Effect of replacing wheat bran with different levels of dried *Sesbania sesban* leaf meal on nutrient digestibility and rumen fermentation characteristics in Awassi lambs

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Abstract

A digestibility trial was conducted on a private sheep farm in Al-Musayyib District, Babylon Province, Iraq, to evaluate the effects of replacing wheat bran with dried *Sesbania sesban* leaf meal (SES) on nutrient digestibility and rumen fermentation characteristics in Awassi lambs. Twenty Awassi male lambs (4–5 months old) with an initial body weight of 21.72 ± 0.54 kg were randomly allocated to four dietary treatments: SES0 (control), SES10, SES20, and SES30, in which dried *Sesbania sesban* leaf meal replaced wheat bran at 0, 10, 20, and 30%, respectively. Lambs were offered the concentrate mixture at 2.5% of live body weight, while wheat straw was provided *ad libitum*. No significant differences ($P > 0.05$) were observed in the digestibility of dry matter, organic matter, or nitrogen-free extract among the dietary treatments. However, the digestibility coefficients of crude protein, crude fiber, and ether extract were significantly higher ($P < 0.05$) in lambs fed the SES20 diet than in the other treatment groups. Dietary treatments had no significant effect ($P > 0.05$) on ruminal pH or ammonia nitrogen concentration. In contrast, total volatile fatty acid concentration and total bacterial count increased significantly ($P < 0.05$), with the highest values recorded in the SES20 group. These findings indicate that replacing wheat bran with 20% dried *Sesbania sesban* leaf meal enhances nutrient digestibility and rumen fermentation without adversely affecting ruminal pH or ammonia nitrogen concentration. Therefore, dried *Sesbania sesban* leaf meal can be considered a promising alternative feed ingredient for Awassi lambs.

Keywords: *Sesbania sesban*; Awassi lambs; Digestibility; rumen fermentation; Alternative feed resources.

Introduction

In many developing countries, the limited and unpredictable availability of conventional feed resources throughout the year is a major constraint to sustainable ruminant production [12]. In addition, one of the major challenges facing ruminant nutrition, particularly during the summer season, is the shortage of green forages in both quantity and quality [8]. Moreover, increasing competition between humans and livestock for cereal grains and other conventional feed ingredients has substantially increased the cost of concentrate feeds used in ruminant production [25]. Consequently, identifying alternative and sustainable feed resources has become essential for improving ruminant productivity while reducing feeding costs [21]. Among the available alternatives, forage tree legumes have attracted considerable attention because of their high nutritional value and adaptability to harsh environmental conditions. *Sesbania sesban* is a multipurpose

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leguminous shrub widely distributed throughout tropical regions of Africa and Asia [16]. In addition to its role in reforestation programs and combating desertification in arid and semi-arid regions [2], *Sesbania sesban* is considered a valuable feed resource for ruminants. It can be fed either fresh or as hay and is characterized by a high crude protein content (up to 18.3%), relatively low fiber concentration, and high digestibility, making it particularly suitable for feeding small ruminants [8]. Moreover, *Sesbania* is highly palatable to sheep, resulting in good voluntary feed intake and making it a practical alternative to conventional feed ingredients [19]. Furthermore, partial replacement of the concentrate mixture with *Sesbania* has been reported to reduce feed costs without compromising animal performance [11; 28]. Numerous studies have evaluated the use of *Sesbania sesban* in ruminant diets; however, the reported responses have been inconsistent. Improvements in nutrient digestibility have been reported in dairy cows [10], sheep [19;23;24], and lambs supplemented with moderate levels of *Sesbania*. Conversely, [7] reported no significant effects on nutrient digestibility in goats or reduced nutrient digestibility when applied in high levels in dairy cows [13]. These inconsistent results need further investigation.

Therefore, the present study was conducted to evaluate the effects of replacing wheat bran with different levels of dried *Sesbania sesban* leaf meal on nutrient digestibility and rumen fermentation characteristics in Awassi lambs.

Material and Methods

All procedures involving animals were carried out in accordance with the ethical guidelines for the care and use of animals in scientific research. The study protocol received ethical approval from the Central Bioethics Committee at the University of Kufa (Approval No. 2327/2026).

