



Effects of Feeding Marula [*Sclerocarya birrea* (L.) Seed Cake on Milk Yield and Composition for Lactating Dairy Cows

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Authors' contributions

This work was carried out in collaboration with all authors. Authors JFM, AMD and GZK designed the study. Author MPM wrote the protocol, carried out the experiment, performed the statistical analysis and wrote the first draft of the manuscript. Authors AMD and GZK supervised the experiment and helped in interpreting the results. All authors read and approved the final manuscript.

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ABSTRACT

Aim: To investigate the effect on milk yield and composition of feeding Marula (*Sclerocarya birrea*) seed cake as a protein source to lactating dairy cows.

Study Design: Nine multiparous Friesian dairy cows in mid-lactation were assigned to a 3 x 3 Latin square design (three replicate).

Place and Duration of Study: Department of Animal Science, Faculty of Agriculture and Consumer Sciences, University of Swaziland, between September 2014 and December 2014 (90 days).

Methodology: The treatments were as follows: Control diet (CD) - containing 100% CP from SBM; Marula seed meal (MSM) - diet containing 100% crude protein (CP) from Marula seed cake (MSC); and soya bean meal (SBM) + Marula seed cake (SBM+MSC) - diet containing 50% CP from MSC and 50% from SBM, and. Milking was done twice daily and the milk yield was recorded. Milk samples were stored at -18°C pending chemical analysis.

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Results: The results of substituting SBM with MSC indicated no significant ($P = .05$) differences in dry matter intake (DMI), milk yield and composition among the diets. Milk yield was 19.5 litres per day across the diets. Milk protein ranged from 3.16% (CD) to 3.29% (SBM+MSC) while milk fat ranged from 3.23% (CD) to 3.73% (MSM).

Conclusion: The study shows that dairy cattle diets can be formulated using MSC as a protein source while maintaining or increasing dry matter intake, milk yield and composition of dairy cows. It indicates that MSC, which is a by-product in oil extraction companies, is a valuable protein feed resource that holds profitable potential in animal production.

Keywords: Marula seed cake; sustainable animal production; milk constituents; dry matter intake; body weight.

1. INTRODUCTION

Agriculture is the backbone and key driver of Swaziland's economy. It is also a major source of employment for rural households, with approximately 70% of the population dependent on agriculture [1]. According to [1], cattle comprise the largest component of Swaziland's livestock population. The total cattle population in the country is about 633,954 of which an estimated 5,224 are dairy cattle [2]. Dairy cattle comprise only a small proportion, about 1%, of the cattle population, however, Swaziland aspires to have a viable, sustainable and competitive dairy industry to adequately meet the demand of her population [3] estimated at 1.1 million [4].

Milk is one of the most popular dairy products in Swaziland and is an important aspect of human diets. On average, Swazis consume about 54.76 litres of milk *per capita* [3]. According to [3], the demand for dairy products is estimated at 60.24 million litres per annum while commercial milk production from the national dairy herd is estimated at 9.76 million litres per annum and the deficit is met through imports and milk produced by the indigenous herd in the traditional sector [3]. Previous reports have shown that this shortage may be attributed to the small population of specialized dairy breeds in the country and high costs of supplementary feed [5]. The increasing human population in the country depicts a severe milk shortage in the near future unless comprehensive programmes in production are designed and implemented. In order to alleviate feed constraint in dairy production there is a need to use non-conventional, less costly and locally available protein rich feedstuffs. One such naturally occurring and abundant feed resource is Marula seed cake, a residue from oil extraction of Marula kernels [6].

The evaluation of Marula (*Sclerocarya birrea* L.) seed cake as a protein source in commercial

cattle fattening diets has been reported where it was concluded that MSC is rich in protein, 470 g/kg DM [7]. In another study MSC was incorporated in dairy meal as a protein supplement to determine the chemical composition and digestibility of the diets in comparison with a commercial diet containing soya bean meal as a protein supplement [8]. In this study it was concluded that MSC is rich in crude protein, 47.21% (w/w), hence it can be successfully incorporated in dairy meals without lowering the nutritive value of the ruminant feed. This study sought to establish information on the effects of feeding this non-conventional feedstuff as a protein source to lactating dairy cows.

