# *Saccharomyces cerevisiae* diet supplementation influences haematological parameters in healthy steers

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## SUMMARY

Intensive farm conditions, overcrowding and limited individual space, high grain feed, transportation, exposure to pathogens and high productivity are several stressors that can threaten animal welfare and the search for different tools to help maintain the balance between high farm productivity and animal welfare is increasingly well established. The effects of yeast *Saccharomyces cerevisiae* diet supplementation on cattle growth performance were widely investigated, but few studies debated about the health status of steers. For this purpose, two groups of Charolaise steers were equally divided according to the type of administered food: the control group (CG), which received the base diet without yeast supplement and the treatment group (YG), which each animal received the base diet with 5g of yeast *Saccharomyces cerevisiae* supplementation (YS) per day. From each group, blood samples were collected at three different time point, before (t0), after 21 (t1) and 42 (t2) days of the start of the study to evaluate changes on haematological parameters, including red blood cells (RBC), haematocrit (HCT), haemoglobin (HGB), mean corpuscular haemoglobin (MCH), mean corpuscular haemoglobin concentration (MCHC), mean corpuscular volume (MCV), white blood cells (WBC), neutrophils (NEU), lymphocytes (LYM), monocytes (MONO), eosinophils (EOS), basophils (BASO), and platelets (PLT). According to two-way analysis of variance (ANOVA), some haematological parameters including RBC ($P<0.01$; $F_{(2,116)} = 9.08$), HGB ($P<0.001$; $F_{(2,116)} = 16.17$), HCT ($P<0.001$; $F_{(2,116)} = 9.67$), MCV ($P<0.05$; $F_{(2,116)} = 29.42$), MCH ($P<0.001$; $F_{(2,116)} = 43.90$), MCHC ($P<0.05$; $F_{(2,116)} = 44.27$), MONO ($P<0.001$; $F_{(2,116)} = 15.34$), EOS ($P<0.001$; $F_{(2,116)} = 8.24$), BASO ($P<0.01$; $F_{(2,116)} = 43.15$) and PLT ($P<0.001$; $F_{(2,116)} = 15.76$) showed a significant effect of time and group. Results gathered in the current study suggest that *Saccharomyces cerevisiae* diet supplementations do not have a significant impact on the health status of cattle.

## KEY WORDS

Live yeast; growth; haematological parameters; steers.

## INTRODUCTION

Nowadays, the livestock systems aim to enhance animal growth and productivity to maximize profit in a rather short time. At this purpose, beef steers are feeding with minimum of roughage and high amount of concentrate with detrimental impacts on animal health (1). Certainly, highly fermentable substrates can lead to the imminent rumen dysfunction by an alteration of pH with negative impact on ruminal microbial ecosystem, ruminal inflammation and metabolism disorder, such as acidosis (2). The yeast supplement can help to minimize the negative effects of altered ruminal fermentation pattern, in order to keep a good health status and welfare of the animal (3). In particular, yeast supplementation improves stability of ruminal pH, digestibility of organic matter and fiber by modifying the microflora of the host's digestive tract and (4). Moreover, yeast supplementation is widely used in intensive dairy cattle farm to support milk production and feed conversion efficiency (5) and in beef cattle farm to improve the digestion of fiber and utilize the lactate by bacteria and to improve growth performance and safeguard liver health (6-9). In spite of that, other authors found similar or reduced growth rate between group feeding with yeast supplementation and control group (10, 11). These controversial results on yeast supplementation effects may depend on diet composition and yeast dose used (12). In livestock system, animals are usually submitted to a high amount of stressors caused by several factors such as high productivity, overcrowded and limited individual space, transportation, vaccination, exposure to pathogens and poor quality nutrition (13). These aspects have a negative impact on immune system and animals become susceptible to several pathogens, hence an extensive use of antibiotics for prophylactic purpose in farms is common. Yeasts have been em-

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ployed as replacement of antibiotics used as growth promotans and interact directly with immune cells, modifying their blood concentration (8, 14).

In view of such consideration, the aim of the present study was to evaluate the effect of time and of treatment on haematological parameters measured in steers feed with a diet supplemented with *Saccharomyces cerevisiae*.

