

Prickly pear meal (*Opuntia ficus-indica*) in the feeding of lactating cows

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Abstract

The objective of this study was to evaluate the effects of increasing levels of inclusion of prickly pear meal (PPM) (*Opuntia ficus-indica*) in diets for lactating dairy cows on nutrient intake, apparent digestibility, milk yield and composition, milk fatty acid profile, and ingestive behavior. Fifteen lactating Holstein × Zebu crossbred cows were assigned to three 5 × 5 Latin squares and fed diets containing 0 (CON), 3.7, 7.4, 11.1, or 14.8% PPM on a dry matter basis. The inclusion of PPM increased NDFap intake ($P = 0.037$) compared with the CON diet and promoted a linear reduction in ether extract intake ($P < 0.001$). Apparent digestibility of ether extract ($P < 0.022$) and TDN ($P = 0.034$) decreased linearly as PPM inclusion levels increased. Milk yield showed a linear reduction with increasing levels of PPM ($P = 0.031$), as did protein yield ($P = 0.023$), lactose yield ($P = 0.022$), and solids-not-fat yield ($P = 0.022$). The milk fatty acid profile was significantly altered, with linear increases in C16:0 ($P < 0.001$) and total saturated fatty acids ($P = 0.004$), as well as reductions in C18 fatty acids ($P < 0.001$), total unsaturated fatty acids ($P = 0.004$), and the unsaturated:saturated fatty acid ratio ($P = 0.007$). Regarding ingestive behavior, the inclusion of PPM reduced the number of rumination periods ($P = 0.036$) and the number of ruminated boluses per day ($P = 0.046$). The inclusion of prickly pear meal in diets for lactating dairy cows negatively alters dietary energy density, reduces milk production, and modifies the milk fatty acid profile by increasing the proportion of saturated fatty acids. Although prickly pear meal represents a viable feed alternative in semi-arid regions, its inclusion must be carefully adjusted to avoid impairments in productive performance.

Introduction

Milk production in tropical and semi-arid regions requires nutritional strategies capable of sustaining the performance of lactating cows under limitations in forage availability and quality, as well as increasing costs of energy concentrates (Souza et al. 2019). This scenario reinforces the need to identify alternative feed resources that ensure adequate nutrient supply and productive efficiency (Silva et al. 2024).

Among plant species used in ruminant nutrition, forage cactus (*Opuntia ficus-indica*) stands out for its high biomass production capacity and physiological efficiency, characteristics associated with crassulacean acid metabolism (CAM), which confers greater water-use efficiency and tolerance to adverse environmental conditions (Heyduk 2022; Sipango et al. 2022). In addition, forage cactus exhibits high biomass yield under semiarid conditions, with substantial dry matter accumulation over multi-year production cycles (Costa et al. 2024), adaptation to low-fertility soils (Sipango et al. 2022), and a nutritional composition characterized by high concentrations of non-fibrous carbohydrates (536 ± 85.1 g/kg of DM) (Siqueira et al. 2022), associated with good dry matter digestibility (Silva et al. 2024).

These characteristics make forage cactus a promising ingredient for formulating diets for lactating cows with moderate milk production levels (approximately 10.4 kg/cow/day), whose nutritional requirements are compatible with the use of alternative feeds (Rashid et al. 2024). However, despite its recognized nutritional potential, the use of forage cactus in natura presents limitations related to its high moisture content (850–950 g/kg), low dry matter density, and reduced physical effectiveness of fiber (Siqueira et al. 2022; Godoi et al. 2024), factors that may restrict dry matter intake, compromise ruminal stability, and hinder diet standardization (Sipango et al. 2022; Silva et al. 2024). Moreover, the chemical composition of fresh forage

cactus may vary according to species, cladode age, and cultivation conditions, imposing additional challenges to nutritional control in productive and experimental systems.

In view of these limitations, the use of prickly pear meal emerges as a technically relevant alternative, as it concentrates dry matter, reduces compositional variability, and facilitates storage, transport, and dietary inclusion. However, compared with in natura use, the effects of prickly pear meal on productive performance and metabolism of lactating cows remain poorly documented.

In this context, it is hypothesized that increasing levels of inclusion of prickly pear meal, due to its higher nutrient concentration, may promote increased nutrient intake and digestibility, enhance microbial protein synthesis, and maintain productive performance of cows, potentially resulting in responses that indicate a nutritionally more appropriate inclusion level.

Thus, this study investigated the effects of increasing dietary inclusion of prickly pear meal (*Opuntia ficus-indica*) on nutrient intake and digestibility, nitrogen balance, microbial protein synthesis, productive performance, milk fatty acid composition, and ingestive behavior in lactating cows.

Material and methods

Ethics and Localization Committee

Animal handling and experimental procedures complied with national and international ethical regulations and received approval from the Animal Ethics Committee of the State University of Southwest Bahia (protocol 169/2017).

The trial was conducted on a commercial farm situated in Macarani-BA, Brazil (15°33'52" S; 40°25'15" W), at 311 m above sea level.