2. MATERIALS AND METHODS

2.1 Site Description

The experiment was conducted between September 2014 and December 2014 (90 days) at the University of Swaziland (UNISWA), Luyengo Farm, which is located in the upper Middleveld of Swaziland (coordinates: 26°32' S – 31° 14' E) with an altitude of 600-800 meters above sea level [9]. This site receives an annual rainfall of 850-1000 mm and mean temperature of 18°C [9] and the summer rains fall in the period of October and March [10]. Chemical analyses of milk composition were done at the Animal Science Nutrition laboratory and Animal Science Dairy Technology laboratory of the University of Swaziland, Luyengo campus.

2.2 Experimental Procedure

2.2.1 Experimental diets

The diets used in this study were formulated as described before [8] and the chemical composition of these diets is presented in Table 1.

Table 1. Chemical composition of dairy meal containing Marula seed cake and soya bean meal

Parameter	n	MSM	SBM+MSC	CD
Metabolisable Energy (MJ/kg DM)	4	1163.98	1114.70	1073.84
Dry matter (%)	4	87.45	88.35	87.52
Crude protein (%)	4	22.22	23.09	24.23
Neutral Detergent Fibre (%)	2	18.49	18.88	18.75
Acid Detergent Fibre (%)	2	8.70	7.81	5.95
Non-fibre carbohydrates (%)	4	45.86	46.52	45.61
Starch (%)	2	33.14	32.28	34.53
Ether extract (%)	4	7.32	5.63	4.15
Ash (%)	4	6.11	5.89	7.26
Nitrogen Free Extracts (%)	4	46.09	47.69	45.67

Where: n – Number of replicate samples, MSM - A formulated diet containing Marula seed cake (MSC) as a protein source, SBM+MSC - A formulated diet containing 50% soya bean meal (SBM) and 50% MSC as a protein source, CD - A formulated diet containing SBM as a protein source
Source: [8]

2.2.2 Experimental animals

Nine multiparous, mid-lactating Friesian cows (380 ± 50 kg of body weight) producing an average of 15 ± 2 litres of milk/day were selected from the dairy herd at UNISWA farm. The dairy cows were sprayed weekly with an acaricide to control ticks and prevent tick-borne diseases. The cows had full access to a clean and abundant water source.

2.2.3 Experimental design

The experiment lasted 90 days, divided into three periods of 30 days, of which the first 7 days of each period were used for adaption of animals to the experimental diets and 21 days for data collection. A Latin square design was used in allocating the experimental diets and is appropriate since the cows were changing in physiological state during the experiment [11]. As a basal diet, cows grazed on rye grass (*Lolium multiflorum* Lam.) then were supplemented with the different experimental diets after milking. After milking at 0600 hours and 1500 hours, the cows were offered a fixed amount (3 kg) of experimental diets (6 kg daily), based on 1 kg for every 3 litres of milk produced.

Table 2. Latin square arrangement of the three experimental diets

Periods	Animals		
	Cows 1	Cows 2	Cows 3
1	CD	SBM+MSC	MSM
2	SBM+MSC	MSM	CD
3	MSM	CD	SBM+MSC

MSM - A formulated diet containing Marula seed cake (MSC) as a protein source

SBM+MSC - A formulated diet containing 50% soya bean meal (SBM) and 50% MSC as a protein source

CD – Control diet containing SBM as a protein source

2.2.4 Sampling

Milking was done two times a day; in the morning at 0400 hours and in the afternoon at 1400 hours using a milking machine and milk yield per cow was recorded after every milking. The cows were weighed weekly to determine any weight loss/gain. Sampling was done according to [12] and was done daily after the adaptation period and was done separately for the morning and afternoon milking. The milk was put in transparent sampling plastic bags and stored at -18°C pending chemical analysis. Prior to analysis, the milk sampled from the same cow, same treatment and same period was thawed and pooled into one sample then taken to the laboratory for chemical analysis.

