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Nutritional Evaluation of Marula (Sclerocarya birrea) Seed Cake as a Protein Supplement in Dairy Meal

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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. Authors AMD, GZK and JFM revised the final draft of the manuscript. All authors read and approved the final manuscript.

Article Information

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Original Research Article

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ABSTRACT

Aims: To investigate the nutritional properties of Marula (Sclerocarya birrea) Seed Cake as a protein supplement in dairy cattle ration.

Study Design: Experimental design used was a completely randomized design (CRD).

Place and Duration of Study: Department of Animal Science, University of Swaziland, between September 2014 and December 2014.

Methodology: Three dairy cattle rations were formulated using Marula seed cake (MSC) as follows: Marula Seed Meal (MSM) = a formulated diet containing Marula seed cake as a protein supplement; soya bean meal + Marula seed cake (SBM+MSC) = a formulated diet containing 50% Marula seed cake and 50% soya bean meal (SBM) as a protein supplement; and control diet (CD) = a formulated diet containing soya bean meal as a protein supplement. Marula seed cake which is the protein supplement and the formulated diets were then analysed for dry matter (DM), crude

protein (CP), ash, ether extract (EE) and crude fibre (CF). Non-fibre carbohydrates (NFC), nitrogenfree extracts (NFE) and metabolisable energy (ME) were calculated. In vitro digestibility of the feed was analysed while the apparent dry matter digestibility (ADMD) was calculated. **Results:** The CP, CF, EE and ash content of MSC were 47.21, 4.79, 28.96 and 6.61% (w/w) respectively, and had ME value of 1544.64 MJ/kg DM. The MSC has an in vitro dry matter digestibility (IDMD) and nitrogen digestibility (ND) of 83.33 and 81.46% (w/w) respectively. The CP content, EE and ME value of the formulated diets ranged from 22.22% in MSM to 24.23% CD, 4.39% in CD to 7.32% MSM (w/w) and 1073.84 MJ/kg in CD to 1163.98 MJ/kg in MSM respectively. The IDMD and ND ranged from 79.09% in CD to 81.52% in MSM and from 77.33% (w/w) in CD to 79.45% (w/w) in SBM+MSC respectively.

Conclusion: Marula seed cake is rich in CP and is digestible and thus can be used as a protein supplement in dairy cattle rations.

Keywords: Dairy cattle; digestibility; marula seed cake; nutritive value; soya bean meal.

1. INTRODUCTION

The dairy industry plays an important role in supplying milk as a near complete diet and a cheaper source of nutrients to consumers. There are a lot of constraints that may limit smallholder dairy farming in the tropics such as unavailable technology, lack of training and finance, low milk price, feed shortage, poor farm management, low productive and reproductive performance of the dairy cows and high disease prevalence [1]. The major causes for low milk production in Swaziland are high livestock disease prevention and control costs; high feed costs [2] which may lead to malnutrition, especially inadequate animal protein intake which is believed to hinder development like in most developing countries [3].

About 90% of dairy farmers in Swaziland are on Swazi Nation Land (SNL), and dairy cattle grazed on this land do not perform well due to poor pasture productivity, especially during the dry season [3]. Good quality pasture contains both rumen degradable protein (RDP) and undegradable dietary proteins (UDP), the latter requirements rise as production rises above 12 litres per day [4]. Farmers would then be compelled to supplement their animals with dairy meal, which could be very costly particularly to small holder dairy farmers. The high costs may result from the costly conventional ingredients in ration formulation, which mainly are nitrogen sources, one of which is soya bean meal. In order to alleviate feed constraint in dairy production there is a need to use nonconventional, less costly and locally available protein rich feedstuffs. One of such naturally occurring and abundant feed resource is Marula seed cake (MSC), a residue from oil extraction of Marula kernels [5].

Although MSC is rich in protein, 470 g/kg DM [6], its nutritional value in dairy farming is largely unknown. One important factor of the nutritive value of feed is digestibility as it determines the relationship between the contents of nutrients and energy that are available to ruminants [7]. Feed sources differ in digestibility and the quality of the feed is principally evaluated by feeding trials, which are often time consuming and expensive.