Animals, experimental design and diets

Fifteen Holstein × Zebu crossbred cows were used (mean days in milk = 90 ± 38 days, in the fourth or fifth lactation, body weight (BW) of 472 ± 9.65 kg, and milk yield of 12 ± 1.83 kg/day). Animals were assigned to three 5×5 Latin squares with five experimental periods. Each period lasted 21 days, with the first 16 days allocated for adaptation and the remaining 5 days for data collection.

Five dietary treatments were evaluated, corresponding to increasing inclusion levels of prickly pear meal (PPM; *Opuntia ficus-indica*) in the total mixed ration. The experimental diets were formulated as follows: a control treatment without PPM inclusion (CON) and four diets containing 3.7, 7.4, 11.1, or 14.8%PPM, expressed on a dry matter (DM) basis.

Sugarcane (*Saccharum officinarum*), cultivar RB 72454, was used as the forage component of the diets throughout the experimental period. During the adaptation phase, all cows received sugarcane supplemented with 0.5% of a urea and ammonium sulfate mixture (9:1, wt/wt), whereas in the experimental phase the inclusion level of this mixture was increased to 1%. Diets were formulated to meet the nutritional

requirements for maintenance and a milk yield of 15 kg/day and to be isonitrogenous, according to NRC (2001) (Table 1).

Each animal was maintained in an individual 19-m² pen containing sand bedding and equipped with a feeder, with water available ad libitum.

Nutrient intake and apparent digestibility of the total tract

Feed was offered twice daily (07:00 and 15:00 h) on an ad libitum basis, with orts maintained between 50 and 100 g/kg. From days 17 to 21 of each experimental period, samples of feed ingredients and orts were collected, frozen at -20 °C, and later used for laboratory analyses. Orts were sampled daily and subsequently pooled to obtain a composite sample per animal and per period.

BW was recorded at the start of the trial and at the end of each experimental period to monitor weight changes and adjust dietary supply when necessary. Fecal sampling was carried out between days 18 and 20 of each period at 9-h intervals, with samples obtained directly from the rectum or immediately after spontaneous defecation. Following collection, fecal material was preserved and combined to form a composite sample for each animal within each period.

Fecal output was estimated using indigestible neutral detergent fiber (i NDF) as an internal marker, following the methodology described by Detmann et al. (2012).

Sample collection and laboratory analysis

Samples of feed ingredients, orts, and fecal material were first dried in a forced-air oven at 55 °C for 72 h and then ground using a Wiley-type mill equipped with a 1-mm screen. Chemical analyses were performed to determine DM (method 930.15), ash (method 942.05), crude protein (CP; method 984.13), and ether extract (EE; method 920.39), following the procedures recommended by AOAC (2000).

Fiber fractions were quantified as neutral detergent fiber corrected for ash and protein (NDF_{ap}), according to the procedures proposed by Mertens (2002) and Licitra et al. (1996), using heat-stable α -amylase without the inclusion of sodium sulfite, with analyses conducted in a fiber analyzer (Ankom Tech Corp., Fairport, NY, USA).

Non-fibrous carbohydrate (NFC) concentration was estimated following the approach described by Hall (2000). Dietary energy values, expressed as total digestible nutrients (TDN) and metabolizable energy (ME), were calculated based on NRC (2000) recommendations.

Nitrogen balance and excretion of purine derivatives

Blood samples were collected on the last day of each experimental period from the mammary vein into heparinized vacuum tubes. Plasma was obtained by centrifugation and stored at -20 °C for subsequent determination of urea concentration.

Spot urine samples were collected on day 21 of each period, approximately 4 h after feeding, during spontaneous urination. Aliquots were preserved with sulfuric acid for analyses of urea, allantoin, and uric

acid, while undiluted samples were stored for determination of total nitrogen and creatinine.

Urea concentrations in plasma, urine, and milk, as well as urinary creatinine and uric acid, were quantified using commercial kits (Bioclin®), whereas allantoin in urine and deproteinized milk was determined by a colorimetric method (Chen and Gomes, 1992). Urinary volume was estimated from spot urine samples based on daily creatinine excretion (24.05 mg/kg BW; Chizzotti et al. 2007).

Total purine excretion was calculated as the sum of urinary allantoin and uric acid plus milk allantoin. Absorbed microbial purines and intestinal flow of microbial nitrogen were estimated using equations proposed by Verbic et al. (1990) and Chen and Gomes (1992), respectively. Microbial crude protein synthesis and its efficiency were calculated based on total digestible nutrient intake.

Nitrogen balance was determined as the difference between nitrogen intake and nitrogen excreted in feces, urine, and milk, with total nitrogen analyzed according to AOAC (2000).

Production, composition, and milk fatty acid profile

Cows were milked twice daily (05:00 and 17:00 h) using a mechanical milking system, and individual milk production was continuously monitored with an automated recording device (MMI6®). Mean daily milk yield was calculated using data obtained during the sampling phase, after adaptation to the experimental diets, and production was standardized to 3.5% fat-corrected milk (FCM; Sklan et al. 1992) and energy-corrected milk (ECM) according to NRC (2000).