2.3 Data Collection

2.3.1 Dry matter intake

Average daily dry matter intake (DMI) was calculated following the method by [13]:

Average daily DMI (kg) = Average DM offered daily – average DM refused daily

Where:

Average DM offered daily = Average weight of feed offered daily (kg) x DM content of feed (%) / 100

Average DM refused daily = Average weight of uneaten feed daily (kg) x DM content of refusals (%) / 100

2.3.2 Milk composition

Milk samples were analysed for milk protein, milk fat, specific gravity, ash, titratable acidity, lactose, total solids and solids non fats. Milk

protein was determined using the Kjeldahl method [14], specific gravity was measured using a lactometer, ash by combustion at 550°C for 4 hours, milk fat was determined by using the Gerber method for fat determination from whence total solids and solids non fats were calculated; titratable acidity by titrating with 0.1 normality sodium hydroxide and lactose by using the DNS test for reducing sugars method [15]. Daily milk energy output was estimated from measured milk yield and concentrations of milk fat, CP and lactose according to [16] equation:

$$NE_L \text{ (Mcal/kg of milk)} = 0.0929 * EE\% + 0.0547 * CP\% + 0.0395 * \text{lactose}$$

2.4 Statistical Analysis

The general linear model (GLM) procedure of statistical analysis system [17] was used to determine variation in milk yield and milk composition, with the following linear model:

$$Y_{ijkl} = \mu + B_i + Y_j + T_k + Q_l + Q_l * T_k + e_{ijkl}$$

Where:

Y_{ijkl} = dependent variable receiving treatment k in row i and column j

μ = overall mean

B_i = i^{th} row effect (effect of the periods)

Y_j = j^{th} column effect (effect of the cows)

T_k = k^{th} treatment effect (effect of the treatments)

Q_l = l^{th} Latin squares effect (effects of Latin squares)

$Q_l * T_k$ = interaction of the effect with Latin squares x treatment

e_{ijkl} = random or residual error

Treatment means were separated using the least significance difference (LSD) in the least squares means statements of the GLM procedure in SAS [17] at 95% confidence level.

3. RESULTS AND DISCUSSION

3.1 Cost of Ingredients

Using Marula seed cake (MSC) as a protein source in dairy cattle diets was economical. A tonne of MSC cost E4500 at the Mpaka oil extraction company whereas a tonne of soya bean meal (SBM) cost E5900 at Arrowfeeds Company, Swaziland. MSC is locally and readily available to farmers in Swaziland [7] during the 'maganu' season. Soya bean meal is imported largely by the animal feed companies. Therefore,

it would be cost effective for farmers to formulate their own dairy cattle diets using MSC as a protein source as it does not impair animal performance, as discussed below.

3.2 Body Weight and Dry Matter Intake

There were no significant ($P = .05$) differences observed in body weight of the animals across the diets (Table 3). Therefore, the different diets had no significant effect on the body weight of the cows. The body weight of the cows ranged from 473.00 kg (CD) to 487.19 kg (SBM+MSC).

Table 3. Body weight (n=9) and dry matter intake (Mean, SEM)

Diet	Body weight (kg)	Dry matter intake (kg/day)
MSM	481.44 ^a , 23.42	5.25 ^a , 0.04
SBM+MSC	487.19 ^a , 20.43	5.30 ^a , 0.04
CD	473.00 ^a , 13.45	5.25 ^a , 0.01

^aMeans in the same column with the same superscripts do not differ significantly ($P = .05$)

MSM- A formulated diet containing MSC as a protein source
SBM+MSC- A formulated diet containing 50% MSC and 50% SBM as a protein source

CD- A formulated diet containing SBM as a protein source

Dry matter intake (DMI) is an indication of the quantity of feed dry matter a cow consumes in a given period of time. Dry matter consists of all the nutrients of a feed except water. Thus an increase in DM intake indicates high nutrient uptake which should result in high milk yield. Dry matter intake is affected by the fibre composition of the feed as it decreases with an increase in fibre. There was no significant ($P = .05$) difference in DMI of the diets (Table 3).

