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Research Article
Productive Performance of Lactating Buffaloes Fed Ration Containing Date Seed and Fibrolytic Enzymes

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Abstract

Background and Objective: Utilization of date seeds (processed date by product) as a feedstuff in diets of farm animals are being in spotlight, this study were carried out to investigate the effect of feeding diets contain cracked date seed with or without fibrolytic enzyme, versus control diet using Egyptian buffaloes. **Materials and Methods:** Fifteen multiparous lactating Egyptian buffaloes (600±30 kg BW) were randomly assigned for 90 days in a completely randomized experimental design. Buffaloes were randomly assigned to 3 groups and fed a basal diet of concentrates, Egyptian clover and rice straw in a ratio of 50:30:20 DM basis (T1), the second group fed (T2) concentrate feed mixture, cracked date seed, Egyptian clover and rice straw as 35:15:30:20, respectively and the third group fed as T2 diet plus fibrolytic enzyme. **Results:** T2 groups had reduced feed intake ($p>0.09$) and DM, OM, NDF and ADF digestibility ($p<0.05$) than control (T1). While, T3 improved fiber digestion (NDF and ADF) compared with T2, with no differences with control (T1). Similarly, T2 resulted in lower ($p<0.05$) daily milk yield, energy corrected milk and milk efficiency ($p<0.05$) compared with T1, whilst, T3 improved the milk yield and ECM and milk efficiency compared with T2 ($p<0.05$) but without differences with T1 ($p<0.05$). **Conclusion:** It could be concluded that using cracked date seed with fibrolytic enzymes in lactating buffalo's diet improved feed conversation and productive performance with no negative effect on animal health.

Key words: Date seed, fibrolytic enzymes, buffalo, nutrients digestibility, milk yield

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

INTRODUCTION

Arid animals' producers suffer from covering their animal's nutritional requirements which led to increase nutritional cost by importing concentrate feeds. So, recently many research studies concerned on utilizing the local agricultural by-products as feedstuffs for ruminant animals[1]. In arid and semi-arid regions, dates are common and main food. Date seed are one of the main by-products can be utilized efficiently by ruminant animals as non-traditional feedstuff. Estimation of date production according to FAO[2] approximately 7.6 million t, average estimates showed that date stones mass ranging 10-15% of total date fruit mass[3]. Utilization of date stone as feedstuff in ruminant diets has been investigated and cleared potential effectiveness in ruminant performance and digestion.

One of the disadvantages of inclusion of date seed in diets, its hard seed coat that makes it difficult to be chewed and digested[4]. Thus different studies were carried out to investigate different treatments to increase nutritive value. Grinding is one of several effective treatments to increase nutrient availability of date seed. Also, previous evaluation of chemical composition of date stones showed its high contain of fiber as 66.1% NDF, 45.5% ADF[4] and low grade protein (up to 8% of DM)[5].

Soliman *et al.*[6] concluded that replacing corn with date seeds by 40 and 60% increased the digestibility coefficients of feed nutrients, daily gain and economic efficiency of lambs. In another study on lactating goat, Al-Suwaiegh[4] suggested that using date seed to replace 20% of concentrate of lactating arid goat diets had no negative effects on their productive performance. Also, Azzaz *et al.*[7] illustrated that replacing 50% of corn grains by powdered date seeds had no negative impact on performance of lactating Nubian goats. In other study illustrated that ground date seeds could replace up to 30% of the ground maize in concentrate feeds of sheep without any adverse effects[5].

Increasing the nutritive value of highly fiber feedstuffs through adding fibrolytic enzymes especially cellulase enzyme were highly investigated. It well stated that fibrolytic enzymes play a direct role in animals feeding by improved digestion in ruminants[8]. Abd El Tawab *et al.*[9] reported that supplementing diets contains date palm fronds with cellulase enzyme had enhanced nutrients digestibility.

Azzaz *et al.*[7] reported that supplementing diets containing powdered date seeds with cellulase enhanced rumen fermentation, nutrients digestibility by Rahmani sheep, also increased milk yield of lactating Nubian goats. So, this study aimed investigate the inclusion of cracked date seed with fibrolytic enzymes in lactating buffaloes' diets on feed utilization and productive performance of lactating Egyptian buffaloes.

