO group had significantly higher levels of TP and IgG at the second sampling (Table 4).

Discussion

To our knowledge, till now no studies have been published using the same yeast/*lactobacillus* product. Thus, it is not possible to compare the results obtained with existing literature testing the same type of product. However, it can be assumed that the SC in the tested product has a similar mode of action as if the product only consisted of SC instead of SC and 'ZooLac'. Furthermore, only very few studies have investigated the effects of postbiotics from *Lactobacillus* spp. in young ruminants (Izuddin et al., 2019). No studies have been published studying the effect of 'ZooLac' alone.

Health. The total number of veterinary treatments did not differ between the calves allocated to either CON or PRO groups. This might be explained by general low morbidity and mortality in this herd compared to similar herds in Denmark.

In several studies it was found that SC can lower faecal score and frequency of diarrhoea in calves (Galvao et al., 2005; Magalhaes et al., 2008; Hill et al., 2009; Alugongo et al., 2017b) while in other no effect of the supplementation was observed (Lesmeister et al., 2004; Huuskonen and Pesonen, 2015). The inconsistency found in the literature concerning the effect of SC on the frequency of diarrhoea might be related to differences in the infection levels, housing conditions and monitoring of diseases in the studies. The supplementation of SC has been found to lower the faecal score when calves were experimentally challenged with *Salmonella enterica* (Brewer et al., 2014; Harris et al., 2017). Similarly, antimicrobial metabolites such as organic acids and bacteriocins from postbiotics have also been found to inhibit the colonization of pathogenic bacteria in the GIT (Izuddin et al., 2019). It was found that the supplementation of postbiotic from *Lactobacillus* spp. lowers the frequency of diarrhoea in piglets (Thu et al., 2011; Loh et al., 2013).

In the current study, the supplementation of the yeast/*lactobacillus* product did not affect the number of treatments against diarrhoea. The general low morbidity in the herd used in the current study is most likely due to a low infection level. This is supported by 23 negative results of the 23 tested faecal samples for antigens from *Escherichia coli* K99, rotavirus, coronavirus and *Cryptosporidium parvum*. Thus, if the infection level in the herd had been higher, the supplementation of the yeast/*lactobacillus* product might have caused a treatment effect on the number of veterinary treatments for diarrhoea

as both SC and postbiotic from *Lactobacillus* spp. have been found to lower the frequency of diarrhoea in production animals.

The number of veterinary treatments for pneumonia was also similar between the two treatment groups. Similar results of the effect of SC on the number of treatments for pneumonia were found by Magalhaes et al. (2008), Hill et al. (2009), and Huuskonen and Pesonen (2015). This could imply that SC mainly has a positive effect on the immune system associated with the GIT. It has not been possible to find studies investigating the effects of postbiotic from *Lactobacillus* spp. on the frequency of pneumonia in young ruminants.

Growth performance. The supplementation of the yeast/*lactobacillus* product had significant effects on final BW and ADG. Similar effects of SC on the growth performance of calves (Lesmeister et al., 2004; Galvao et al., 2005) and of postbiotic from *Lactobacillus* spp. on the growth performance of piglets (Thu et al., 2011; Loh et al., 2013) and newly-weaned lambs (Izuddin et al., 2019) are found.

It is also important to notice that the CP levels in the MR and concentrate differed slightly between the two treatments due to the addition of the yeast/*lactobacillus* product. The content of CP was 3.5% higher in the MR fed to CON group in comparison to the MR fed to the PRO group. Additionally, the CP level in the CON concentrate was also 4.5% higher in comparison to the PRO concentrate. Even though this could have affected the growth performance, the PRO group, despite the lower the CP supply, still had a significantly higher growth performance. The levels of CP in both diets should be sufficient to secure a normal growth performance of calves at this age and level of gain (Hill et al., 2007).

Several studies have investigated the effects of supplementation of SC on FCR of calves. The studies found no effect of the supplementation on FCR (Quigley et al., 1992; Hill et al., 2009; Hučko et al., 2009; Huuskonen and Pesonen, 2015; Alugongo et al., 2017b). Izuddin et al. (2019) found that the supplementation of postbiotic from *Lactobacillus* ssp. increased the digestibility of DM, CP and NDF, but also significantly increased the intake of DM, OM and NDF of newly-weaned lambs. Thus, FCR was not affected in the study.

In the current study, the calves allocated to the diet with the yeast/*lactobacillus* product had a significantly lower FCR while the energy intake was similar between the treatments. However, it is important to emphasize that the roughage intake was not measured as the treatments had a shared hay-rack in the hutch. It was not possible to change the design of the hutches as the study was carried out in a real production setup. This is often a limitation of production trials in comparison to more experimental setups. Thus, the PRO group could potentially have ingested a higher amount of roughage as the SC in the product is known to increase the number of cellulolytic bacteria and NDF degradation rate in the functionally developed rumen (Callaway and Martin, 1997; Ding et al., 2014). Additionally, postbiotic from *Lactobacillus* spp. has also been found to increase total bacteria, total protozoa and major cellulolytic bacteria *in vitro* (Izuddin et al., 2019). However, the contribution of energy from roughage (i.e. the hay supplied in the current study) is most likely neglectable due to the age of calves during the production trial compared to the contribution of energy coming from concentrate and MR. The calves in the current study are still developing their rumen around weaning, but we cannot exclude that this could have affected FCR.