The digestibility of feeds can be evaluated using different techniques [8]; including the biological methods which require the use of fistulated animals (in vivo) and laboratory (in vitro) methods which simulate the rumen environment using rumen liquor from donor animals. The in vitro digestibility methods are less expensive and easier hence extensively used in determining the digestibility of different feed ingredients [9]. Tilley and Terry [10] developed a two-stage in vitro technique which closely simulate physiological conditions of ruminants including potential effects of ruminal fermentation.

Feed is the most costly expense in animal production, especially in dairy farming and this has triggered the need for less expensive sources of feed. The ever-increasing cost of commercial feeds arises mainly from the high cost of feed ingredients, especially the conventional ones like cereal grains, oil seed cakes and fish meal, particularly when imported. Other scholars [11] have observed that utilisation of non-conventional feedstuffs in crop-livestock farming systems is hampered by lack of knowledge of their nutritive value and digestibility. Therefore, generation of knowledge on nutritional value and use of locally available feedstuffs is critical for successful exploitation of these. In turn this may result in reduction of feed

costs, and thus making them affordable by smallscale farmers.

Marula seed cake is produced as an industrial waste in Swaziland. Even though it is rich in protein, its nutritional and digestibility values in dairy farming have not yet been determined. It is thus imperative that the nutritional value of this non-conventional feed resource be investigated.

2. MATERIALS AND METHODS

2.1 Site Description

The experiment was conducted at the Faculty of Agriculture and Consumer Sciences, University of Swaziland (UNISWA), 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 [12]. This site receives an annual rainfall of 850-1000 mm and mean temperature of 18°C [12].

2.2 Experimental Diets and Design

The diets were formulated as follows:

- 1. MSM: A formulated diet containing Marula seed cake (MSC) as a protein supplement.
- 2. SBM+MSC: A formulated diet in which 50% of soya bean meal was substituted with 50% of Marula seed cake.
- 3. CD: A formulated diet containing soya bean meal (SBM) as a protein supplement (positive control diet).

Samples were taken from each diet for proximate analysis. The experimental design used was a completely randomised design.

Other feed ingredients used in formulating MSM, SBM+MSC and CD include wheat bran, yellow maize meal, urea, hominy chop and vitaminmineral premix.

2.3 Chemical Analysis

Marula seed cake and the formulated diets were analysed for DM, CP, ash, EE and CF. The DM content was determined by drying samples at 105°C in an oven for 48 hours, CP using the Kjeldahl method [13]; ash by combustion at 550°C overnight and then calculating the organic matter from the ash weight (g) and dry feed (g), and EE by extraction with anhydrous ether using a Soxhlet apparatus. The CF was estimated by using a Buckner flask and funnel, neutral detergent fibre (NDF) by using a neutral detergent solution and acid detergent fibre (ADF) by using an acid detergent solution [14]. Mineral composition of the diets was determined by Central Analytical Laboratories (CAL) in South Africa with the help of Arrowfeeds Swaziland. Non-fibre carbohydrates (NFC) were determined using an equation from Grant and Kononoff [15] and Iqbal et al. [16]:

$$
NFC\% DM = 100 - (\% NDF + \% CP + \% EE + \% ash) \tag{1}
$$

Nitrogen free extract (NFE) was determined by the equation of Forejtova et al. [7] and Moran [17]:

$$
NFE (%)DM) = DM% - (CP% DM + EE% DM + CF% DM + ash% DM)
$$
 (2)

Metabolisable energy (ME) was estimated following the equation of Stevenson and Graham [18]:

ME (MJ/kg DM) =
$$
0.82^*((0.24^*CP) + (0.39^*EE) + (0.18^*R))^*
$$
 in vitro OMD (3)

Where: $R = OM\%$ - (CP + EE); CP, EE and R are as %DM.