MATERIALS AND METHODS

Animals' managements and experimental diets: Fifteen homogenised multiparous lactating Egyptian buffaloes (3±1 parity) in early lactation were assigned randomly to three experimental groups (5 animals each) with live body weight (600±30 kg): Buffaloes in the first group were fed diet containing (per kg DM) 500 g concentrate feed mixture, 300 g Egyptian clover (*Trifolium alexandrinum*) and 200 g rice straw (T1) to meet nutrients requirements according to Jayanegara *et al.*[10], the second group fed (per kg DM) 350 g concentrate feed mixture, 150 g cracked date seed, 300 g Egyptian clover and 200 g rice straw (T2) and the third group fed as T2 diet plus fibrolytic enzyme (2 g kg^{-1} DM which contains10358 IU of cellulase[11] and 1732 IU (T3) (Table 1). The experiment extended for 90 days started one week post parturition. Buffaloes were fed twice daily at 07:00 and 15:00 h. The animals were kept in a free stall and had free access to water.

Feed intake and nutrient digestibility: Feed intake was daily recorded. At the last week of the first, second and third months of the experiment, samples of feces were collected to determine nutrient digestibility using acid insoluble ash as an internal indigestibility marker and coefficients of digestion were calculated according to Ferret *et al.*[12], samples were collected from each animal of experimental groups twice daily at 07:00 and 15:00 h, dried at 60°C in a forced-air oven for 48 h. Dry matter of feed and feces samples was determined by drying at 105°C for 48 h nitrogen (method 954.01). Ether extract (method 920.39) according to AOAC[13] official methods. Neutral detergent fiber (NDF) was determined by the procedure of Van Soest *et al.*[14]. Acid Detergent Fiber (ADF) was analyzed according to AOAC[13] (method 973.18). Organic matter (OM = 100-ash) were calculated[15].

Sampling and analysis of blood plasma: Blood samples were collected from the jugular vein at days 30, 60 and 90 of the experiment, 10 mL of blood samples were taken 4 h after feeding from animals of each treatment by heparinized syringe. Samples were centrifuged at 4000 ×g for 20 min; collected plasma was immediately transported to laboratory and freezed at -20°C until analyzed. By using specific kits (Stanbio Laboratory, Boerne, Texas, USA) and following manufacturer instructions, plasma concentrations of total

protein, albumin, urea, total lipids, glucose, aspartate aminotransferase (AST) and alanine aminotransferase (ALT) were determined spectrophotometrically using T80 UV/VIS Spectrometer, PG Instruments Ltd., UK.

Milk sampling and milk composition: Animals were milked twice daily at 07:00 and 16:00 h, samples were collected at each milking. Milk yield was recorded daily. Samples were analyzed for total solids, fat, protein, lactose, solids non-fat using infrared spectroscopy (Bentley 150, Infrared Milk Analyzer, Bentley Instruments, USA)[16]. Energy-corrected milk (ECM) was calculated according to Sjaunja *et al.*[17]:

$$\text{ECM (kg/day)} = \text{Milk (kg/day)}\times[38.3\times\text{fat (g kg}^{-1})+24.2\times \text{protein (g kg}^{-1})+16.54\times\text{lactose (g kg}^{-1})+20.7]/3140$$

Statistical analysis: Data were statistically analyzed according to a completely randomized design using GLM procedure of SAS software (Version 9.2). Significant differences between means of treatments were carried out by the Duncan's test and the significance threshold was set at $p<0.05$.

RESULTS

Chemical composition of date seed: Results of chemical analysis showed that date stone high in NDF, ADF and low in CP (Table 1). The chemical composition of the experimental rations reflected no differences in CP, NDF, ADF and EE.

Nutrients digestibility: Digestibilities of NDF ($p = 0.029$) and ADF ($p = 0.01$) were higher for T1 and T3 than T2. Also, T1 recorded higher values in DM ($p = 0.034$) and OM ($p = 0.0143$) compared with other treatments. While there were no differences between treatments on CP and EE digestibilities ($p>0.05$) (Table 2).

Feed intake, milk yield and composition: No differences ($p>0.05$) were recorded in feed intake between experimental rations.

Higher daily yields of milk ($p = 0.049$) and ECM ($p = 0.0016$) were observed with the control group (T1) compared with T2 (Table 3) but no differences ($p>0.05$) were observed between T1 and T3. The milk content of total solids, protein, lactose and ash were not affected ($p>0.05$) by the treatments. Conversely, greater daily outputs of solids non-fat ($p = 0.042$) were noted with diet contained date seed (T2) compared with control (T1) but there was no difference ($p>0.05$) between T2 and T3. The milk daily yields of total solids recorded the highest value in T1 ($p = 0.0091$) compared with T2 and T3. Also T1 recorded higher yield in milk fat comparing with T2, with no difference with T3. Other milk constituents solids non-fat, protein and lactose did not recorded differences among treatments. Milk efficiency expressed as milk yield/feed intake was greater ($p<0.05$) with control (T1) than for other groups T2 and T3. Also, ECM yield/feed showed a high efficiency in T1 compared with T2 ($p>0.05$), otherwise there was no difference between T3 with other treatments.