Milk sampling was performed over four consecutive days (days 18 to 21 of each experimental period). Milk composition (fat, protein, and lactose) was determined by mid-infrared spectrometry (Lactoscan®, Entelbras, São Paulo, Brazil). Additional aliquots were frozen at -20 °C for urea determination or freeze-dried for subsequent analysis of the milk fatty acid (FA) profile.

Milk urea concentration was analyzed using a commercial enzymatic kit (Doles®, Goiânia, Brazil). Milk lipids were extracted and converted to fatty acid methyl esters using sodium methoxide as the transesterification catalyst and acetyl chloride for acidification, with hexane as the solvent. Fatty acid methyl esters were quantified by gas chromatography (Focus GC, Thermo Scientific, Milan, Italy) equipped with a flame ionization detector and a capillary column (SP-2560, Supelco). Injector and detector temperatures were set at 250 °C and 280 °C, respectively, and the oven temperature was programmed from 140 °C to 220 °C at 1 °C/min, with a final hold of 25 min. Hydrogen served as the carrier gas at a flow rate of 1.5 mL/min, and duplicate injections (1 µL) were performed. Fatty acids were identified by comparison of retention times with commercial standards (GLC-674, Nu-Chek Prep, Inc.) and expressed as mg/100 mg of total fatty acids based on peak area normalization.

Eating behavior

Ingestive behavior was assessed through continuous visual monitoring over a 24-h period on day 17 of each experimental phase. Observations were carried out at 5-min intervals by trained personnel positioned to avoid interference with animal activity, and artificial lighting was provided before the observation period to ensure proper acclimation.

Rumination characteristics were evaluated during three daily intervals (10:00–12:00, 14:00–16:00, and 19:00–21:00 h) by recording the number of chewing movements per ruminal bolus and the duration of rumination using a digital stopwatch. Behavioral measurements followed the procedures described by Bürger et al. (2000).

Statistical analysis

Data were analyzed using the MIXED procedure of SAS OnDemand for Academics® (2014) (SAS Institute Inc., Cary, NC, USA), using the following model:

$$Y_{ijkl} = \mu + S_i + A_i(S)_j + P_k + T_l + e_{ijkl}$$

where Y_{ijkl} is the dependent variable, μ is the overall mean, S_i is the fixed effect of Latin square i ($i = 1-3$), $A_i(S)_j$ is the random effect of cow within square j ($j = 1-5$), P_k is the fixed effect of experimental period k ($k = 1-5$), T_l is the fixed effect of diet l ($l = 1-5$), and e_{ijkl} is the residual error.

Treatments corresponded to five levels of inclusion of PPM: control (0%), 3.7%, 7.4%, 11.1%, and 14.8% of total dietary dry matter. Orthogonal contrasts were applied to evaluate treatment effects, including the comparison between the CON and diets containing PPM (4 -1 -1 -1 -1), as well as the linear (0 -3 -1 1 3), quadratic (0 1 -1 -1 1), and cubic (0 -1 3 -3 1) effects of inclusion levels. Differences were considered statistically significant at $P \leq 0.05$, and trends were considered when $0.05 < P \leq 0.10$.

Results

Dry matter intake (DMI) showed a tendency to increase in animals fed PPM compared with the CON diet ($P = 0.070$) (Table 2). Conversely, lower NDF_{ap} intake was observed in animals receiving the CON diet ($P = 0.037$). EE intake exhibited a linear decreasing effect with increasing inclusion of PPM in the diet ($P < 0.001$).

The inclusion of PPM did not affect the digestibility of DM, CP, NDF_{ap} , or NFC ($P > 0.05$). However, EE digestibility showed a tendency to decrease compared with the CON diet ($P = 0.081$), in addition to a linear decreasing effect as PPM inclusion levels increased ($P = 0.022$). TDN showed a tendency for a linear effect with PPM inclusion ($P = 0.068$).

No effect of PPM was observed on nitrogen balance or microbial protein synthesis ($P > 0.05$) (Table 3).

Regarding productive performance, cows fed the CON diet showed a tendency toward higher milk yield (kg/day) compared with those receiving PPM ($P = 0.092$), as well as a linear decreasing effect with increasing levels of PPM in the diet ($P = 0.031$). Similarly, linear decreasing effects were observed for protein yield (kg/day) ($P = 0.023$), lactose yield (kg/day) ($P = 0.022$), and defatted dry extract (DDE) yield (kg/day) ($P = 0.022$), along with a tendency for a linear decreasing effect on total dry extract (TDE) yield (kg/day) in milk ($P = 0.090$). Additionally, a tendency for a quadratic effect was observed for protein (%) ($P = 0.076$), lactose (%) ($P = 0.081$), and DEE (%) ($P = 0.093$) concentrations in milk (Table 4).

