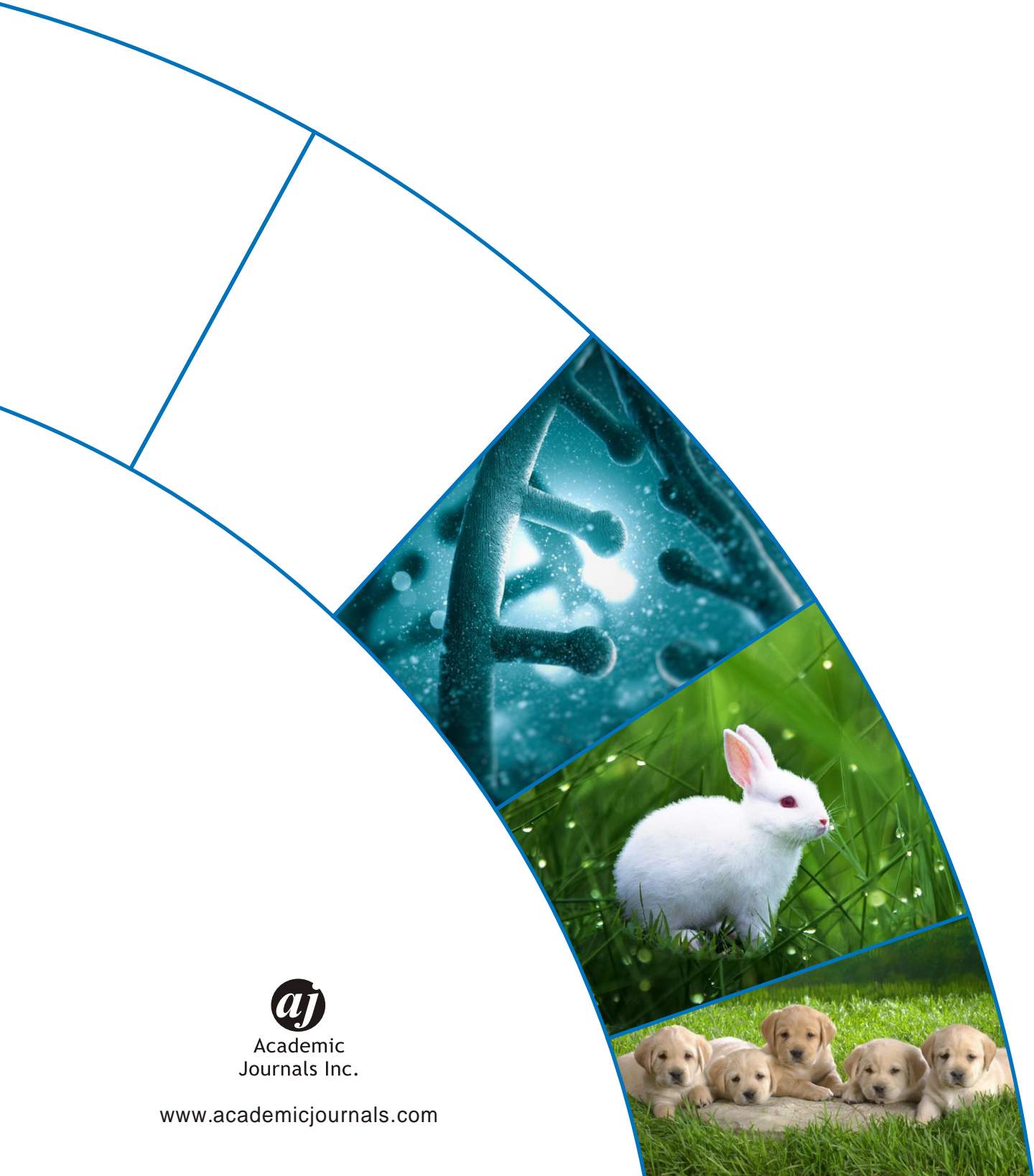


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Research Article

Effect of Dietary Yeast on Male Dromedary Camels During the Rut: Behavioral and Biochemical Insights

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Abstract

Background and Objective: Camels used as meat-producing animals in Egypt, but suffer from economic losses represented by a 25% decrease in body weight due to a partial loss of appetite during rutting season. This study examined the impact of dietary addition of dried baker's yeast (*Saccharomyces cerevisiae*) on behavioral and biochemical parameters of male Dromedary camels during a rutting period.