Blood metabolites. Metabolic responses are valuable in providing information on the nutritional status of the animal and its rumen development. The levels of glucose, BHB and NEFA in the blood serum are good indicators of energy metabolism in calves (Alugongo et al., 2017a).

Before the calf becomes ruminating, glucose is the main energy source due to the limited utilization of volatile fatty acids (VFAs) in the undeveloped rumen epithelium (Baldwin et al., 2004). The concentration of glucose in the serum was not affected by the treatments. Similar results are found by Quigley et al. (1992) and Magalhaes et al. (2008) when SC was supplemented to the diet of calves. The similar intakes of MR and concentrate can explain the lack of treatment effect as the concentration of glucose is influenced by the calf's energy consumption. Increased consumption of energy leads to increased glucose absorption (Magalhaes et al., 2008). This is supported by Galvao et al. (2005) and Izuddin et al. (2019). Galvao et al. (2005) found that calves supplemented with SC in the diet had a significantly higher level of glucose in the serum, both prior and after weaning. However, in the study, the calves did also have a significantly higher intake of concentrate in both periods in comparison to the control group. Similarly, Izuddin et al. (2019) found that newly-weaned lambs supplemented with postbiotics from *Lactobacillus* spp. had a significantly higher level of glucose in comparison to the control group, but these lambs did also have a higher intake of concentrate.

The contribution of VFAs to the energy requirements of the calf increases as the calf consumes a larger amount of solid feed and starts ruminating. The ketogenesis in the ruminal epithelium is believed to result in an elevated concentration of BHB in the blood. In the current study, the supplementation of the yeast/*lactobacillus* product did not affect the level of BHB in the blood serum. In several studies a similar effect on the level of BHB in the blood of calves was found when SC was supplemented to the diet (Lesmeister et al., 2004; Galvao et al., 2005; Magalhaes et al., 2008; Xiao et al., 2016). In addition, the total concentration of VFAs in the rumen of newly-weaned lambs was also unaffected by the supplementation of postbiotics from *Lactobacillus* spp. (Izuddin et al., 2019).

A high concentration of NEFA indicates adipose mobilization as they are mobilized to maintain caloric homeostasis during times of fasting (Adewuyi et al., 2005). The treatments did not affect the concentration of NEFA in the serum at the blood samplings. This indicates a similar and low adipose tissue mobilization between the treatments. This complies with the result found by Quigley et al. (1992) when SC was supplemented to calves.

The level of urea was also unaffected by the supplementation of the product. Similar result was found by Quigley et al. (1992). A lower urea level in the blood could indicate a higher microbial activity and incorporation of ammonia into microbial protein (Robinson and Erasmus, 2009). This could imply that the yeast/*lactobacillus* product has no effect on the synthesis of microbial protein in the rumen of calves.

Similar concentrations of IgG and IgA in the serum at the first sampling imply that the calves were well sorted based on their herd of origin and age to take differences in colostrum management and half-life of the concentrations into account (Roodposhti and Najafgholi, 2012). Measurement of TP by refractometer can be used as a reasonably accurate assessment of passive transfer of immunity status. Tyler et al. (1996) suggested that a TP concentration ≥52 g/l indicates an adequate passive transfer of immunity. Thus, with a mean TP level of 54 g/l, the calves generally had a high immune status at the beginning of the production trial. This can explain the relatively low morbidity during the production trail.

β-glucan, which constitutes 50–60% of the polysaccharides in the yeast cell wall, has been suggested as a potential immunomodulatory agent (Kogan and Kocher, 2007). β-glucans have been found to activate macrophages, neutrophils, natural killer cells, B and T lymphocytes, and increase phagocytosis and cytokine production in macrophages *in vivo* and *in vitro* (Jensen et al., 2008). The activation of B lymphocytes might explain the significantly higher concentration of IgG in the serum of the PRO group found at the second sampling. Roodposhti and Najafgholi (2012) found that the supplementation of polysaccharides isolated from the SC cell wall also tended to increase the concentration of IgG in the blood plasma of calves. Similar effects of SC on the concentration of IgG are found by White et al. (2002) in the serum of piglets and by Zanello et al. (2013) in the colostrum of sows. The supplementation of postbiotic from *Lactobacillus* has also been found to increase the concentration of IgG in broilers (Humam et al., 2019). However, to our knowledge, the potential mode of action is unknown.

Conclusions

The supplementation of the product containing *Saccharomyces cerevisiae* and postbiotic from *Lactobacillus acidophilus* in milk replacer for four weeks and in the concentrate for six weeks did not affect the overall health of young rosé veal calves around weaning. The number of veterinary treatments for diarrhoea and pneumonia was similar between the two treatments.

However, average daily gain and body weight after six weeks were significantly higher in calves fed the supplemented diet. Furthermore, the serum concentrations of immunoglobulin G and total protein were significantly higher in the serum of calves supplemented with the yeast/*lactobacillus* product after 4 to 5 weeks of treatment. Other blood metabolites measured were unaffected by the treatments.

Acknowledgments

The study was carried out within the research project 'Robust calves' granted by The Danish Milk Levy Fund and The Danish Cattle Levy Fund. We are indebted to the skilled work done by the private farmer and his staff where the trial took place. Lene Jensen, DVM is acknowledged for taking the blood samples and performing the clinical assessments of calves. Torben Larsen and his staff are acknowledged for analysing the blood samples, and Nuria Canibe is acknowledged for measuring CFU/g of intestinal digesta by microbial enumeration by plating. The yeast/*lactobacillus* products were supplied free of charge to the farmer by ChemVet (Silkeborg, Denmark).

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