Cows fed the CON diet showed higher concentrations of C10:0 ($P = 0.030$), C12:0 ($P = 0.023$), C18:0 ($P < 0.001$), C20:0 ($P = 0.039$), and saturated C18 fatty acids ($P < 0.001$), as well as a tendency toward higher C18:1n7 concentration ($P = 0.076$). In contrast, these cows showed lower concentrations of C16:0 ($P = 0.013$) and total C16 ($P = 0.013$) in milk compared with those fed PPM (Table 5).

Additionally, PPM inclusion promoted linear increases in the concentrations of C16:0 ($P < 0.001$), total C16 ($P < 0.001$), and total saturated FA ($P = 0.004$). Conversely, linear decreasing effects were observed for C18:0 ($P < 0.001$), C18:1n7 ($P = 0.002$), C18:1n9 ($P < 0.001$), C20:0 ($P = 0.025$), FA > C16 ($P < 0.001$), saturated C18 fatty acids ($P < 0.001$), unsaturated C18 FA ($P < 0.001$), total unsaturated FA ($P = 0.004$), and the U/S ratio ($P = 0.007$), as well as a tendency for a linear decreasing effect on C18:2n6 ($P = 0.078$).

Regarding ingestive behavior, cows fed the CON diet showed a greater number of rumination periods ($P = 0.036$) and a higher number of ruminated boluses per day ($P = 0.046$), as well as a tendency toward a lower number of chews per bolus (NCB) ($P = 0.076$) and less time spent per ruminated bolus (TBR) ($P = 0.091$), compared with cows fed PPM. Similarly, a tendency for a linear effect was observed for the number of rumination periods per day ($P = 0.072$), and a tendency for a quadratic effect was observed for total chewing time per day (TCT) ($P = 0.065$) with increasing inclusion of PPM in the diet (Table 6).

Discussion

The increasing inclusion of PPM in sugarcane-based diets promoted gradual and consistent changes in dietary chemical composition, mainly characterized by increased NDF_{ap} content and reductions in non-fibrous carbohydrates, ether extract, and energy density, as evidenced by lower TDN and metabolizable energy concentrations. These changes reflect the progressive replacement of ground corn with an ingredient containing a higher fibrous fraction and lower energetic value, with direct implications for intake regulation, nutrient utilization, and productive performance of lactating cows.

Within this nutritional context, the tendency toward increased DMI observed in cows fed PPM containing diets compared with the control diet suggests an adaptive response to reduced dietary energy density. In dairy cows, adjustments in voluntary intake represent one of the primary compensatory mechanisms to mitigate reductions in metabolizable energy supply, provided that no physical or metabolic constraints to intake are present (Allen 2020). The fiber derived from PPM, despite its high concentration, is characterized by high ruminal fermentability and low physical effectiveness (Alves et al. 2023), which likely minimized rumen fill limitations and allowed this intake adjustment. This response is consistent with the cell wall characteristics of *Opuntia ficus-indica*, which is rich in pectins and highly fermentable polysaccharides that favor faster ruminal passage rate and reduced digesta retention, even under high dietary fiber levels (Gomes et al. 2023).

Although NDFap intake was lower in the control diet due to its reduced fiber content, the progressive inclusion of PPM increased the intake of this fraction without compromising total DMI. Moreover, NDFap intake remained within the range of 1.2 to 1.3% of BW, which is considered adequate for moderately producing cows, as it stimulates rumination and supports ruminal stability without limiting voluntary intake

(Allen 2000). These results indicate that the fiber supplied by PPM, despite its greater quantitative contribution, was functionally compatible with maintaining intake and ruminal activity.

Concurrently, ether extract intake decreased linearly with increasing dietary PPM levels, directly reflecting the lower lipid content of this ingredient compared with corn. In addition, ether extract digestibility also declined linearly, a response that can be attributed to the combination of lower absolute dietary lipid content, greater association of ether extract with the fibrous matrix, and changes in the ingested fatty acid profile. Diets containing a higher proportion of long-chain saturated FA, such as C16:0, tend to exhibit reduced emulsification efficiency and intestinal absorption, thereby decreasing apparent lipid digestibility (Boerman et al. 1998; Rico et al. 2017). Under low-fat dietary conditions, endogenous losses represent a greater relative proportion of fecal ether extract, further contributing to reduced apparent digestibility (Palmquist et al. 2017). In addition, increased dietary fiber content favors physical entrapment of lipids, limiting access of ruminal and pancreatic lipases. Collectively, these factors explain the reduced energetic contribution of ether extract and the consequent linear reduction in total digestible nutrients, despite the maintenance of digestibility of the other dietary fractions.

Despite these changes in diet composition and energy value, the inclusion of PPM did not affect nitrogen balance, plasma, urinary, or milk urea concentrations, nor microbial protein synthesis. These findings indicate that nitrogen metabolism was adequately supported across all treatments, suggesting that synchronization between ruminally degradable nitrogen availability and fermentable energy supply was sufficient to sustain microbial efficiency, even under reduced dietary energy density (Lu et al. 2019).