Animal, Diet and Digestion trail:

A field experiment was conducted on a private sheep farm located in Al-Musayyib District, Babylon Province, Iraq (32° 45′ 24.7″ N, 44° 16′ 11.6″ E), using twenty healthy Awassi male lambs. Before the start of the experiment, all animals were treated against internal and external parasites. Fresh leaves of *Sesbania sesban* were collected from gardens and parks in Babylon Province, sun-dried for 72 h, ground using an electric grinder, and stored in airtight containers until incorporation into the experimental diets. The lambs were individually housed in semi-shaded pens (2 × 2 m), each equipped with separate feeders for concentrate and wheat straw and an automatic water supply. The experiment consisted of a 21-day adaptation period followed by a 30-day digestibility trial, during which feces were collected for the final 7 consecutive days. All lambs were offered a concentrate mixture consisting of barley grain, yellow corn grain, wheat bran, soybean meal, limestone, common salt, and a vitamin–mineral premix at 2.5% of their live body weight, while wheat straw was provided *ad libitum*. The experimental diets were as follows: SES0, a control diet containing no Sesbania leaf meal; SES10, a diet in which 10% of the wheat bran was replaced with dried Sesbania leaf meal; SES20, a diet in which 20% of the wheat bran was replaced with dried Sesbania leaf meal; and SES30, a diet in which 30% of the wheat bran was replaced with dried Sesbania leaf meal. Feces excreted were collected using fecal collection bags, transferred to plastic buckets, weighed, and recorded. A representative subsample equivalent to

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15% of the daily fecal output was collected from each lamb and stored at −18 °C. At the end of the collection period, subsamples from each animal were composited, dried in a forced-air oven at 60 °C for 48 h until constant weight, ground, and stored in airtight plastic containers for subsequent chemical analyses. Clean drinking water and mineral blocks were available to all lambs throughout the experimental period.

Chemical analysis:

Representative samples of wheat straw, concentrate mixture, dried *Sesbania sesban* leaf meal, and feces (dried at 60 °C for 48 h) were ground using a laboratory mill fitted with a 1-mm sieve prior to chemical analysis. The samples were analyzed for dry matter (DM), ash, crude protein (CP), crude fiber (CF), and ether extract (EE) according to the standard procedures [3]. Organic matter (OM) content was calculated as the difference between dry matter and ash, while nitrogen-free extract (NFE) was calculated by difference. The ingredient and chemical composition of the experimental diets are presented in Table 1.

Table 1. Feed ingredients (%) and Chemical composition of experimental diet used in the study (On DM% basis)

Feed ingredients (%)	Wheat straw	Sesbania leaves	SES0	SES10	SES20	SES30
Barley grain	-	-	35	35	35	35
Yellow corn grain	-	-	15	15	15	15
Wheat bran	-	-	40	30	20	10
Soybean meal	-	-	8	8	8	8
Sesbania leaf meal	-	-	0	10	20	30
Limestone	-	-	0.5	0.5	0.5	0.5
Common salt	-	-	0.5	0.5	0.5	0.5
Premix	-	-	1	1	1	1
Chemical composition (%)						
Dry matter (DM)	91.12	90.68	98.22	89.39	89.56	89.72
Ash	7.83	8.28	5.65	5.79	5.92	6.06
Organic matter (OM)	92.17	91.72	94.35	94.21	94.08	93.94
Crude Protein (CP)	2.62	16.83	15.62	15.59	15.57	15.54
Crude fiber (CF)	40.96	24.58	7.43	8.75	10.08	11.41
Ether extract (EE)	1.81	2.85	3.25	3.1	2.94	2.79
NFE	46.78	47.65	68.05	66.77	65.49	64.20
ME(MJ/100gDM)	0.95	1.08	1.28	1.26	1.25	1.23

0 SES: Control diet (containing 0% dried Sesbania leaf meal); 10 SES: Diet containing 10% dried Sesbania leaf meal; 20 SES: Diet containing 20% dried Sesbania leaf meal; 30 SES: Diet containing 30% dried Sesbania leaf meal; NFE: Nitrogen free extract= OM-(CP+CF+EE); ME: Metabolizable energy (MJ / 100g DM) = 0.12×CP + 0.31×EE + 0.05 ×CF+ 0.14 ×NFE [15].