The DMI of the concentrate diets observed in this study are in agreement with the findings of [18] who observed that cows fed high-concentrate low forage ratio consumed 5.1 kg of DM concentrate in the parlour. The DMI obtained by [19] ranged from 21.7 kg/day for diets containing 32% NDF to 23.4 kg/day for diets containing 28% NDF, and these values are higher than those obtained in this study. This may be attributed to the fact that [19] fed a total mixed ration compared to this study where the cows were allowed to graze on rye grass then supplemented with dairy meal.

Oil has an effect on fibre digestion [20] and consequently dry matter intake. [21] concurred with this statement when reporting that high fat content (>5%) can coat the fibre thus reducing the hydrophilicity of feed particles and therefore

interfering with its digestion by rumen microbes. Marula seed cake has an appreciable level of crude fat content of 28.96% DM [8] while soya bean meal has small amounts of fat (1.7% observed by [22]; 2.07% from the soya bean meal used in this study as reported by [8]). Therefore, since the digestibility of a feed, and consequently its DMI, is affected by the fat content of the feed, MSM would be expected to have lower DMI values in this study, however, this was not so. This may be attributed to the alleviation of negative oil effects by the concentration of divalent cations [23] in the MSM (1.02% Ca and 0.34% Mg) and SBM+MSC (0.99% Ca and 0.34% Mg) diets [8]. [24] observed that with an increase in lipid supplementation comes a decrease in the contents of soluble and ionisable Ca. Calcium supplements are assumed to provide ionic Ca in the rumen that combines with free fatty acids rendering them largely inactive [25], however, this is subject for further research. Another possible reason for the unobserved detrimental effects of high oil content could be that the oil from Marula seed cake has a lower degree of unsaturation, which does not inhibit ruminal fermentation nor decrease fibre digestibility and milk fat test [25].

3.3 Milk Yield

The average milk yield obtained is presented in Table 4. Substituting soya bean meal with Marula seed cake had no significant ($P = .05$) effect on the morning and afternoon milk yield. The average daily milk yield was also not affected ($P = .05$) by the use of MSC as a protein source.

There was no significant ($P = .05$) difference in milk yield across diets in this study as observed by other researchers [26,18]. It has been reported elsewhere that the synthesis of lactose and overall milk yield is reduced if too little propionate is produced, which can occur during the feeding of high fibre diets [21]. [21] also

stated that when the energy level is in excess relative to the protein level, the rate of microbial protein synthesis declines with resultant decrease in milk yield. The excess energy may be converted to body condition rather than milk. The excess energy may be from the high amount of EE contained in Marula seed cake [8]. This energy may be beneficial to high producing dairy cows in early lactation as they have a great demand for energy [20].

Based on these results, in the 305 days lactation period, substituting 100% soya bean meal with 100% Marula seed cake may result in 5,947.50 litres of milk produced per cow, and substituting 50% soya bean meal with 50% Marula seed cake may result in 5,956.65 litres of milk produced per cow and these do not differ from the 5,959.70 litres of milk per cow per lactation produced from feeding soya bean meal. If farmers could venture into dairy farming with the inexpensive and locally available Marula seed cake as a protein source, milk production in Swaziland could be improved and the deficit of 50.48 million litres of LMEs [3] could be reduced.

3.4 Milk Composition

In all the parameters measured, there was no significant ($P = .05$) effect of replacing soya bean meal with Marula seed cake (Table 5).

Petit [26,27] also found no differences in milk composition after feeding cows with different dairy rations. [18] observed no significant effects of concentrates on milk fat and protein. Reports of replacing oilseeds and using non-conventional feedstuffs in animal rations have shown that these feedstuffs may be effectively included in rations without detrimental effects on animal performance in terms of milk yield and composition [28,26,29,30]. [31] stated that a cow's diet has a fairly small effect on milk composition, except for milk fat content and composition.