From a productive standpoint, the linear reduction in milk yield with increasing PPM levels can be physiologically explained by the progressive decrease in dietary energy density, which limited the amount of energy available to the mammary gland. Milk synthesis is highly dependent on energy supply, particularly for lactose production, which acts as the primary osmotic regulator of milk volume secretion (Anger et al. 2024). When energy intake is insufficient, glucose availability to the mammary gland is reduced, thereby limiting lactose synthesis and, consequently, milk yield (Allen and Piantoni 2013). The reduced energetic contribution of ether extract exacerbates this process, given that lipids represent the most energy-dense fraction of the diet (Palmquist et al. 2017).

In addition to effects on milk yield, the inclusion of PPM altered the milk fatty acid profile, characterized by reductions in C10:0 and C12:0, increases in C16:0, and relative decreases in long-chain and unsaturated fatty acids, resulting in a higher total concentration of saturated FA, a lower proportion of unsaturated FA, and a reduced U/S ratio. The reduction in C10:0 and C12:0 is associated with a lower relative availability of lipogenic precursors for de novo synthesis in the mammary gland, such as acetate and β -hydroxybutyrate (Matamoros et al. 2020), as well as with increased supply of preformed FA. Conversely, PPM is rich in C16:0, and the greater dietary supply of this long-chain FA leads to increased intestinal absorption and direct incorporation into milk fat, shifting lipid composition toward C16:0 and increasing total saturated FA (Rocha Filho et al. 2021). This response promotes a relative dilution of unsaturated FA and may also modify ruminal biohydrogenation patterns, resulting in a more saturated milk fat profile, consistent with dietary modulation of milk lipid composition in dairy cows (Gallardo and Teixeira 2023).

From a behavioral perspective, cows fed the control diet exhibited a greater number of rumination periods and a higher number of ruminal boluses per day, behavior consistent with the greater physical effectiveness of sugarcane fiber. The inclusion of PPM reduced these parameters and promoted a tendency toward fewer chews and shorter chewing time per bolus, indicating that, despite the higher NDF_{ap} content, the fiber from PPM presents lower physical resistance and requires less mastication effort. This characteristic is typical of feed ingredients with poorly lignified cell walls and high structural fragility (Zebeli et al. 2012; Heering et al. 2023).

Nevertheless, total chewing time remained adequate, suggesting that PPM inclusion did not compromise ruminal health but rather promoted adjustments in intake and rumination patterns consistent with the physical nature and high fermentability of the supplied fiber.

Conclusion

The increasing inclusion of prickly pear meal increased dry matter intake while maintaining nutrient digestibility and microbial protein synthesis. However, the reduction in dietary energy density limited the productive performance of the cows, indicating that high inclusion levels compromise milk yield. Therefore, prickly pear meal can be used as an alternative feed ingredient, provided that its inclusion level is properly adjusted to balance increased intake with an adequate energy supply.

Declarations

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Competing Interests

The authors have no relevant financial or non-financial interests to disclose.

Author Contributions

All authors contributed to the conception and design of the study. Jacqueline Firmino de Sá: Data curation and Functions/Writing - original version. Fabiano Ferreira da Silva: Conceptualization, formal analysis, methodology, project administration, resources, and validation. Robério Rodrigues Silva: Conceptualization. Jose Esler de Freitas Júnior and Lara Maria Santos Brant: Formal analysis and Writing - review and editing. Paulo De Jesus Santos and Luan Brito Matos: Research. All authors read and approved the final manuscript.

Data Availability

The datasets generated during and/or analyzed during the current study are not publicly available due to [WHY DATA ARE NOT PUBLIC] but are available from the corresponding author on reasonable request.

Ethics approval

This study was performed in line with the principles of the Declaration of Helsinki. Approval was granted by the Ethics Committee of the State University of Southwest Bahia (UESB), under approval protocol 169/2017.

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Tables

Table 1. Percentage of inclusion of ingredients and chemical composition of experimental diets.