Materials and Methods: Data from male Dromedary camels in a rut with an average age of 5-7 years and weighing between 385 and 400 kg were collected after three months of dietary addition of dried baker's yeast compared to control (basal feed only). The behavioral observation was performed two days a week, (1 hr per day) for both groups via a continuous focusing sampling. At the end of the experiment, 16 blood samples were collected for biochemical analysis. **Results:** The addition of yeast to the diet significantly increased the frequency and duration of the different behaviors, however, standing and sexual behavior showed no significant change; only aggressive behavior appeared to be lower in yeast-fed camels. Due to yeast supplementation, growth performance parameters have shown significant improvement ($p < 0.05$); in most of the studied traits, except for the leftover trough. Increases in the total protein, albumin, globulin and Albumin/Globulin (A/G) ratio and glucose, urea, calcium and phosphorus levels were observed in the yeast-fed camels; serum cholesterol, triglycerides, cortisol, testosterone, T_3 and T_4 decreased with yeast supplementation, whereas the liver enzymes (AST and ALT) and creatinine levels did not alter. **Conclusion:** Dietary supplementation of yeast provided beneficial effects on behaviors, growth performance and blood parameters of camels during rut, moreover, the recommended dose for camels at rut represented 40 g active dried yeast.

Key words: Dromedary camels, behavior, performance, biochemical parameters, rut season

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Competing Interest: The authors have declared that no competing interest exists.

Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

Dromedary camel (*Camelus dromedarius*) is one of Egypt's most important domestic animals with a 152,946-head population¹. Camels can produce more milk and meat and retain production at extreme temperatures and during droughts and poor pastures longer than other animal species². Camels are seasonal breeders with mating occurring in the coldest months of the year, which is one of the major factors restricting camel reproductive performance during the limited breeding season³. Male camel husbandry switches to a more intensive system that is kept isolated and used for programmed matching or artificial insemination⁴.

Throughout breeding season, male camels exhibit morphological, behavioral and endocrinological peculiarities, increases in pacing and anxiety and excessive restlessness and aggressiveness⁵. The male camel's rut period is between 50 and 100 days⁶ or can vary from 4-6 months⁷ with multiple consequences, such as partial loss of appetite⁸ and a tendency to off-feed⁹ with a subsequent decrease in body weight up to 25%¹⁰. Probiotics are one of the feeding strategies used to boost animal health and production for different livestock species¹¹. Significant positive effects of probiotics on growth efficiency, nutrient digestibility, intestinal microflora balance and immune function enhancement have previously been reported¹². They are considered to be live micro-organisms that when supplied in sufficient amounts, provide health benefits for the host¹³.

Live yeast, yeast cell wall and yeast cell wall extracts have been used as a natural protein sources in ruminant feeding to enhance animal performance, health and immune responses¹⁴. The main components of yeast cell walls consist of polysaccharides, such as α or β glucans¹⁵, which can serve as feed additives to improve feed intake¹⁶ by enhancing dry matter intake¹⁷, improving nutrient utilization¹⁸ and increasing average daily weight gains in addition to final body weight¹⁷, which ultimately leads to improvements in animal performance¹⁹.

However, no applied research based on enhancing camel behavior during the rut season using yeast has been reported; thus, current research reported new findings and additional data are presented concerning young Maghraby calves²⁰ or dairy camels²¹. Therefore, current research aimed to study the effects of feeding yeast on behavior, performance and blood parameters in camels during rut season as a new way to improve its productive performance during this period.

MATERIALS AND METHODS

Study area and animal management: This work was performed with 16 healthy male dromedary camels with weights ranging from 385-400 kg and ages from 5-7 years. The study was carried out from February-May, 2019 at the private camel farm in Manfloat City, Assiut Governorate, Egypt. The animals were randomly divided into two groups (eight camels per group), 1st group; control group that was fed basal diet only and 2nd group; yeast group that was fed basal diet + 40 g active dried yeast supplying 20×10^9 Colony Forming Units (CFU) of *Saccharomyces cerevisiae* per head daily with the same space allowance of 15 m² per camel in semi-covered floor area with sand floors²² and 0.60 cm in feeding areas. Camels were fed with approximately 3% of their live weight²³. This diet consists of 40% concentrate containing 11% proteins and 60% hay, which was divided into two equivalent weights, given twice daily between 8.00 and 9.00 am and 4.00 and 5.00 pm, while at 11:00 am and 12:00 pm, green fodder was used as 1.5 kg per day per camel. Such pens are ventilated naturally and use natural light sources throughout the experimental period.