Rumen Fermentation Parameters

Rumen fluid samples were collected from all lambs on the final day of the digestibility trial at 3h after the morning feeding. Approximately 20 mL of rumen fluid was collected from each animal using a stomach tube attached to a hand-operated suction pump. Immediately after collection, the pH of each sample was

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measured using a calibrated digital pH meter (HANNA, China). The rumen fluid was then filtered through four layers of cheesecloth to remove feed particles. Before preservation, an aliquot of freshly filtered rumen fluid was serially diluted and cultured on the appropriate medium for determination of the total bacterial count using the standard plate count method. Results were expressed as colony-forming units per milliliter (CFU/mL). The remaining filtrate was preserved by adding a few drops of 0.1 N sulfuric acid ($H_2 SO_4$) and phosphoric acid ($H_3 PO_4$), followed by centrifugation at 5,000 rpm for 20 min. The resulting supernatant was transferred into clean tubes and stored at −18 °C until chemical analysis. Ruminal ammonia nitrogen (NH_3 -N) concentration was determined according to the standard procedures [3]. Total volatile fatty acid (TVFA) concentration was determined by steam distillation using a Markham microdistillation apparatus, following the method described by [27].

Statistical Analysis

Data were analyzed using analysis of variance (ANOVA) under a completely randomized design (CRD) with SAS software [22]. When significant treatment effects were detected, means were compared using Duncan's multiple range test [6]. Differences were considered statistically significant at $P < 0.05$.

Results and Discussion

Nutrient Digestibility Coefficients

Results The effects of feeding different levels of dried *Sesbania sesban* leaf meal on nutrient digestibility coefficients in Awassi lambs are presented in Table 2. Dietary inclusion of dried *Sesbania* leaf meal had no significant effect ($P > 0.05$) on the digestibility of dry matter (DMD), organic matter (OMD), or nitrogen-free extract (NFED). Although the SES20 treatment recorded numerically higher values for DMD (73.99%), OMD (75.99%), and NFED (80.48%) than the other treatments, these differences were not statistically significant. In contrast, the digestibility coefficients of crude protein (CPD), crude fiber (CFD), and ether extract (EED) were significantly affected ($P < 0.05$) by dietary treatment. Lambs fed the SES20 diet exhibited the highest digestibility coefficients for CP (74.17%), CF (64.71%), and EE (61.76%), whereas the lowest values were recorded in the SES30 treatment. These results indicate that replacing wheat bran with dried *Sesbania* leaf meal at a 20% inclusion level enhanced the utilization of protein, fiber, and fat without adversely affecting the overall digestibility of dry matter or organic matter.

Table 2. The effect of feeding different levels of dried Sesbania sesban leaves on diet digestibility coefficients in Awassi lambs (% ± SE)

Digestibility (%)	Treatments (% Sesbania leaf meal replacing wheat bran)				P-value
	SES0	SES10	SES20	SES30	
DMD%	71.63 ± 0.95	72.64 ± 3.10	73.99 ± 2.73	69.79 ± 0.23	N.S.
OMD%	74.81 ± 0.69	75.30 ± 2.30	75.99 ± 1.75	72.62 ± 1.03	N.S.
CPD%	73.52 ± 1.27^{a}	72.26 ± 1.05^{ab}	74.17 ± 0.53^{a}	69.64 ± 1.32^{b}	*
CFD%	62.15 ± 0.99^{ab}	62.61 ± 0.46^{ab}	64.71 ± 0.49^{a}	61.88± 0.55^{b}	*
EED%	61.38 ± 0.83^{ab}	61.47 ± 0.58^{ab}	61.76 ± 0.82^{a}	58.19 ± 1.48^{b}	*
NFED%	79.82 ± 0.56	80.15 ± 0.60	80.48 ± 1.10	79.46 ± 0.29	N.S.