## MATERIALS AND METHODS

### 2.1 Animal and experimental design

Sixty Charolaise steers, 10 months of age, initial body weight 518±16.24, were selected from a farm located in the Northeast of Italy (45° 24' N: 11° 52' E, 12 m above sea level) and were enrolled in the study. All animals are clinically healthy and free from external and internal parasites. Their status was evaluated based on rectal temperature, heart and respiratory rate, fecal consistency and haematochemical profile. Animals were kept into pens and had free access to water.

Animals were divided into two equal groups, 30 animals each: the treatment group (YG) received the base diet with 5g of *Saccharomyces cerevisiae* supplementation; the control group (CG) received the base diet without yeast supplementation. The viable cells of *Saccharomyces cerevisiae* were a strain (NCYC Sc 47) produced by batch fermentation in a growth medium typical of those used for the industrial production of yeasts and with guaranteed concentration of 1010 CFU/g.

All treatments, housing and animal care were carried out in accordance with the standards recommended by the EU Directive 2010/63 EU for animal experiments.

### 2.2 Sampling and laboratory analysis

Blood samples were collected by jugular venepuncture, from both groups, into vacutainer tubes with EDTA anticoagulant agent, at day 1 (t0) and at 21 (t1) and 42 (t2) days of the start of experimental period. The sampling was carried out by qualified and experienced personnel, avoiding unnecessary injuries and stress to the animals. EDTA whole blood samples were processed in the laboratory within 2 hours by means of an au-