Behavioral observations: The behavioral observation session was conducted two days a week for both groups (each session lasted 1 hr per day). In total, 48 (1 hr \times 2 days \times 2 weeks \times 2 groups) observations were made. Behaviors were recorded using a clearly defined method of continuous focusing sampling²⁴. During the experimental period, each scan yielded the duration and frequency of the several behavioral states: feeding (collect food from feeders), ruminating (a bolus of regurgitating food goes back into his mouth and the camel chews and swallows it again while standing or lying down), lying in a crouching position or fully stretching camel and standing (camel standing on three or four legs). The observer also recorded the sexual and aggressive behavior of male camels according to the procedure defined by Fatnassi *et al.*⁷.

Scoring of trough leftovers: Visual observations were conducted daily for scoring of trough leftovers. The scores were assessed on a scale from 1-5 on which: 1 = no food, 2 = scattered food, which was most of the apparent trough, 3 = thin and even layer on the bottom of trough, 4 = trough with food (20% of the food supplied in the previous meal) and 5 = full trough, above 50% of the amount provided in the previous meal²⁵ after 2 hrs of feeding.

Body weight and body gain: The evaluated variables were Body Weight (BW), which was calculated as the average weight gain was calculated as the difference between the final and initial BW during the experimental period.

Body condition score: Scores of 1-5 were adopted to determine Body Condition Score (BCS)²⁶: (1) very lean, (2) below average, (3) average or ideal, (4) above average and (5) very fat.

Blood sampling and serum preparation: Blood samples from the jugular vein of 16 camels were obtained in vacuon tubes containing the anticoagulant, ethylenediaminetetraacetic acid (EDTA) were centrifuged at 3000 rpm 10 min⁻¹ to obtain plasma which was stored at -20°C before analysis²⁷.

Total serum protein, albumin, glucose, urea, triglycerides and total cholesterol in addition to Aspartate and Alanine aminotransferase (AST and ALT, respectively), were measured with a colorimetric method using a trade kit developed by the Egyptian Biotechnology Company. In addition, serum globulin and ratio of albumin/globulin were calculated according to Ducan *et al.*²⁸. Serum tri-iodothyronine (T₃), thyroxin (T₄), testosterone and cortisol hormonal levels were measured by commercial enzyme-linked immunosorbent assay (ELISA) kits.

Statistical analysis: In this study, the data collected were statistically analyzed by means of SPSS 22.00 Software. For comparing groups, a T-test was used. The data is displayed as Mean ± Standard Error (SE), based on average values, for each group.

RESULTS

Behavioral responses: In yeast-fed camels, the frequency of all behavioral patterns was significantly higher than that in controls, except for standing behavior and sexual activity in Fig. 1a, as yeast feeding in male camels did not affect the frequency of standing behavior and sexual activity, but there was a decrease in aggressive behavior frequency as shown in Fig. 1a. Besides, there was a significant difference in the duration of feeding, rumination and lying, but not for standing; between the yeast-fed group and the control, as shown in Fig. 1b.

Based on the addition of yeast, the estimated trough leftover scores decreased significantly ($p < 0.01$); while BCS, BW and BWG increased significantly in yeast fed camels relative to the control group in Table 1.

Table 1: Effect of yeast supplementation on camel performance during the receiving period

Parameters	Dietary treatment	
	Control	Yeast
Scoring of trough leftovers	3.4 ± 0.2 ^a	2.7 ± 0.1 ^b
Body condition score	3.1 ± 0.3 ^b	3.9 ± 0.5 ^a
Average initial body weight (kg)	385.0 ± 1.2	390.0 ± 2.2
Average final body weight (kg)	432.0 ± 2.1 ^b	465.0 ± 2.4 ^a
Average body weight gain (kg)	47.0 ± 0.9 ^b	75.0 ± 1.2 ^a

^{a,b}Means with different superscripts within a row differ significantly ($p < 0.05$)

Table 2: Effect of feeding *Saccharomyces cerevisiae* on physiological body functions in Dromedary camels during rut season