Means within same raw with different letter are significantly different ($P \leq 0.05$), N.S.: Not significant; *: ($P \leq 0.05$);0 SES: Control diet (containing 0% dried Sesbania leaf meal); 10

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SES: Diet containing 10% dried Sesbania leaf meal; 20 SES: Diet containing 20% dried Sesbania leaf meal; 30 SES: Diet containing 30% dried Sesbania leaf meal; DMD: Dry matter digestibility, OMD: Organic matter digestibility, CPD: Crude protein digestibility, CFD: Crude fiber digestibility, EED: Ether extract digestibility, NFED: Nitrogen free extract digestibility .

The improvement observed at the 20% inclusion level may be attributed to the high nutritional value of *Sesbania sesban* leaves, particularly their protein content, which may have stimulated ruminal microbial growth and enzymatic activity, thereby enhancing nutrient digestion. However, increasing the inclusion level to 30% resulted in a decline in CP, CF, and EE digestibility, possibly because higher concentrations of secondary plant metabolites, such as tannins and saponins, can reduce microbial activity and nutrient availability through the formation of tannin–protein complexes and inhibition of fibrolytic microorganisms. Therefore, the present findings suggest that a moderate inclusion level (20%) of dried *Sesbania* leaf meal is more beneficial than higher inclusion levels for improving nutrient utilization in Awassi lambs. Similar findings were reported by several previous studies reporting that replacing part of the concentrate mixture with *Sesbania sesban* leaves had no significant effect on nutrient digestibility in sheep [4;8;28], goats [11; 14], and dairy cows [13]. [7] also found that nutrient digestibility in Zaraibi goats was maintained at moderate levels of *Sesbania* hay inclusion but declined significantly when the inclusion level reached 80% of the diet. Conversely, [21; 23; 24] reported significant improvements in nutrient digestibility in sheep fed diet supplementation with dried *Sesbania* leaves. These improvements were attributed to the high crude protein content and lower fiber concentration of *Sesbania*, which enhanced nutrient availability, stimulated ruminal microbial growth, and improved feed degradation [18]. In contrast, [17] observed reduced digestibility of OM, CP, EE, and CF at 20–30% inclusion of *Sesbania* hay in diet of Ossimi lambs, while [1] reported a reduction only in CP digestibility in sheep diet. These adverse responses were attributed to the presence of antinutritional compounds, particularly tannins and saponins, which can reduce ruminal microbial activity and nutrient utilization by forming tannin–protein complexes [9; 20].

Overall, the available evidence indicates that the response to *Sesbania sesban* supplementation is influenced by several factors, including the level of dietary inclusion, the composition of the basal diet, animal species, and the concentration of secondary plant metabolites, particularly tannins and saponins [8; 24]. In the present study, replacing wheat bran with 20% dried *Sesbania sesban* leaf meal improved the digestibility of crude protein, crude fiber, and ether extract without adversely affecting the digestibility of dry matter, organic matter, or nitrogen-free extract. However, increasing the inclusion level to 30% tended to reduce nutrient digestibility, possibly because higher concentrations of tannins and other phenolic compounds can form tannin–protein complexes, thereby reducing ruminal microbial activity and nutrient utilization.

Rumen Fermentation Parameters:

Table 3 presented rumen fermentation parameters in Awassi lambs fed different levels of dried *Sesbania* leaf meal. Dietary supplementation with dried *Sesbania* leaf meal had no significant effect ($P > 0.05$) on ruminal pH or ammonia nitrogen (NH_3 -N) concentration. Ruminal pH remained within the normal range (6.53–