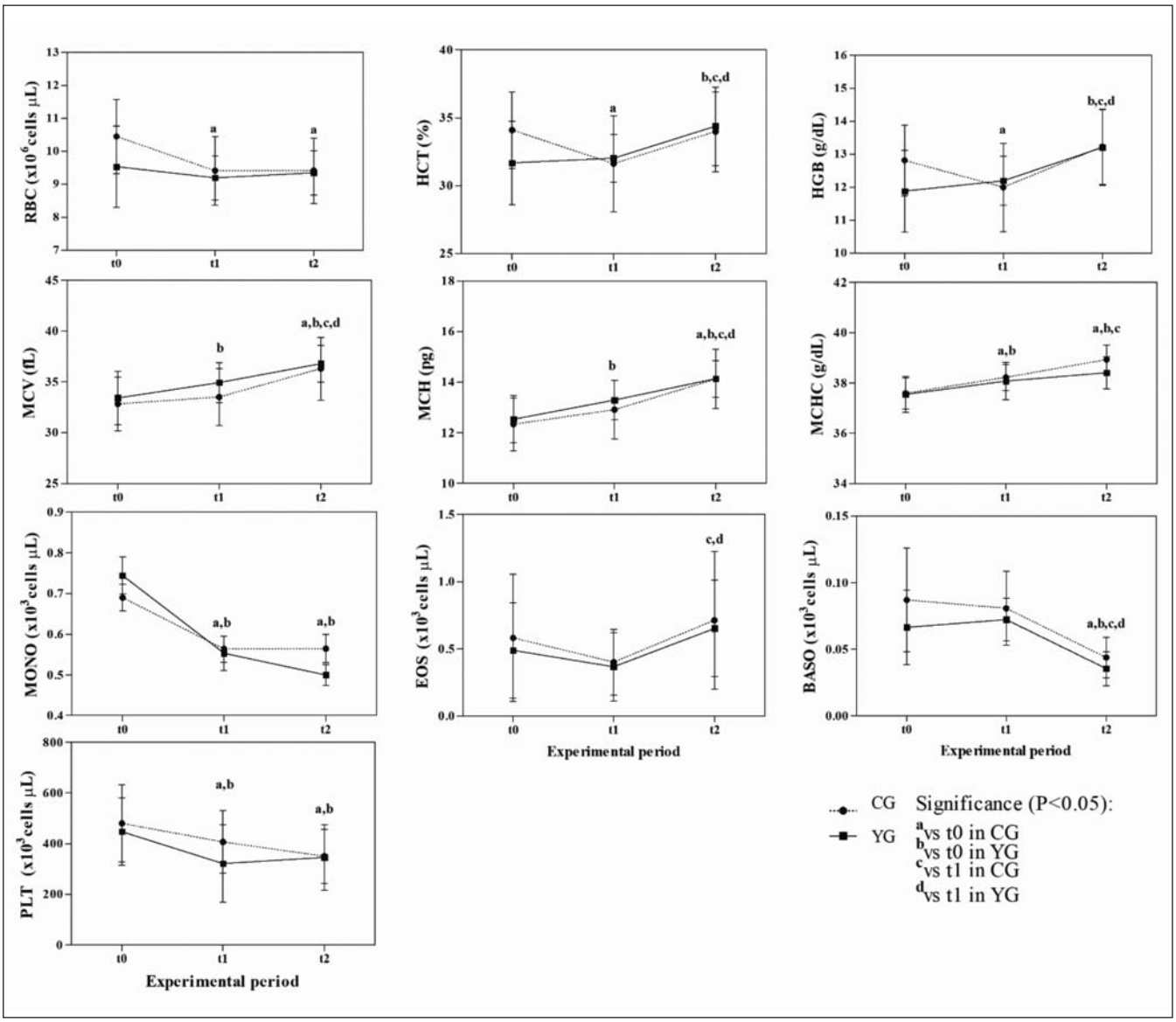


**Figure 1** - Mean values ± SD of haematological parameters, together with differences related to time, measured in steers feeding with the base diet with 5g of Saccharomyces cerevisiae (YG) and in control steers (CG) feeding with the base diet without yeast supplementation.

**Table 1** - Mean values ± SD of haematological parameters measured in steers feeding with the base diet with 5g of Saccharomyces cerevisiae (YG) and in control steers (CG) feeding with the base diet without yeast supplementation.

| Parameters | Group | t0 | t1 | t2 |
|---|---|---|---|---|
| RBC (x10$^6$ cells/μL) | CG | 10.31±1.19 | 9.42±1.11 | 9.42±0.99 |
| | YG | 9.64 ±1.14 | 9.20±0.70 | 9.35±0.68 |
| HGB (g/dL) | CG | 12.71±1.22 | 12.10±1.31 | 13.23±1.15 |
| | YG | 11.92±1.17 ** | 12.21±0.77 | 13.21±1.17 |
| HCT (%) | CG | 33.80±3.36 | 31.70±3.44 | 33.99±2.95 |
| | YG | 31.66±2.97 ** | 32.04±1.80 | 34.39±2.94 |
| MCV (fL) | CG | 32.95±2.66 | 33.80±2.86 | 36.30±3.09 |
| | YG | 33.02±2.54 | 34.93±2.05 | 36.79±1.83 |
| MCH (pg) | CG | 12.39±1.01 | 12.91±1.13 | 14.13±1.17 |
| | YG | 12.44±0.89 | 13.29±0.80 | 14.13±0.74 |
| MCHC (g/dL) | CG | 37.60±0.66 | 38.20±0.50 | 38.93±0.58 |
| | YG | 37.69±0.70 | 38.08±0.77 | 38.41±0.66 ** |
| WBC (x10$^3$ cells/μL) | CG | 10.02±1.91 | 9.27±1.53 | 10.05±1.82 |
| | YG | 10.28±2.35 | 9.37±1.57 | 9.77±1.54 |
| NEU (x10$^3$ cells/μL) | CG | 2.44±0.94 | 2.49±0.80 | 2.95±0.90 |
| | YG | 2.76±1.82 | 2.70±1.04 | 2.70±0.74 |
| LYM (x10$^3$ cells/μL) | CG | 6.10±1.20 | 5.68±1.04 | 5.69±1.19 |
| | YG | 5.98±1.38 | 5.62±1.30 | 5.79±1.18 |
| MONO (x10$^3$ cells/μL) | CG | 0.68±0.18 | 0.57±0.19 | 0.56±0.19 |
| | YG | 0.73±0.22 | 0.55±0.24 | 0.50±0.14 |
| EOS (x10$^3$ cells/μL) | CG | 0.59±0.48 | 0.40±0.26 | 0.71±0.51 |
| | YG | 0.61±0.55 | 0.37±0.26 | 0.65±0.37 |
| BASO (x10$^3$ cells/μL) | CG | 0.08±0.04 | 0.08±0.03 | 0.04±0.02 |
| | YG | 0.07±0.03 | 0.07±0.02 | 0.04±0.01 |
| PLT (x10$^3$ cells/μL) | CG | 467.79±159.25 | 406.65±133.57 | 350.27±106.39 |
| | YG | 446.49±138.82 | 321.89±158.23 * | 345.45±131.24 |

*Significant effect of group: *P<0.05 and ** P<0.01*

tomated hematology analyzer (HeCo Vet C; SEAC, Florence, Italy) for the evaluation of complete blood count including RBC, HCT, HGB, MCH, MCHC, MCV, WBC, NEU, LYM, MONO, EOS, BASO, PLT. Leukocyte identification and counting was performed on all whole blood samples by manual analysis.