Parameters	Dietary treatment	
	Control	Yeast
Blood chemicals		
Total protein (g 100 mL ⁻¹)	6.80 ± 0.4 ^b	7.70 ± 0.3 ^a
Albumin (g 100 mL ⁻¹)	2.70 ± 0.2 ^b	3.10 ± 0.4 ^a
Globulin (g 100 mL ⁻¹)	4.10 ± 0.4 ^b	4.60 ± 0.5 ^a
A/G ratio	0.66 ± 0.01 ^b	0.68 ± 0.016 ^a
Glucose (mmol L ⁻¹)	64.00 ± 1.3 ^b	71.00 ± 0.9 ^a
Liver enzymes activity (U L⁻¹)		
AST	82.00 ± 1.5	87.00 ± 0.6
ALT	112.00 ± 2.6	120.00 ± 1.8
Kidney function test (mmol L⁻¹)		
Urea	8.20 ± 0.7 ^b	9.40 ± 0.7 ^a
Creatinine	1.60 ± 0.2	1.70 ± 0.1
Heart function (mmol L⁻¹)		
Cholesterol	59.00 ± 2.5 ^a	46.00 ± 1.7 ^b
Triglycerides	48.50 ± 0.8 ^a	41.00 ± 0.66 ^b
Blood hormone (mmol L⁻¹)		
Cortisol	79.30 ± 3.7 ^a	68.80 ± 1.8 ^b
Testosterone	1.60 ± 0.2 ^a	1.10 ± 0.3 ^b
T ₃	1.40 ± 0.1 ^a	0.90 ± 0.2 ^b
T ₄	3.20 ± 0.4 ^a	2.70 ± 0.5 ^b
Mineral level (g kg⁻¹)		
Calcium	2.20 ± 0.2 ^b	2.80 ± 0.3 ^a
Phosphorous	1.80 ± 0.1 ^b	2.30 ± 0.2 ^a

^{a,b}Means with different superscripts within a row differ significantly ($p < 0.05$).

AST: Aspartate transaminase, ALT: Alanine aminotransferase

Blood parameters: Results in Table 2 shown that, the total protein, albumin and globulin in addition to the A/G ratio, glucose concentrations and urea levels increased significantly ($p < 0.05$) when 40 g active dried yeast (containing 10⁹ CFU *S. cerevisiae* per gram) was added to the diet of the camels; however, there was no significant impact on the AST and ALT activities and creatinine level with the treatment.

Findings suggest decreases in serum cholesterol, triglycerides, cortisol and testosterone concentrations, while the T₃ and T₄ levels increased significantly in yeast-fed rut camels in comparison with controls (Table 2).

In addition, yeast supplements increased the serum calcium and phosphorous concentrations to a significantly higher level ($p < 0.001$) than controls, as shown in Table 2.

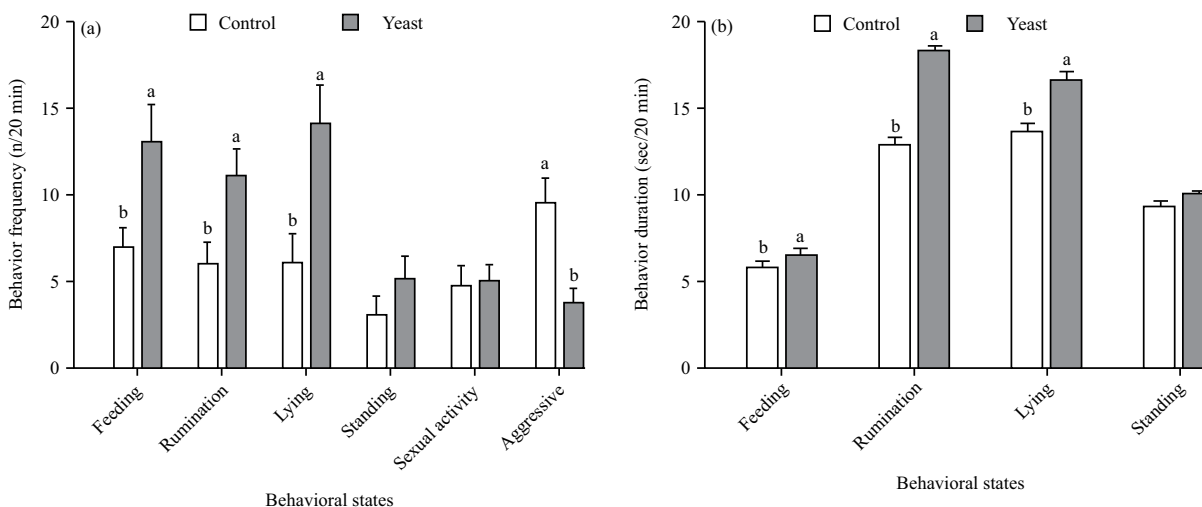


Fig. 1(a-b): Behavioral states in male Dromedary camels during rut season

(a) Frequency of the different behavioral states ($n\ 20\ min^{-1}$) in male Dromedary camels in control and yeast-fed camels during rut season and (b) Duration of the different behavioral states ($sec\ 20\ min^{-1}$). Different superscript letters indicate significant differences ($p < 0.05$)