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6.65) across all treatments. Moreover, all pH values recorded in the present study were within the optimal range required for the growth and activity of cellulolytic microorganisms [26].The absence of a significant effect of dried *Sesbania* leaf meal on ruminal pH is consistent with previous studies reporting that the inclusion of *Sesbania* leaves or *Sesbania* hay did not significantly affect ruminal pH in sheep [21;8], goats [7], or dairy cows [13].In contrast, other studies have reported either a significant decrease [28] or a significant increase [17] in ruminal pH following dietary supplementation with *Sesbania* leaves in sheep. Likewise, NH_3 -N concentrations in the rumen were comparable among treatments, suggesting that replacing wheat bran with dried *Sesbania* leaf meal maintained adequate ruminal nitrogen availability for microbial protein synthesis. Ruminal ammonia nitrogen (NH_3 -N) is the primary source of nitrogen required for microbial protein synthesis in the rumen and serves as an important indicator of ruminal protein degradation [5]. However, the NH_3 -N concentration observed in the present study did not decrease to a level that could adversely affect rumen fermentation or nitrogen metabolism, indicating that dietary inclusion of dried *Sesbania* leaf meal maintained an adequate supply of ruminal nitrogen to support normal microbial activity. Comparable results were obtained in goats [7], in sheep [8; 17] with no significant change in ruminal NH_3-N concentration when feeding sesbania. In contrast, other studies reported a decrease in ruminal NH_3 -N concentration when feeding sesbania in dairy cows [13] in sheep [21; 28].

Regarding total volatile fatty acid (TVFA) concentration and total bacterial counts in the current study were showed significantly affected ($P < 0.05$). Lambs fed the SES20 diet exhibited the highest TVFA concentration (11.57 mmol/100 mL), which was significantly greater than that of the control (SES0) and SES30 groups, while the SES10 treatment showed intermediate values. The increased TVFA concentration indicates enhanced microbial fermentation and greater production of fermentation end-products from dietary carbohydrates. Similar results were reported by [17;21] when incorporate sesbania leaves in the diet of Ossimi sheep. In contrast, [28] and [8] reported no significant changes in TVFA concentrations when sheep fed sesbania leaves. These discrepancies may be attributed to differences in diet composition, the concentrate-to-forage ratio, the level of *Sesbania* inclusion, and other experimental conditions among studies.

Similarly, the total bacterial count increased significantly with the inclusion of dried *Sesbania* leaf meal. The highest bacterial population was observed in the SES20 treatment (2.61×10^8 CFU/mL), followed by SES10, whereas SES30 showed an intermediate response and the control diet recorded the lowest bacterial count. These findings suggest that moderate inclusion of dried *Sesbania* leaf meal promoted ruminal microbial growth, possibly by providing an improved balance of fermentable nutrients and nitrogenous compounds. However, increasing the inclusion level to 30% did not further enhance bacterial proliferation, indicating that the optimal inclusion level under the present experimental conditions was 20%.

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Table 1. The effect of feeding different levels of dried Sesbania leaf meal on rumen fermentation parameters in Awassi lambs (Mean ± SE)

Fermentation parameters	Treatments (% Sesbania leaf meal replacing wheat bran)				P-value
	SES0	SES10	SES20	SES30	
pH	6.65± 0.07	6.57± 0.08	6.53±0.07	6.60±0.06	N.S.
NH3-N (mg.100mL−1)	24.46± 0.31	24.19± 0.40	23.57±0.37	24.30±0.38	N.S.
TVFA (mmol.100mL−1)	10.42±1.91^{c}	11.15±0.33^{ab}	11.57±0.11^{a}	10.70±0.23^{bc}	*
Total bacterial counts (CFU ×10^{8}.mL-1)	2.18±0.19^{c}	2.46± 0.13^{ab}	2.61± 0.1 ^{a}	2.32±0.11^{bc}	*

Means with different superscripts in the same row are significantly different ($P < 0.05$); NS: Not significant; NH_3-N : ruminal ammonia nitrogen; TVFA: Total volatile fatty acids 0 SES: Control diet (containing 0% dried Sesbania leaf meal); 10 SES: Diet containing 10% dried Sesbania leaf meal; 20 SES: Diet containing 20% dried Sesbania leaf meal; 30 SES: Diet containing 30% dried Sesbania leaf meal5%.

Conclusion

Collectively, the results of the present study demonstrate that replacing wheat bran with dried *Sesbania sesban* leaf meal at a 20% inclusion level improved nutrient digestibility, enhanced ruminal fermentation, and increased total bacterial counts without adversely affecting ruminal pH or NH_3 -N concentration. These findings indicate that a moderate inclusion level of dried *Sesbania* leaf meal can be considered a suitable alternative feed ingredient for Awassi lambs, whereas higher inclusion levels may reduce the nutritional benefits because of increased concentrations of antinutritional compounds.

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