Blood chemistry measurements: The inclusion of date seed in experimental ration (T2 and T3) did not affect ($p>0.05$) concentrations of plasma glucose, total protein, total lipids, albumin, globulin, AST and ALT concentrations (Table 4).

Table 1: Chemical composition (g kg^{-1} of DM) of concentrate feed mixture, date kernel, rice straw and Egyptian clover

Parameters	Concentrate	Clover Hay	Rice straw	Date stone	T1	T2	T3
DM	908.8	921.7	920.3	899.3	914.9	914.2	914.2
OM	900.8	861.6	832.2	989.2	875.3	881.9	881.9
CP	136.6	137.7	29.0	49.5	115.4	108.9	108.9
EE	34.5	35.4	23.2	64.3	32.5	34.7	34.7
NDF	291.4	205.9	377.9	371.1	283.1	289.0	289.0
ADF	155.2	147.5	228.5	283.0	167.6	177.1	177.1

Table 2: Nutrient digestibility (DM %) of experimental diets

Parameters	T1	T2	T3	SEM	Pr>
DM	662.9^{a}	601.2^{b}	595.9^{b}	1.11	0.034
OM	615.2^{a}	558.1^{b}	541.1^{b}	1.02	0.0143
CP	609.0	624.8	605.3	1.79	0.899
NDF	683.4^{a}	496.6^{b}	600.2^{a}	3.87	0.029
ADF	631.0^{a}	525.2^{b}	605.3^{a}	4.15	0.01
EE	697.0	650.4	649.9	5.10	0.105

Table 3: Body weight, feed intake, milk yield and composition of buffaloes fed experimental diets

Parameters	T1	T2	T3	SEM	Pr>
Milk yield (kg/day)	9.44[a]	7.81[b]	8.04[ab]	0.316	0.049
ECM (kg/day)	13.27[a]	9.50[b]	11.10[b]	0.534	0.0016
Total solids (%)	16.63	16.16	16.71	0.365	0.82
Solids non-fat (%)	9.69[b]	10.94[a]	10.01[ab]	0.222	0.042
Fat (%)	6.94	5.22	6.70	0.373	0.1178
Protein (%)	3.50	4.05	3.53	0.115	0.077
Lactose (%)	5.49	5.89	5.78	0.095	0.21
Total solids (g/day)	1569.18[a]	1259.82[b]	1335.10[b]	49.61	0.0091
Solids non-fat (g/day)	914.32	850.42	805.90	26.55	0.274
Fat (g/day)	654.88[a]	409.40[b]	529.23[ab]	36.81	0.0055
Protein (g/day)	330.16	313.10	282.48	9.65	0.139
Lactose (g/day)	518.26	449.43	467.23	15.94	0.175

Table 4: Blood plasma measurements of buffaloes fed experimental diets

Parameters	T1	T2	T3	SEM	Pr>
Glucose (mg dL^{-1})	68.00	68.00	71.00	1.71	0.76
Protein (g dL^{-1})	4.55	4.31	4.19	0.22	0.80
Albumin (g dL^{-1})	2.11[b]	2.49[a]	2.32[ab]	0.061	0.03
Globulin (g dL^{-1})	2.44	1.81	1.55	0.227	0.27
A/G ratio	2.11	2.49	2.32	0.57	0.36
Urea (mg dL^{-1})	38.00[c]	52.00[b]	61.00[a]	2.56	0.0001
AST	23.00[a]	17.00[b]	27.00[a]	1.22	0.0026
ALT	18.00	14.00	20.00	1.203	0.11
Total lipids (mg dL^{-1})	553.00[b]	531.00[b]	615.00[a]	10.66	0.0014

DISCUSSION

Chemical composition of date seed showed high contents of fiber fractions NDF and ADF. These values were lower that reported by other studies[4,18,19]. These variations in fiber contents components could be related to date species variety, environmental conditions, season, geographical zone and pedological conditions[20-22].