Table 4. Effect of diet (n=9) on milk yield (Mean, SEM)

Milk yield	MSM	SBM+MSC	CD
Morning (Litres)	10.37 ^a , 0.76	10.61 ^a , 0.85	10.56 ^a , 0.72
Afternoon (Litres)	9.13 ^a , 0.75	8.92 ^a , 0.72	8.98 ^a , 0.77
Average daily (Litres)	19.50 ^a , 1.49	19.53 ^a , 1.55	19.54 ^a , 1.46

^{a,b} Means on the same row with the same superscripts do not differ significantly ($P = .05$).

MSM - A formulated diet containing Marula seed cake (MSC) as a protein source

SBM+MSC - A formulated diet containing 50% soya bean meal (SBM) and 50% MSC as a protein source

CD - A formulated diet containing SBM as a protein source

Table 5. Effect of the diets on milk composition (n=9) (Mean, SEM)

Milk parameter	MSM	SBM+MSC	CD
NE _L (MJ/kg of milk)	3.40 ^a , 0.08	3.18 ^a , 0.14	3.10 ^a , 0.12
Titratable acidity (%)	0.16 ^a , 0.01	0.17 ^a , 0.01	0.16 ^a , 0.01
Lactometer reading	29.87 ^a , 0.16	29.86 ^a , 0.17	29.90 ^a , 0.18
Specific gravity	1.0299 ^a , 0.00	1.0297 ^a , 0.00	1.0299 ^a , 0.00
Fat (%)	3.73 ^a , 0.18	3.40 ^a , 0.28	3.23 ^a , 0.18
Lactose (%)	7.20 ^a , 0.31	6.67 ^a , 0.32	6.71 ^a , 0.41
Crude protein (%)	3.28 ^a , 0.06	3.29 ^a , 0.12	3.16 ^a , 0.10
Ash (%)	0.70 ^a , 0.08	0.69 ^a , 0.06	0.76 ^a , 0.08
Total solids (%)	12.73 ^a , 0.22	12.34 ^a , 0.34	12.14 ^a , 0.24
Solids non-fats (%)	9.00 ^a , 0.06	8.93 ^a , 0.07	8.91 ^a , 0.07

^a Means on the same row with the same superscripts do not differ significantly ($P = .05$).

MSM- A formulated diet containing MSC as a protein source

SBM+MSC- A formulated diet containing 50% MSC and 50% SBM as a protein source

CD- A formulated diet containing SBM as a protein source

3.4.1 Milk fat

The fat content of milk is determined by the proportion in which volatile fatty acids such as acetate and propionate are produced [21]. Acetate is used for the synthesis of milk fat and is an end product of fibre fermentation hence a decrease in dietary fibre and fibre digestion causes a reduction in milk fat test [21,32]. Milk fat is the most sensitive constituent of milk to dietary manipulation and could be changed over a range of 3% units [33]. According to [31], the fat content and composition is affected by the amount and type of fat included in the diet, and by the composition of the non-lipid diet.

Substituting soya bean meal with Marula seed cake caused no significant difference in milk fat percentages. Some well documented studies conducted using different formulated rations in dairy production also found no significant difference on milk fat content [34]. MSM and SBM+MSC had a numerically higher milk fat percentage. [35] observed that an increase in milk yield is followed by a decrease in the percentages of milk protein and milk fat, especially in the early stages of lactation, but this was not so in the present study as the cows were in mid lactation. Cow's, Friesian, milk has a typical average fat composition of 3.65% [36, 35]. Compared to these studies, the milk fat contents of MSM and SBM+MSC were above the average.

3.4.2 Milk protein

Milk protein is the second most variant constituent of milk; however, no significant differences were observed in milk protein from this study. Diets containing Marula seed cake as

a protein source had a numerically higher CP% compared to the control. However, [33] reported that the inability of a diet to noticeably alter milk protein content is due to the low transfer efficiency, 25-30%, of dietary protein to milk.

The milk protein concentrations of Marula seed cake containing diets obtained in this study are slightly greater than the protein values obtained by [37] for cows supplemented with concentrates with high pasture allowance (3.11% DM). The differences may be attributed to the different protein sources used, they used roasted soybeans; therefore their protein values are similar to the protein content of the control (3.16% DM) from this study. Also, the energy content of the diets which is the fuel for manufacturing the milk constituents was higher for this study. [38] reported a milk protein range of 2.37 - 4.26% with an average of 3.30% while [36] and [27] reported milk protein of 3.5%, which is within and above the average reported by [38].