## 2.3 Statistical analysis

The obtained data were expressed as mean ± standard deviation (SD).

For each group, a separate analysis of variance (ANOVA) with repeated measures was applied to determine the influence of feed supplementation and of time (t0, t1, t2) on haematological parameters in both groups. Bonferroni multiple comparison tests were applied for post hoc comparisons. All the statistical analyses were performed using the Statistica 8 software (Statsoft Inc., Tulsa, OK, USA). *P values*<0.05 were considered statistically significant.

# RESULTS

Table 1 showed mean values ± SD and significant effect of groups of the blood haematological parameters (RBC: $F_{(1,116)}$=7.29; MCV: $F_{(1,116)}$=4.72; MCHC: $F_{(1,116)}$=5.92; MPV: $F_{(1,116)}$=5.35; BASO: $F_{(1,116)}$=11.9). As showed in Fig. 1, two-way analysis of variance (ANOVA) showed a significant effect of time on RBC ($F_{(2,116)}$=9.08), HGB ($F_{(2,116)}$=16.17), HCT ($F_{(2,116)}$=9.67), MCV ($F_{(2,116)}$=29.42), MCH ($F_{(2,116)}$=43.90), MCHC ($F_{(2,116)}$=44.27), MONO ($F_{(2,116)}$=15.34), EOS ($F_{(2,116)}$=8.24), BASO ($F_{(2,116)}$=43.15), PLT (P<0.001; $F_{(2,116)}$=15.76) and HCT (P<0.01; $F_{(2,116)}$=9.67). No significant effect of time (P>0.05) was observed on WBC, NEU and LYM in both groups throughout the experimental study.

# DISCUSSION

A lot of works has demonstrated that yeast and its product fermentation supplementation improved ruminal fermentation, feed efficiency, energy status and body weight of cattle and the organism reaction for inflammation due to feed high grain rations (15-18). The yeast supplementation can provide favourable effects in terms of profitability, but there are few studies regarded effects on health status of steers (19, 20). Piccione et al (21) found a significant lower acute phase response in treated group respect to control group during the finishing phase. Moreover, Idowu et al (16) found a positive effect on health, during and after administration of yeast fermentation products, related to their ability to reduce inflammatory stress. Other authors suggest that cattle resulted better prepared for exposure to a

pathogen hereafter previous immune stimulation by feed supplementation with a yeast fermentation product (22). Regarding haematology results, a significant decrease of RBC from t2 and t1 compared to t0 was observed in CG but no change was observed in YG. Results concerning HGB, suggest that its concentration showed an increase during the experimental study as a consequence of a continuous increase in muscle mass of steers (23). HGB, HCT and PLT concentrations values were result higher in CG respect YG during t0, probably for transient hydration differences between groups (21). Likewise, a significant decrease of MONO concentrations from YG and from CG were observed and it can be partially attributed to a reduction in inflammatory stress throughout experimental study. A study carried out on growing beef cattle showed that supplementation of hydrolysed yeast did not influence erythrocytes parameters, WBC, lymphocytes, or eosinophils, while neutrophils and monocytes were increased with hydrolysed yeast supplementation (24). Similarly, Adili et al. (25) reported that neutrophils were increased by the addition of hydrolysed yeast to dairy cows. Neutrophils can protect livestock against the most common infectious diseases (26). Kim et al. (27) observed that Holstein calves fed hydrolysed yeast showed enhanced neutrophils. Similarly, Wang et al. (28) indicated that live yeast increases the expression of genes that improve the function of neutrophils, especially those that code for the IL-4 receptor and IL-1B in dairy cattle. Pedro et al. (29) found that Dectin-1 activation increases the expression of pro-inflammatory cytokines in monocytes in response to  -glucan in yeast products. In addition, modulation of monocyte activation has also been related to bovine neutrophil degranulation (30). These results indicate that the addition of yeast to the cattle has the possibility of reducing inflammatory factors via enhanced neutrophils and monocytes in growing beef cattle.

## CONCLUSION

The results gathered in the current study suggest that the base diet with addition of 5gr yeast (*Saccharomyces cerevisiae* NCYC Sc 47) supplementation did not negatively affect the overall health status of steers as suggested by haematological changes herein found. However, further studies are needed in order to evaluate the impact of a higher concentration of yeast supplement on steers wellness at all stages of their farming life.