2.4 In vitro Digestibility

In vitro digestibility of feed samples was estimated by the method of Tilley and Terry [10] using faecal liquor as an inoculum and a 48 hours incubation period [19]. Artificial saliva (buffer solution) was prepared according to the suggested composition for synthetic saliva by McDougall [20]. This experiment was repeated two times for the determination of organic matter digestibility (OMD) and nitrogen degradability (ND).

The digestibility of dry matter was calculated as follows [10]:

In vitro dry matter digestibility (%) =
$$
[A - (B - C) / A]
$$
 * 100 (5)

Where:

 $A =$ dry weight of sample

 $B =$ dry weight of residue after digestion $C =$ dry weight of reagent blank

The organic matter (OM) of the residues and feed was determined by ashing at 550°C

overnight. Organic matter digestibility (OMD) was calculated according to the following formula [10]:

In vitro OMD (
$$
\%
$$
) = $[A - (B - C) \times 100] / A (6)$

Where:

 $A = weight of OM$ in sample

 B = weight of OM of sample residue

 C = weight of OM of control residue

The nitrogen (N) content of the residues and feed was determined using Kjeldahl method. The degradability of protein was then calculated as follows:

In vitro ND (%) =
$$
[A - (B - C) \times 100] / A
$$
 (7)

Where:

 $A = weight of N in sample$

 B = weight of N of sample residue

 C = weight of N of control residue

The apparent digestibility (%ADMD) of the diets was calculated according to the equation of Mertens [21]:

Apparent DM digestibility (%ADMD) = 100 * (Feed DM - Faecal DM) / (Feed DM) (8)

2.5 Statistical Analysis

The general linear model (GLM) procedure of statistical analysis system [22] was used to determine variation between diets, with the following linear model:

Each observation= Overall mean + Treatment effect + Error effect

Treatment means were separated using the probability difference in the least squares means statements of the GLM procedure in SAS [22] at 95% confidence level and the results were reported as mean and standard error of the mean.

3. RESULTS AND DISCUSSION

3.1 Chemical Composition of Marula Seed Cake

The proximate analysis of MSC was done to determine its nutritive value and the results are presented in Table 1. According to the results, MSC has a high ME content, and is rich in CP and EE.

1. Parameters are in % of DM unless noted; 2. $n =$ number of replicate samples used; 3. SEM = Standard error of the mean

Feed Master, Swaziland, provided results of proximate analysis of SBM whose DM content was 88.46%, CP of 47.80% DM, CF of 4.69% DM, EE of 2.07% DM and ash (mineral) of 6.60% DM. These parameters of SBM are not significantly ($P = .05$) different from those of MSC except the EE content.

Lactating dairy cows require large amounts of dietary energy for maintenance, production and reproduction [16]. These energy demands may be met by inclusion of MSC in dairy rations. The energy content of MSC was found to be 1544.64 MJ/kg DM, which is capable of supplying enough energy to high producing cows which are typically in a negative energy balance, especially in early lactation (the first 100 days after calving) [23].

The DM content of MSC obtained in this study was 93.30% and is the portion of the feed that contains nutrients. The DM for SBM was found to be 90% [24], same as that from McDonald et al. [25], and DM obtained from Feed Master, Swaziland was 88.46%. These results show that MSC may supply as much DM to the animals as SBM.

The CP content of MSC in this study was found to be 47.21% DM which is comparable to that of 47% DM obtained elsewhere [6]. The similarity may be attributed to the fact that MSC was purchased from the same company thus the extraction techniques of oil from the Marula seed are the same. In a study conducted before [26], the protein content of Marula seed obtained was 6.17%. This huge difference may be due to the use of the whole nut [26] without removing the oil and kernel thus the protein concentration may have become diluted by the oil and the hard shell of the kernel.