DISCUSSION

This experiment evaluates whether yeast products can positively affect behavioral, performance and biochemical parameters in Dromedary camels during rut season. The findings together showed that when camels get live yeast, they draw their meals closer together and eat more frequently. In support of current hypothesis, yeast influenced camel feeding behavior during rut season. When supplemented, camels prefer to have six more feeding sessions a day than when they have no added yeast. These results are similar to Kroger *et al.*²⁹, who found that feeding duration was increased with live yeast supplementation than with no yeast and that interval between meals in yeast-supplemented cows was shorter than in non-supplemented ones. This is similar to a finding documented by Kayser *et al.*³⁰, who found that dairy bull calves raised with live yeast had a higher frequency of feed trough visits while eating smaller quantities per visit. The increased duration of eating by addition of yeast agreed with Alzahal *et al.*³¹, who reported that across ruminant species, yeast supplementation increased Dry Matter Intake (DMI) by $0.44\ g\ kg^{-1}$ of BW. In comparison, Stadler *et al.*²⁵ did not find any differences in feeding time or duration as a consequence of a lack of effect on ruminal propionate production. The higher fiber digestibility, associated with live yeast addition, can help speed up feed movement and thus increase appetite and feed intake. The decrease in intervals among the foods as reported by Bach *et al.*³² may also explain why yeast addition can also help regulate the pH of rumen if translated into higher meal frequencies³³.

The current study findings suggest a positive effect of living yeast supplementation on ruminal duration time and frequency as camels tended to ruminate for $54\ sec\ 20\ min^{-1}$ longer when they were yeast-supplemented. Given that the frequency of meals with yeast supplementation has increased, this may lead to more frequent rumination. Also, yeast fed cows appears to spend an additional 25.4 min ruminating during the day³⁴. The live yeast supplement has been used to increase feeding activity in addition to improving rumination with reduced ruminating times³⁵. Compared to these findings, even with yeast supplementation, feeding and ruminating activity did not alter between groups²⁵.

Perhaps changes in feeding behavior and time spent ruminating might be related to the rate and site of digestion of starch, in particular. If *S. cerevisiae* influences the site of starch digestion³⁶, such changes will result in less release of propionate, a powerful hypophagic compound in ruminants that alters feeding behavior³⁷ and could affect how dairy cows respond to diets supplemented with yeast products.

As with more time spent eating, the camels with the yeast-fed diet spent more time lying down frequently, but the time and frequency of standing were not affected in camels receiving yeast supplementation. The findings were consistent with previous research³⁵, which concluded that live yeast yields positive results for various reproductive and welfare indicators, including a longer lying time when yeast was added to the daily diet, 30 days before calving. In comparison, the results were inconsistent with DeVries and Chevaux³⁴, who noticed no effect on cows' lying time or frequency when *S. cerevisiae* was added to the diet.

Contradictory results regarding sexual activity, yeast had no impact on the sexual behavior score. Even though McRobert³⁸ has shown that the presence of yeast in the diet increases the chance of mating in *Drosophila melanogaster*. Starmer *et al.*³⁹ found that males of *Drosophila buzzatii* that fed on yeast and grew had a higher number of mating episodes than those with un-supplemented diets. The presence of yeast in virgin *D. melanogaster* females' diet increases the possibility of copulating and eliciting more attempted copulations per min⁴⁰.

Data from this study showed that the frequency of aggression in yeast-fed camels was lower than in the control group. The present experiment supports a horse experiment in which *S. cerevisiae* exerts a calming effect on behavior⁴¹. In contrast, Commun *et al.*⁴² confirmed that yeast does not affect behavioral aspects, including lying, standing and aggressive behavior in sheep.