The experimental diets used in this study were similar in chemical composition. Replacing concentrate with date seed led to significant decrease ($p<0.05$) on DM, OM, NDF and ADF digestibility, the reduction of digestibility could be reflect the difference between concentrate non-structural carbohydrates content and the increase of structural carbohydrate in date seed. The increase of fiber contents of date seed may contribute the reduction of digestibility in the rumen due to a limited availability of fermentable OM in the rumen[23]. Also, cracked date seed to small particles may increase passage rate, in addition fibre from non-forage sources often ferments and passes rapidly from the rumen. As a consequence they are less effective at stimulate chewing[24]. In another studies reviewed the anti-nutritional components (oxalate, tannin, flavonoids, saponin) in date palm[25] may act to reduce nutrient intake, digestion, absorption and utilization and may produce other adverse effects[26].

Supplementing diets contain date seed with fibrolytic enzymes (cellulase) showed an enhancement in fiber digestion (NDF and ADF), the increase of diet NDF and ADF contents in the experimental diets due to inclusion of date seed could explain the reduction of OM digestibility in T_2 and T_3 compared with T_1. Cellulase and hemicellulose enzymes hydrolyze the glycosidic bond between carbohydrates or between a carbohydrate and a non-carbohydrate molecule[27]. Hydrolysis of the glucoside results in the formation of a sugar and another compound and the 'hydrolase' signifies that C-O, C-N or C-C bonds can be broken during hydrolysis. The enzymes used in the current experiment are produced from ruminal micro-organism[11]. Efficient breakdown of cellulose in the rumen usually requires a number of glycosyl hydrolases enzymes including endoglucanases (endo-1,4-β-D-glucan hydrolase, EC 3.2.1.4), exoglucanases (exo-1,4-β-D-glucan cellobiohydrolase, EC 3.2.1.91) and β-glucosidases (β-D-glucosidase, EC 3.2.1.21), which work synergistically to hydrolyze cellulose[28]. It well established that rumen fibrolytic micro-organisms (bacteria and fungi) had the higher capacity to breakdown the cellulytic materials due to its capability to produce complete glycosyl hydrolases enzymes, so supplanting diets with fibrolytic enzymes produced from ruminal bacteria should enhance fiber digestion.

The current blood metabolites results showed that all values were within the normal reference ranges[29]. Feeding date seed with or without fibrolytic enzymes did not affect plasma glucose, total protein, total lipids, albumin and globulin suggesting good potential effect of date seed as alternative feedstuff. Values of liver enzymes, AST and ALT were within normal physiological ranges revealing normal liver activity and function. The result suggests no liver pathological lesions[30].

Recorded values of dry matter intake showed no differences between treatments, these observations agreed with Azzaz *et al.*[7]. Also, reflect an acceptable palatability of cracked date seed in diets[4]. Replacing concentrate with cracked date seed (T2) reduced daily yields of milk (by about 18%) and ECM (about 28%) compared with control (T1), without affecting milk total solids, fat, protein and lactose contents. The decrease in milk production in T2 might due to the reduction in DM, OM, NDF and ADF digestibility. Also, reflected as reduction in milk efficiency T2 by about 12 and 25% (as milk yield/feed intake and ECM yield/feed intake, respectively). While supplementing diets with fibrolytic enzymes enhanced fiber (NDF and ADF) digestion which cleared as improve in milk production and efficiency (milk yield/feed intake and ECM yield/feed intake). Supplementing diets with fibrolytic enzymes improve fiber digestion and energy status, which could influence milk yield and composition[31]. The enhancement in milk production and feed efficiency observed in T3 compared with T2 might be attributable to greater NDF digestibility in the rumen and the similar trend was concluded by Azzaz *et al.*[7]. Improvements in feed conversion efficiency were due to improvement in milk yield. Improved feed efficiency indicates better utilization of nutrients when diet contain cracked date seed was supplemented with fibrolytic enzymes, finally, Improved production of milk yield in T3 compared with T2 caused by addition of enzymes was most likely a result of improved fiber fractions digestibility rather than a change in feed intake, adding fibrolytic enzymes to diet contained cracked date seed make the utilization of that diet similar to control diet (T1) did not contain cracked date seed.

CONCLUSION

Under the conditions of the present study, using the cracked date seed in the diet of lactating buffaloes could reduce the utilization of nutrients and feed efficiency and milk production. While supplementing diet contain cracked date seed with fibrolytic enzymes improved animal performance and milk production and feed efficiency. It could be concluded that inclusion cracked date seed with adding fibrolytic enzymes do similar effect on milk yield, with no negative effect on the health status of buffaloes.

SIGNIFICANCE STATEMENT

This study showed that inclusion of cracked date seed in the diet of lactating buffaloes could be alternative feedstuff for corn especially with supplementing diets with fibrolytic enzymes with no negative impacts on productive performance of lactating buffaloes.

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