The CP content of MSC as identified in this study is comparable to that of SBM of 47.30% [27] and that obtained by Feed Master, Swaziland, of 47.80%. Therefore, from the CP results of this study, SBM and its potential replacer, MSC, are believed to supply the same amount of CP to the animals. The high EE content of MSC obtained in this study may be due to inadequate extraction of oil from the Marula seed performed in the Swazi Secrets company. Willis [24] found the crude fat content of SBM to be 2.5% while that obtained from Feed Master, Swaziland, was 2.07%. These values show that MSC contains high quantities of EE compared to SBM.

Oilseeds (such as whole cotton seed) can be fed to animals with no observable impairment of the rumen function [28]. [29] noted that the oil from oilseeds is slowly released into the ruminal contents when the seed is masticated, which may help decrease detrimental effects on rumen fermentation and increase the efficiency of milk fat synthesis. [17] further stated that these fats are useful because they increase the energy density of the diet, particularly in early lactation, thus helping to reduce live weight loss.

The oil or fat content of dairy rations should be no more than 5% since high fat content impair rumen function [30]. The results of this study have shown that the EE of MSC is high (28.96%) for dairy rations, and even more so if the ration contains an energy source. The major sources of energy in ruminant diets are carbohydrates and fats [31]. The principal value of fat is that it is a more concentrated source of energy than protein or carbohydrates [17] so MSC can be a beneficial source of both energy and protein in a ration.

The CF content of MSC (4.79%) is comparable to that of the conventional SBM of 4.2% [24] and that obtained from Feed Master of 4.69%. Thus, MSC and SBM have similar CF contents. The ADF content of MSC was 14.74% which is comparable to the findings of [6] who reported an ADF of 13.1% for MSC.

The observed ash (mineral) content of MSC was 6.61% DM, which includes a calcium (Ca) content of 0.15% DM and phosphorus (P) content of 1.32% DM. The ash content is slightly greater than that of 5.55% DM obtained by [6] but this difference is negligible which may be due to the use of the same MSC source. The mineral content of SBM obtained from Feed Master was 6.06% and the Ca and P contents of SBM observed by [24] were 0.30% and 0.24%, respectively. MSC and SBM differ in overall mineral and CA contents; however, MSC can supply five times as much P as SBM.

3.2 Chemical Composition of the Formulated Diets

Proximate analysis was carried out to determine the chemical composition of MSM, SBM+MSC and CD (Table 2). There were significant $(P = .05)$ differences in the ME contents of the diets. The MSM diet had a significantly ($P = .05$) higher ME whereas SBM+MSC and CD were not significantly different. Therefore, substituting SBM with MSC significantly increases the ME of the diet. However, for all the other parameters determined, there were no significant ($P = .05$) differences observed across the diets (Table 2).

Energy measures the feeds ability to help the cow function and is the most important nutrient for milk production [17]. If a feed has high energy content, then more energy will be available to the animal for use. [32] stated that poor digestion results in feeds with low ME so they need more time in the digestive tract hence the stomach becomes full for long. Consequently, energy intake is depressed due to poor feed digestibility, which in turn may reduce milk protein production [15]. [33] reported that the availability of energetic precursors to produce intracellular energy transfer molecules gives dietary energy a positive effect on milk protein synthesis.

Fats are a source of energy for the cow and during microbial fermentation of fats in the rumen some vitamins required by the cow are produced [17]. Previous reports have stated that a cow's diet should contain no more than 5% fats or oils as this may reduce the palatability of the diet thus impairing rumen function [16,28]. The EE content of MSM and SBM+MSC exceeded this limit but it was speculated that this oil content would not affect animal performance. Possible explanation for this assumption could be the high concentration of Ca in MSM (1.02%) and SBM+MSC (0.99%) which if more soluble and

ionisable, promotes the attachment of bacteria to feed particles, thus increasing ruminal digestion [34].

According to [35], a 500 kg lactating cow producing 19.5 litres of milk per day and 3.5% milk fat should consume 142.67 MJ ME per day, MSC supplies much more than is required. The diets used in this study meet and exceed this energy requirement thus MSC when used as a protein source has the potential of increasing the energy content of feeds.