The leftover score between the yeast-fed camels and control group differ significantly ($p < 0.05$), which may be due to yeast's effect as a feeding stimulant. Such results were inconsistent with the finding of Stadler *et al.*²⁵ in which the leftover score did not differ ($p > 0.05$) for heifers with or without live yeast included within the feed.

Enhanced feeding activity with yeast addition leads to a significant increase in BCS, BW and BWG in camels as does the increasing growth for bulls with a high-concentration diet supplemented by *S. cerevisiae* active live yeast⁴³. Similarly, Maysoon *et al.*²¹ found significant increases in BW and BWG of dams' and calves' BW in those receiving *S. cerevisiae*-supplemented diets (0.5 g per head per day) compared to control. Kalmus *et al.*⁴⁴ and Al Ibrahim *et al.*⁴⁵ reported contrasting results in which early-lactation dairy cows consume diets with *S. cerevisiae* and BCS was not affected. Similarly, when live yeast (25 g per cow per day) was fed to a group of cows of different parities during early and mid-lactation, Tristant and Moran⁴⁶ found no effect on BCS or BW. Increased performance results may be linked to increased food intake enhanced animal microbial ecological uptake and better weight gains⁴⁷. Yeast and its products caused a significant improvement in the efficiency of beef animals in addition to increased BW⁴⁸. Live yeast works within the rumen to increase fiber digestion; however, the method has not been clearly defined. Cagle *et al.*⁴⁹ propose that yeast cells contain or can produce chemical compounds that directly stimulate the growth of cellulolytic bacteria and resulted in higher non-digestible fiber NDF digestibility and more production of Volatile Fatty Acids (VFAs) for energy⁵⁰.

It should be that due to the fat deposition in the yeast-supplemented bulls, variations in our findings and previous studies may have also been influenced by the increased availability of acetate and butyrate, which are the main lipogenic precursors of adipose tissue in ruminants⁵¹.

Adding *S. cerevisiae* caused a significant increase in the function of liver immunity (total protein, albumin, globulin and A/G) in addition to blood glucose levels. Similarly, Din⁵² reported that total protein, albumin and globulin levels increased significantly after adding *S. cerevisiae* to growing rabbits' diet. At the same time, *S. cerevisiae* caused an improvement in rabbits' appetite and increased their food digestion. Thus, improved dietary utilization leads to an increase in feed intake and contributes to higher levels of blood metabolites. Including *S. cerevisiae* in the diets of lactating buffalo induced an insignificant increase in levels of total protein, albumin, globulin and A/G ratio from 6.64-7.09, 3.30-3.50 and 3.34-3.37g dL⁻¹, respectively, after 10 g hrs⁻¹ day⁻¹ Baker's yeast, this finding contrasts current study⁵³. The reason for the high blood levels of liver immunity function (total protein, albumin and globulin), as observed in the yeast-treated groups can be attributed to yeast-related enhancement of microbial protein synthesis in the rumen. This process thereby increases the population and cellulolytic bacteria activity and enhancement of fiber digestion and lactate use in the rumen and increasing the flow of bacteria. The increased levels of globulin may also be caused by enhanced lipolysis enzyme activity and increased utilization of dietary lipid in yeast preparations⁵⁴.

A major impact of yeast supplementation on glucose concentrations is also consistent with those in the Dromedary camels that showed a substantial rise in blood sugar levels as a result of live yeast supplementation⁵⁵. In contrast to the previous observation, Piva *et al.*⁵⁶ stated that glucose, total protein and blood plasma albumin were not affected by yeast supplementation. It was reported by Saleem *et al.*⁵⁷ that routine introduction of 10 g yeast to lactating vaccine diets did not affect plasma glucose levels in cattle. The controversial results confirmed that the glucose levels in yeast-treated camels were not significantly altered relative to control by Geng *et al.*⁴³. In male Dromedary camels, Morad *et al.*⁵⁸ reported an opposite trend in which the glucose concentration declined significantly in probiotic-treated lambs.

Dietary yeast addition to male camels did not affect liver enzyme activity; these results are consistent with previous findings in which yeast supplementation had no significant

effect on dairy cow AST and ALT levels⁵⁵. On the contrary, in the study of Al saied⁵⁹, a decrease in activity of the active ALT enzyme in native Egyptian bovine animals as a consequence of yeast supplementation and a substantial increase in AST enzyme activity were reported.