## References


1. Dias A.L.G., Freitas J.A., Micai B., Azevedo R.A., Greco L.F., Santos J.E.P. (2018). Effect of supplemental yeast culture and dietary starch content on rumen fermentation and digestion in dairy cows. J Dairy Sci, 101: 201-221.
2. Nagaraja T.G., Chengappa M.M. (1998). Liver abscesses in feedlot cattle: a review. J Anim Sci 76: 287-298.
3. Adams D.C., Galyean M.L., Kiesling H.E., Wallace J.D., Finkner M.D. (1981). Influence of viable yeast culture, sodium bicarbonate and monensin on liquid dilution rate, rumen fermentation and feedlot performance of growing steers and digestibility in lambs. J Anim Sci, 53: 780-788.
4. Shen Y., Wang H., Ran T., Yoon I., Saleem M.A., Yang W. (2018). Influence of yeast culture and feed antibiotics on ruminal fermentation and site and extent of digestion in beef heifers fed high grain rations. J Anim Sci, 96(9): 3916-3927.
5. Poppy G.D., Rabiee A.R., Lean I.J., Sanchez W.K., Dorton K.L., Morley P.S. (2012). A meta-analysis of the effects of feeding yeast culture produced by anaerobic fermentation of saccharomyces cerevisiae on milk production of lactating dairy cows. J Dairy Sci, 95:6027-6041.
6. Wiedmeier R.D., Arambel M.J., Walters J.L. (1987). Effect of yeast culture and aspergillus oryzae fermentation extract on ruminal characteristics and nutrient digestibility. J Dairy Sci, 70: 2063-2068.
7. Callaway E.S., Martin S.A. (1997). Effects of a Saccharomyces cerevisiae culture on ruminal bacteria that utilize lactate and digest cellulose. J. Dairy Sci. 80:2035- 2044.
8. Broadway P.R., Carroll J.A., Sanchez N.C. (2015). Live Yeast and Yeast Cell Wall Supplements Enhance Immune Function and Performance in Food-Producing Livestock: A Review. Microorganisms. 3(3): 417-27.
9. Alberghina D., Fiore E., Piccione G., Marafioti S., Morgante M., Gianesella M. (2016). Evaluation of hepatic markers and body weight gain in growing and finishing steers. Comp Clin Pathol, 25: 721-725.
10. Tripathi M.K., Karim S.A., Chaturvedi O.H. Verma D.L. (2008). Effect of different liquid cultures of live yeast strains on performance, rumen fermentation and microbial protein synthesis in lambs. J Anim Physiol Anim Nutr, 92: 631-639.
11. Armato L., Gianesella M., Fiore E., Arfuso F., Rizzo M., Zumbo A., Giudice E., Piccione G., Morgante M. (2016). Effect of live yeast & yeast cell wall Saccharomyces cerevisiae diet supplementation on faeces chemical composition and growth performance in growing and finishing beef steers. LAR, 22: 203-210.
12. Lòpez-Soto M.A., Valdès-Garcìa Y.S., Plascencia A., Barreras A., Castro-Perez B.I., Estrada-Angulo A., Rios F.G., Gòmez-Vazquez A., Corona L., Zinn R.A. (2013). Influence of feeding live yeast on microbial protein synthesis and nutrient digestibility in steers fed a steam-flaked corn-based diet. Acta Agric Scand, AAnim Sci, 63, 39-46.
13. Lynch E., McGee M., Earley B. (2019). Weaning management of beef calves with implications for animal health and welfare. J App Anim Res, 47:167-175.
14. Ran T., Shen Y.Z., Saleem A.M., AlZahal O., Beauchemin K.A., Yang W.Z. (2018). Using ruminally protected and nonprotected active dried yeast as alternatives to antibiotics in finishing beef steers: growth performance, carcass traits, blood metabolites, and fecal Escherichia coli. J Anim Sci, 96(10): 4385-4397. Erratum in: J Anim Sci, 96(12): 5345.
15. Armato L., Gianesella M., Morgante M., Fiore E., Rizzo M., Giudice E., Piccione G. (2016). Rumen volatile fatty acids × dietary supplementation with live yeast and yeast cell wall in feedlot beef cattle, Acta Agric Scand, AAnim Sci, 66(2): 119-124.
16. Idowu M.D., Taiwo G., Pech Cervantes A., Bowdridge S.A., Ogunade I.M. (2022). Effects of a multicomponent microbial feed additive containing prebiotics and probiotics on health, immune status, metabolism, and performance of newly weaned beef steers during a 35-d receiving period. Transl Anim Sci, 6(2):txac053.
17. Adeyemi J.A., Harmon D.L., Compart D.M.P., Ogunade I.M. (2019). Effects of a blend of Saccharomyces cerevisiae-based direct-fed microbial and fermentation products in the diet of newly weaned beef steers: growth performance, whole-blood immune gene expression, serum biochemistry, and plasma metabolome1. J Anim Sci, 97(11):4657-4667.
18. Ogunade I.M., McCoun M., Idowu M.D., Peters S.O. (2020). Comparative effects of two multispecies direct-fed microbial products on energy status, nutrient digestibility, and ruminal fermentation, bacterial community, and metabolome of beef steers. J Anim Sci, 98(9):skaa201.
19. Burdick Sanchez N.C., Young T.R., Carroll J.A., Corley J.R., Rathmann R.J., Johnson B.J. (2013). Yeast cell wall supplementation alters the metabolic responses of crossbred heifers to an endotoxin challenge. Innate Immun, 20: 104-112.
20. Shen Y., Davedow T., Ran T., Saleem A.M., Yoon I., Narvaez C., Mcallister T.A., Yang W. (2019). Ruminally protected and unprotected *Saccharomyces cerevisiae* fermentation products as alternatives to antibiotics in finishing beef steers1. J Anim Sci, 97(10): 4323-4333.
21. Piccione G., Badon T., Bedin S., Giannetto C., Morgante M., Giudice E., Gianesella M., Fiore E. (2021). Evaluation of yeast supplementation in steers housed under suitable temperature-humidity index, Biol Rhythm Res, 52(9), 1313-1321.
22. Burdick Sanchez N.C., Carroll J.A., Broadway P.R., Edrington T.S., Yoon I., Belknap C.R. (2020). Some aspects of the acute phase immune response to a lipopolysaccharide (LPS) challenge are mitigated by supplementation with a *Saccharomyces cerevisiae* fermentation product in weaned beef calves. Transl Anim Sci, 24,4(3):txaa156.
23. Owens F.N., Gill D.R., Secrist D.S., Coleman S.W. (1995). Review of some aspects of growth and development of feedlot cattle. J Anim Sci, 73(10): 3152-72.