The nitrogen and CP contents of the formulated diets were not different ($P = .05$). Marula seed cake and soya bean meal supply similar dietary nitrogen hence crude protein to the animal.

Feed intake is limited by fibre content and fibre is needed for digestion in the rumen [36]. The CF, NDF and ADF of MSM, SBM+MSC and CD were not significantly different ($P = .05$). The NDF and ADF contents of the diets containing 50% and 100% Marula seed cake obtained by [6] (31% and 6.31%; and 32% and 8.57%, respectively) are higher than the ones obtained in this study possibly due to the use of different ingredients in the formulation. [37] also obtained higher NDF values because the diets were a total mixed ration (TMR).

The NFC and starch content of the diets were not different. The NFC should range between 30- 40% but a range of 40-45% is typical in rations with less forage (<40%) [15], which was the case with the diets in this study. Other researchers [37] obtained starch contents of 29.9% and 30.1% for TMR diets containing 28% NDF and starch content of 25.9% and 26.5% for TMR diets containing 32% NDF. These values are comparable to those obtained in this study indicating that the starch content from this study is in agreement with documented literature. Observed from these results is that starch content is not influenced by the forage content hence NDF content of the feed.

3.3 Mineral Composition

Table 3 reports the mineral composition of the formulated diets. There were no significant $(P = .05)$ differences in the Ca, P and magnesium (Mg) contents. Significant ($P = .05$) differences were observed for copper (Cu), iron (Fe), potassium (K), manganese (Mn), sodium (Na), sulphur (S) and zinc (Zn). The CD diet had significantly ($P = .05$) higher Cu, K, Mn and S contents, while MSM had the least. The highest $(P = .05)$ Mn content was more than twice that of the least. The MSM and SBM+MSC were not significantly ($P = .05$) different in S content. Iron content differed significantly ($P = .05$) across the diets with SBM+MSC containing the highest. The MSM had a significantly $(P = .05)$ higher Fe compared to CD. The MSM contained high $(P = .05)$ amounts of Na and the least $(P = .05)$ Zn content.

Table 2. Chemical composition of formulated diets

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

1. The chemical composition is in % of DM unless stated; $2. n =$ Number of replicate samples; 3. MSM- A formulated diet containing MSC as a protein supplement; 4. SBM+MSC- A formulated diet containing 50% MSC and 50% SBM as a protein supplement; 5. CD- A formulated diet containing SBM as a protein

supplement

Mineral	MSM		SBM+MSC		CD	
	Mean	SEM	Mean	SEM	Mean	SEM
%Calcium	1.02	0.03	0.99	0.06	0.94	0.01
%Phosphorus	0.67	0.04	0.68	0.01	0.57	0.01
% Sodium (Na)	0.40^{b}	0.01	0.34°	0.01	0.35°	0.01
% Magnesium (Mg)	0.34	0.01	0.33	0.01	0.32	0.01
% Potassium (K)	0.71°	0.01	0.82^{b}	0.01	0.87 ^c	0.01
% Sulphur (S)	0.24°	0.01	0.25^{ab}	0.01	0.27^{b}	0.01
Copper (Cu) (ppm)	26.44^a	0.01	29.88^{b}	0.01	35.77°	0.01
Iron (Fe) (ppm)	279.11^{b}	0.01	319.91°	0.01	237.51^a	0.01
Manganese (Mn) (ppm)	61.81 ^a	0.01	99.51^{b}	0.01	146.21°	0.01
Zinc (Zn) (ppm)	187.31^{a}	0.01	292.51°	0.01	263.21^{b}	0.01

Table 3. Mineral composition (n=2) of formulated diets

 a,b,c Values on the same row bearing the different superscripts differ significantly (P = .05)

The concentrations of Ca, P and Mg in the diets were not different. Replacing SBM with MSC significantly $(P = .05)$ increased Na and Fe in MSM and SBM+MSC and Zn in SBM+MSC and significantly ($P = .05$) reduced K, S, Cu, Mn.