Ingredient (g/Kg DM)	Levels of prickly pear meal in the diet (% DM)				
	0	3.7	7.4	11.1	14.8
Sugarcane	653.9	654.3	654.4	654.7	654.6
Ground corn	253.5	215.9	178.6	140.9	103.5
Prickly pear meal ^a	0.00	37.0	74.0	111.0	148.0
Soybean meal	70.2	70.4	70.6	71.0	71.5
Mineral ^b	11.2	11.2	11.2	11.2	11.2
Dicalcium phosphate	8.20	8.20	8.20	8.20	8.20
Limestone	3.00	3.00	3.00	3.00	3.00
Chemical composition (g/kg DM)					
Dry matter	478.7	478.2	477.9	478.0	478.3
Crude Protein	176.0	174.6	173.3	171.9	170.6
NDF _{ap} ^c	404.4	415.2	425.8	436.3	446.8
Ether extract	22.2	21.2	20.2	19.3	18.4
Ash	63.2	69.0	74.8	80.7	86.6
NFC ^d	334.2	320.0	305.8	291.8	277.6
TDN ^e	71.8	70.6	69.3	68.0	66.7
ME (Mcal/kg of DM) ^f	2.59	2.55	2.50	2.46	2.41
FA (g/100 g of FA)^g					
16:0	14.38	13.29	15.92	16.00	17.42
16:1	0.11	0.09	0.12	0.15	0.19
17:0	0.26	0.16	0.21	0.31	0.46
17:1	0.14	0.06	0.09	0.16	0.26
18:0	4.53	3.20	4.36	4.60	4.88
18:1n-9c	48.73	49.45	47.68	44.27	33.80
18:2n6	29.18	30.61	28.24	30.58	38.09
18:3n-3	1.00	1.20	1.15	1.34	1.89
20:0	0.81	0.71	0.82	0.71	0.60

22:0	0.11	0.17	0.11	0.21	0.21
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^aComposition of prickly pear meal 86.97g of DM, 5.14g of CP, 40.97g of NDFap, 1.48g of EE, 17.21g of ash and 34.93g of NFC. ^bComposition: Calcium 200 g; Cobalt 200 mg; Copper 1,650 mg; Sulfur 12 g; Iron 560 mg; Fluorine (max) 1,000 g; Phosphorus 100 g; Iodine 195 mg; Magnesium 15 g; Manganese 1,960 mg; Nickel 40 mg; Selenium 32 mg; Sodium 68 g; Zinc 6,285 mg. ^cNeutral Detergent Fiber corrected for ash and protein. ^dNon-fiber carbohydrates. Calculated according to Hall (2000). ^eTotal digestible nutrients. Calculated according to the NRC (2001). ^fMetabolizable energy. Calculated according to the NRC (2001). ^gFatty Acids.

Table 2 – Dry matter and nutrient intake and total tract digestibility of dry matter and nutrients of cows fed diets with different levels of prickly pear meal

Item	Levels of prickly pear meal in the diet (% DM)					SEM ^a	p-value ^{*b}		
	0	3.7	7.4	11.1	14.8		CON vs. PPM	L	Q
Intake (kg/day)									
Dry matter	12.24	12.98	12.35	13.19	13.06	0.253	0.070	0.454	0.431
Crude protein	2.36	2.40	2.25	2.37	2.30	0.048	0.676	0.527	0.467
NDF _{ap} ^c	5.58	5.96	5.67	6.20	6.07	0.123	0.037	0.239	0.623
Ether extract	0.33	0.36	0.32	0.31	0.28	0.007	0.615	<0.001	0.531
NFC ^d	4.36	4.50	4.43	4.60	4.63	0.079	0.117	0.209	0.651
TDN ^e	8.80	9.23	8.72	9.20	9.02	0.144	0.422	0.895	0.531
Intake (%BW)									
Dry matter	2.63	2.74	2.63	2.74	2.76	0.046	0.286	0.594	0.369
NDF _{ap} ^c	1.20	1.26	1.21	1.29	1.28	0.022	0.148	0.361	0.536
Total tract digestibility									
Dry matter	62.15	62.49	60.30	60.14	60.35	0.596	0.205	0.120	0.204
Crude protein	70.88	71.93	68.63	71.30	69.74	0.730	0.777	0.563	0.562
NDF _{ap} ^c	50.26	50.55	48.46	49.18	48.78	0.723	0.356	0.296	0.392
Ether extract	68.69	66.89	65.24	59.96	57.47	1.475	0.081	0.022	0.895
NFC ^d	88.36	89.14	89.31	88.30	88.88	0.455	0.608	0.678	0.834
TDN ^e	66.30	66.29	65.03	64.14	63.67	0.794	0.116	0.068	0.828

* Statistical differences were considered significant at $P \leq 0.05$ and trends at $0.05 < P \leq 0.10$.

^a Standard error of the mean, ^bCON vs. PPM, control vs. treatment; L, linear effect of PPM; Q, quadratic effect of PPM. ^cNeutral Detergent Fiber corrected for ash and protein. ^d Non-fiber carbohydrates. ^eTotal digestible nutrients.

Table 3- Nitrogen balance, urea nitrogen concentration, and microbial efficiency of cows fed diets containing different levels of prickly pear meal