Data also showed that urea concentrations for yeast supplemented-groups were higher than the control group with substantial differences ($p < 0.05$); meanwhile, addition of *S. cerevisiae* did not significantly affect blood creatinine concentrations. Similarly, Masek *et al.*⁶⁰ found that higher rates of yeast supplementation led to higher plasma urea-N ($p < 0.05$), while the overall amount of protein in the blood was stable and comparable between groups of 0.1 and 2 g kg⁻¹ concentrated feed mixtures supplemented with yeast in Baladi goats. The findings are consistent with the results recorded for sheep during the milking period from Abd El-Tawab *et al.*⁶¹ and Baiomy⁶². On the other hand, blood components for yeast-added dairy ewes were not substantially different⁶³. Dietary ingredients, dietary nutrient composition and strain and yeast culture dosage caused variations in this study and previous studies.

Based on the results of this study, triglycerides and cholesterol levels showed lower concentrations due to yeast supplementation in camels. These data are consistent with earlier studies by Bakr *et al.*⁵⁵, while an increase in serum triglycerides and cholesterol rates were recorded by Khayyal *et al.*⁵³ with unchanged cholesterol levels as reported by Galip⁶⁴. This decrease in triglyceride and cholesterol levels may be caused by an increase in lipid metabolism⁶⁵ or a higher fibrolytic active effect induced by yeast promoting lipid synthesis⁶⁶, cholesterol synthesis inhibition or direct cholesterol assimilation⁶⁷.

Furthermore, camels exhibit decreased levels of hormones, such as cortisol, testosterone, T₃ and T₄ after addition of yeast to the diet. Including *S. cerevisiae* to the diet concurrently improved the appetite and increased food digestion, thereby enhancing dietary use that led to increased feed intake, which allowed the production of blood metabolites. Accordingly, *S. cerevisiae* has beneficial effects on protein and fat metabolism in addition to thyroid hormone secretions and no harmful effects on liver; kidney and heart function as stated in the previous results.

In the current study, the trend for cortisol, T₃ and T₄ levels in Dromedary camels following yeast supplementation are consistent with data previously obtained by Mostafa *et al.*⁶⁸. On the contrary, the plasma levels of T₃ and T₄ were not affected by yeast addition in the studies of Alsaied⁵⁹ and

Ghoneim and Moselhy⁶⁹. However, the levels of T₃ and T₄ hormones increased significantly with the addition of active dry yeast to the growing rabbits' diet⁵³.

Testosterone results were similar to those from a study by Cha *et al.*⁷⁰, who recorded that that zinc-enriched yeast *S. cerevisiae* strains could play an important role in sperm physiology as a result of the marked elevation of serum testosterone concentration. Similar testosterone patterns were previously observed in Se-enriched yeast supplemented diet with improved testosterone levels in testis tissue and serum and promotion of the expression of testosterone-related genes in goats⁷¹. On the other hand, other studies revealed no significant differences for the concentration of testosterone after yeast supplementation in lambs⁷².

Calcium levels and P in camels that were supplemented with *S. cerevisiae* in their rations were elevated, a finding that was consistent with the Dolezal *et al.*⁷³, who reported that Ca levels were elevated in dairy cows supplemented with yeast culture in the rations, whereas no changes in electrolyte indices in transition cows resulting from the supplementation with fermented yeast products were noted⁷⁴. High Ca and P levels may be due to improved digestibility and mineral absorption as stated by Pisoni *et al.*⁷⁵.

The variations between some previous studies and the findings of this study may be due to feeding strategies, environmental factors, diet composition, type of forage, yeast type and dose. Moallem *et al.*⁷⁶ have reported that yeast products might be more stress-efficient than in normal conditions.

CONCLUSION

Results have shown that the inclusion of *S. cerevisiae* as a feed additive to regular diets of male camels is effective for ingestive behavior and improvement in some blood parameters during their rut period. Yeast addition appears to cause an improvement in the immune responses of male camels as represented by total protein, albumin and globulin. It is therefore recommended that yeast be added to the regular diet of rut camels in order to improve their health.

SIGNIFICANCE STATEMENT

This study discovered yeast (*Saccharomyces cerevisiae*) addition could be beneficial for male Dromedary camels' behaviors and specific blood parameters during the rutting period by potentiating immune responses as represented by

total protein, albumin and globulin. This study will help the researchers uncover the critical areas where changes in behaviors and blood parameters may occur that many researchers could not explore. Thus a new theory on behavioral and blood parameters changes in male camels triggered by dietary yeast addition in the management field may arrive.

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