3.4 Digestibility of the Feed

The results are presented in Table 4. There were no significant ($P = .05$) differences between the IDMD of MSC and SBM.

Table 5 presents the digestibilities of MSM, SBM+MSC and CD. There were no significant $(P = .05)$ differences among diets for the IDMD. Significant ($P = .05$) differences were observed in the ADMD of the diets. The CD diet had a significantly lower ADMD than the MSM and SBM+MSC diets. Substituting SBM with MSC significantly ($P = .05$) increase ADMD. There were no significant differences observed in the ND of the diets.

Digestibility of a feed indicates the portion of a feed that is available to the cow for use [38]. It is a measure of the overall feed quality because feeds with lower digestibility are poor and more DM intake would be required in order for the animal to obtain the required quantity of nutrients [15]. Digestibility of feed is a function of its chemical (fat, carbohydrate and protein contents) and physical (particle size) characteristics, as these attributes affect capability of digestive enzymes to colonise and digest the feed particles [39]. Feed degradability in the rumen is related to the CF content of the feed and the extent to which it is lignified [40].

The IDMD of the diets was slightly lower than that obtained by [41] of 91.6%. This may be attributed to the differences in the ingredients used to formulate the diets and the composition of the diets. Their concentrate diet had a lower NDF and ADF contents (17.4% DM and 6.5% DM, respectively) and also a higher NFC content (59.2% DM) which may have caused the feed to be easily broken down.

This study demonstrated a positive effect of substituting SBM with MSC on ADMD. The digestibilities of diets containing MSC as a protein source were significantly higher than that of diet containing SBM as a protein source. Thus, significantly more DM is broken down and hence nutrients are accessed by cows from the MSM and SBM+MSC diets than in the CD diet.

A feed may have a high CP content only to find that it is of poor quality and most of the protein is just excreted in the faeces. Thus ND measures the amount of nitrogen available for use by the cow [21]. The ND of MSC was 81.46% whilst SBM has standardised digestibility coefficients for essential amino acids ranging from 85 to 94% [42]. According to the Institute for Natural Resources in Africa (1988) as cited by [43], SBM has high amount (more than 60%) of RDP, good amino acid balance and high cell-wall digestibility. The ND was similar across the diets. The ND of MSM and SBM+MSC diets from this study was higher than that of 70.6% observed by [41] for cows supplemented with concentrates containing SBM as a protein source with high pasture allowance. The differences may be due to the use of MSC as a protein source instead of the roasted soybean they used.

The OMD of MSC as a protein supplement was 73% and was similar to the OMD of sunflower meal obtained by [44] determined using in vivo method (72.3%) and that determined using in vitro enzymatic method (74.0-75.7%). The OMD of MSC was also comparable to that of SBM of 86.5% [27]. This shows that MSC can supply as much organic matter for use by the animal as does other oilseeds.

Table 4. Digestibilities (n=3) of MSC and SBM

^a Means on the same column with the same superscripts do not differ significantly ($P = .05$). nd – not determined

Table 5. Digestibilities (n=3) of formulated diets

 a,b Means within the same column having similar superscripts do not differ significantly (P = .05)

High fat content (>5%) can coat the fibre thus reducing the hydrophylicity of feed particles and interfering with its digestion by rumen microbes [17]. Marula seed cake as a protein supplement has an appreciable level of EE content (28.96% DM) while SBM has small amounts of fat (1.7% [25]; 2.07% obtained by Feed Master, Swaziland). Therefore, since the digestibility of a feed is affected by the fat content of the feed, MSM would be expected to have lower digestibility 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 [40] in the MSM (1.02% Ca and 0.34% Mg) and SBM+MSC (0.99% Ca and 0.34% Mg) diets. [34] 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 to render them largely inactive [28], however, this is subject for further research.

4. CONCLUSION

The results have indicated that Marula seed cake is highly digestible and rich in protein and crude fat. Thus it can be solely used or incorporated in dairy meal as a protein supplement when formulating dairy cattle diets.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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