24. Gunun N., Sanjun I., Kaewpila C., Foiklang S., Cherdthong A., Wanapat M., Polyorach S., Khota W., Kimprasit T., Kesorn P., Milintawisamai N., Gunun P. (2022). Effect of Dietary Supplementation of Hydrolyzed Yeast on Growth Performance, Digestibility, Rumen Fermentation, and Hematology in Growing Beef Cattle. Animals, 12, 2473.
25. Adili S., Sadeghi A.A., Chamani M., Shawrang P., Forodi F. (2020). Autolysed yeast and yeast extract effects on dry matter intake, blood cells counts, IGG titer and gene expression of IL-2 in lactating dairy cows under heat stress. Acta Sci Anim Sci, 42: e48425.
26. Bassel LL., Caswell J.L. (2018). Bovine neutrophils in health and disease. Cell Tissue Res, 371: 617-637.
27. Kim E.T., Lee H.G., Kim D.H., Son J.K., Kim B.W., Joo S.S., Park D.S., Park Y.J., Lee S.Y., Kim M.H. (2020). Hydrolyzed yeast supplementation in calf starter promotes innate immune responses in Holstein calves under weaning stress condition. Animals, 10: 1468.
28. Wang Y.Q., Puntenney S.B., Burton J.L., Forsberg N.E. (2009). Use of gene profiling to evaluate the effects of a feed additive on immune function in periparturient dairy cattle. J Anim Physiol Anim Nutr, 93: 66-75.
29. Pedro A.R.V., Lima T., Fróis-Martins R., Leal B., Ramos I.C., Martins E.G., Cabrita A.R.J., Fonseca A.J.M., Maia M.R.G., Vilanova M., Correia A. (2021). Dectin-1-mediated production of pro-inflammatory cytokines induced by yeast -glucans in bovine monocytes. Front Immunol, 12: 689879.
30. Hussen J., Koy M., Petzi W., Schuberth H.J. (2016). Neutrophil degranulation differentially modulates phenotype and function of bovine monocyte subsets. Innate Immun., 22: 124-137.

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