Item	Levels of prickly pear meal in the diet (% DM)					SEM ^a	p-value ^{*b}		
	0	3.7	7.4	11.1	14.8		CON vs. PPM	L	Q
Nitrogen (N) utilization (g/day)									
Ingested-N	407.35	394.36	411.95	412.35	406.63	6.85	0.951	0.583	0.442
Fecal-N	114.13	110.00	113.83	111.52	109.42	4.11	0.743	0.910	0.711
Urinary-N	197.51	192.11	205.44	199.84	199.05	1.90	0.739	0.427	0.102
Milk-N	57.09	57.47	55.96	53.88	54.08	1.07	0.498	0.234	0.710
Retained-N	38.62	34.78	36.72	47.11	44.08	6.93	0.903	0.571	0.869
Urea nitrogen concentrations (mg/dL)									
Plasma	6.23	7.34	7.85	6.25	6.44	0.41	0.398	0.223	0.838
Urine	12.15	11.33	11.63	10.60	12.62	3.62	0.850	0.561	0.891
Milk	5.66	5.28	5.42	4.94	5.88	1.07	0.638	0.584	0.457
Microbial CP synthesis (g/day)									
Microbial crude protein	1163.9	1256.91	1178.26	1203.98	1187.41	180.02	0.672	0.331	0.431
Microbial efficiency									
Microbial CP (g/kg of TDN)	133.62	137.52	136.69	149.74	133.86	22.33	0.444	0.540	0.279

* Statistical differences were considered significant at $P \leq 0.05$ and trends at $0.05 < P \leq 0.10$.

^aSEM, Standard error of the mean. ^bCON vs. PPM, control vs. treatment; L, linear effect of PPM; Q, quadratic effect of PPM

Table 4- Productive parameters and milk composition of cows fed diets with different levels of prickly pear meal

Item	Levels of prickly pear meal in the diet (% DM)					SEM ^a	p-value ^{*b}		
	0	3.7	7.4	11.1	14.8		CON vs. PPM	L	Q
Milk yield (kg/day)	11.54	11.42	11.20	11.06	10.70	0.212	0.092	0.031	0.750
FCM (3.5%) (kg/day) ^c	12.20	12.17	12.37	12.20	11.93	0.215	0.926	0.475	0.379
Composition (Kg/day)									
Protein	0.37	0.37	0.36	0.35	0.34	0.007	0.183	0.023	0.764
Fat	0.47	0.44	0.46	0.45	0.44	0.009	0.497	0.883	0.345
Lactose	0.52	0.52	0.51	0.49	0.49	0.009	0.182	0.022	0.782
DDE ^d	0.99	1.00	0.98	0.95	0.93	0.018	0.189	0.022	0.803
TDE ^e	1.44	1.44	1.43	1.40	1.38	0.025	0.481	0.090	0.824
Composition (%)									
Protein	3.17	3.23	3.19	3.13	3.18	0.017	0.767	0.137	0.076
Fat	3.84	3.96	4.12	4.16	4.15	0.097	0.117	0.338	0.538
Lactose	4.52	4.59	4.54	4.45	4.53	0.024	0.763	0.115	0.081
DDE ^d	8.66	8.82	8.74	8.56	8.70	0.047	0.663	0.129	0.093
TDE ^e	12.51	12.78	12.83	12.73	12.85	0.124	0.117	0.867	0.834
MEC (kg/day) ^f	12.18	12.17	12.25	12.05	11.80	0.209	0.684	0.244	0.516
BWV (kg/day) ^g	-0.20	-0.02	0.22	0.24	0.21	0.170	0.198	0.395	0.218

*Statistical differences were considered significant at $P \leq 0.05$ and trends at $0.05 < P \leq 0.10$).

^aStandard error of the mean. ^bCON vs. PPM, control vs. treatment; L, linear effect of PPM; Q, quadratic effect of PPM. ^cMilk yield was corrected to 3.5% fat: FCM = $(0.432 + 0.165 \times \text{milk fat content}) \times \text{milk yield (kg/day)}$. ^dDefatted dry extract. ^eTotal dry extract. ^fEnergy-corrected milk: MEC = $0.327 \times \text{milk production (kg/day)} + 12.86 \times \text{fat production (kg/day)} + 7.65 \times \text{protein production (kg/day)}$. ^gBody weight variation.

Table 5 - Fatty acid composition of milk from cows fed diets with different levels of prickly pear meal