The findings of this study fall within the milk protein range with Marula seed cake containing diets slightly below the 3.30% average. According to [32], increasing the supply of dietary crude protein has little effect on milk protein percentage. Since the milk protein percentages agree with documented literature, it means that the diets used in this study provided sufficient energy to meet the requirement for milk protein synthesis.

3.4.3 Lactose

Lactose, also known as milk sugar, is the least variant milk constituent and is the principal carbohydrate found in milk [31]. Its principal biological function is the regulation of water

content and is thus a major regulator in milk causing water to move into milk in the mammary gland [38]. Therefore, lactose is the most constant constituent in milk averaging 4.6% [32]. In this study, high milk lactose, ranging from 6.67% (SBM+MSM) to 7.20% (MSM) was observed which, according to [39] should also mean an increase in milk yield.

3.4.4 Ash

Ash is the white or nearly white substance which remains after incinerating the dry residue remaining after removing water from milk. This contains the mineral substances of the milk in question. Due to some chemical changes that occur during the incinerating process, the ash contains carbonates, oxides and phosphates that are not present as such in the original food [40]. The mineral content of milk was 0.70% (MSM), 0.69% (SBM+MSC) and 0.76% (CD). These values are similar to 0.69% ash obtained by [41]. [31] reported a mineral content of 0.7% while [42] reported a mineral content of 0.74% thus the mineral content of the milk from this study agrees with those documented in literature. Using Marula seed cake as a protein source in place of soya bean meal seemed to slightly reduce the mineral content of the milk but still the mineral content was above that from other studies.

3.4.5 Total solids and solids non-fats

Milk total solids are the remainder after removal of water. Solids-non-fats (SNF) comprise the protein, lactose and minerals in milk. The milk samples collected for the different diets showed that MSM, SBM+MSC and CD had total solids and solids-non-fats of 12.73% and 9.00%; 12.34% and 8.93%; and 12.14% and 8.91% respectively. These values follow the trend for the milk yield obtained in this study as the milk yield, though not much different, was the least for MSM (19.50 litres/day) and highest for CD (19.54 litres/day). The milk total solids and solids non-fat content from this study was within the normal range and comparable to previous reports [27,31].

4. CONCLUSION

The results of this study provided evidence that soya bean meal can safely be replaced with Marula seed cake as a protein source without negatively affecting milk yield and composition in lactating dairy cows. It can be concluded that this feed ingredient is inexpensive and locally

available thus considered economical compared to soya bean meal which is imported and cannot be sold as a feed ingredient to individual farmers to help them formulate their own diets.

5. RECOMMENDATION

The use of Marula seed cake (MSC) as a protein source in both 50% and 100% level of inclusion is recommended as this is economical compared to the use of soya bean meal which is not locally available. The use of Marula seed cake would enable farmers to formulate their own cheaper rations and improve the profitability of their dairy enterprises. However, due to the current limitation of Marula seed cake supply, the researchers recommend the use of 50% MSC, especially to commercial feed companies which use large quantities of protein during feed production. On the other hand, the researcher promotes the use of 100% MSC as a protein source to individual farmers as they do not have access to SBM. Although the protein content of Marula seed cake is high, the presence of lipids may limit its utilisation due to detrimental effects of high lipid level in feedstuffs. Thus a recommendation for the efficient extraction of oil from the seed before the cake is used for ruminant nutrition. Those techniques efficient enough to extract the highest proportion of oil would produce Marula seed cake with higher crude protein concentration. It is also recommended that Marula trees should be cultivated and propagated in order to increase the supply of MSC in Swaziland. Further study on the amino acid profile and feeding trial through all the lactation stages is recommended.

ETHICAL APPROVAL

This experiment was examined and approved by the Research Ethics Committee of the University of Swaziland.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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