Item	Levels of prickly pear meal in the diet (% DM)					SEM ^a	p-value ^{*b}		
	0	3.7	7.4	11.1	14.8		CON vs. PPM	L	Q
C4:0	2.85	2.87	2.88	3.11	2.91	0.045	0.341	0.382	0.111
C6:0	2.52	2.47	2.30	2.37	2.52	0.058	0.442	0.763	0.199
C8:0	1.66	1.69	1.61	1.69	1.65	0.030	0.952	0.783	0.659
C10:0	4.07	3.87	3.79	3.66	3.68	0.083	0.030	0.211	0.709
C12:0	4.52	3.75	3.93	3.58	2.96	0.218	0.023	0.104	0.276
C14:0	14.62	14.70	14.79	14.24	14.52	0.175	0.847	0.354	0.711
C16:0	40.20	39.56	40.79	44.03	45.69	0.508	0.013	<0.001	0.789
C18:0	7.25	6.68	5.66	4.72	4.21	0.205	<0.001	<0.001	0.451
C18:1n7	0.98	0.98	0.89	0.83	0.70	0.031	0.076	0.002	0.730
C18:1n9	14.46	15.60	14.82	13.41	12.42	0.360	0.610	<0.001	0.876
C18:2n6	1.25	1.99	1.33	1.14	1.21	0.142	0.638	0.078	0.254
C18:2 <i>cis</i> -9 <i>trans</i> -11	0.27	0.25	0.30	0.24	0.23	0.015	0.585	0.410	0.346
C18:2 <i>trans</i> -10 <i>cis</i> -12	0.15	0.17	0.15	0.24	0.16	0.020	0.539	0.730	0.460
C18:3n-3	0.12	0.11	0.12	0.12	0.13	0.006	0.722	0.508	0.823
C20:0	0.14	0.13	0.12	0.12	0.10	0.004	0.039	0.025	0.428
C22:0	0.11	0.11	0.13	0.16	0.14	0.011	0.295	0.284	0.325
Others									
<C16	32.74	31.98	33.04	32.03	31.67	0.399	0.461	0.525	0.373
>C16	27.06	28.46	26.18	23.94	22.64	0.557	0.146	<0.001	0.647
C16 total	40.20	39.56	40.79	44.03	45.69	0.508	0.013	<0.001	0.789
C18									
Saturated C18	7.25	6.68	5.66	4.72	4.21	0.205	<0.001	<0.001	0.451
Unsaturated C18	17.22	19.10	17.61	15.99	14.84	0.431	0.722	<0.001	0.841
Total									
Saturated	79.90	77.75	78.43	80.04	80.88	0.415	0.497	0.004	0.926

Unsaturated	20.10	22.25	21.57	19.96	19.13	0.415	0.497	0.004	0.925
U/S ^c	0.25	0.29	0.28	0.25	0.24	0.007	0.477	0.007	0.939

*Statistical differences were considered significant at $P \leq 0.05$ and trends at $0.05 < P \leq 0.10$.

^a Standard error of the mean. ^bCON vs. PPM, control vs. treatment; L, linear effect of PPM; Q, quadratic effect of PPM. ^c Unsaturated to saturated ratio.

Table 6- Eating behavior of cows fed diets with different levels of prickly pear meal

Item	Levels of prickly pear meal in the diet (% DM)					SEM ^a	P-value ^{*b}		
	0	3.7	7.4	11.1	14.8		CON vs. PPM	L	Q
Time spent (minutes/day)									
Feeding	358.4	353.4	380.9	384.4	347.7	6.540	0.596	0.751	0.150
Rumination	527.3	508.3	521.7	513.4	516.6	8.814	0.463	0.800	0.745
Idling	554.3	578.3	537.4	542.2	575.7	10.958	0.862	0.926	0.648
Feeding behavior (number of periods/day)									
Feeding	13.89	15.24	15.65	15.39	14.36	0.378	0.140	0.387	0.347
Rumination	20.47	19.75	18.72	18.88	17.79	0.356	0.036	0.072	0.965
Idling	26.61	27.09	25.55	26.83	25.04	0.433	0.628	0.216	0.886
Time per period of feeding behavior (hour)									
Feeding	0.45	0.40	0.45	0.43	0.40	0.016	0.403	0.932	0.211
Rumination	0.45	0.45	0.48	0.44	0.51	0.018	0.223	0.128	0.378
Idling	0.36	0.36	0.37	0.35	0.38	0.014	0.901	0.605	0.465
Feeding efficiency									
Dry matter /hour(g)	2123.9	2182.2	2074.8	2133.8	2182.6	50.79	0.853	0.885	0.408
NDF/hour(g) ^c	1005.3	1012.7	916.3	986.4	1014.1	24.07	0.607	0.671	0.123
Rumination efficiency									
Dry matter /hour (g)	1403.7	1520.4	1521.0	1603.3	1460.2	33.45	0.100	0.736	0.278
NDF/hour (g) ^c	660.0	697.3	678.7	738.0	683.2	15.21	0.256	0.901	0.557
Chewing									
TCT (minutes/day) ^d	14.76	14.33	15.05	14.96	14.41	0.183	0.862	0.926	0.065
NBR (number/day) ^e	579.06	520.51	526.05	550.16	506.12	13.30	0.046	0.853	0.293
NCD (number/day) ^f	31760	30477	31910	31983	31020	756.3	0.778	0.767	0.362

NCB (number/bolus) ^g	56.79	59.59	61.03	58.89	61.69	1.250	0.076	0.588	0.699
TBR (second/bolus) ^h	56.72	59.60	60.72	56.99	62.95	1.121	0.091	0.410	0.170

*Statistical differences were considered significant at $P \leq 0.05$ and trends at $0.05 < P \leq 0.10$.

^aStandard error of the mean. ^bCON vs. PPM, control vs. treatment; L, linear effect of PPM; Q, quadratic effect of PPM. ^cNeutral Detergent Fiber. ^dTotal chewing time. ^eNumber of boluses ruminated per day. ^fNumber of chews per day. ^gNumber of chews per boluse. ^hTime spent per boluse ruminated.