

DIGESTIBILITY AND FEEDING VALUE OF IMPROVED TROPICAL GRASSES AND *Leucaena leucocephala* CULTIVARS FOR GOAT NUTRITION IN VIETNAM

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Supporting Information



ABSTRACT: This study evaluated the nutritive value and digestibility of five tropical grass cultivars, including Ruzi (*Brachiaria ruziziensis*), Mulato II (*Brachiaria ruziziensis* × *B. decumbens* × *B. brizantha*), and three *Panicum maximum* cultivars (Purple, Mombasa and Mun River), together with five *Leucaena leucocephala* cultivars (Tarramba, Peru, Cunningham, KU15 and KU56). *In vitro* digestibility was determined using ANKOM DAISYII incubator system, and an *in vivo* feeding trial evaluated the effects of increasing dietary inclusion of Tarramba leaves (0, 10, 20 and 30% of dietary dry matter) in Mombasa-based diets on nutrient intake and apparent digestibility in goats. Grass cultivars differed significantly ($P < 0.05$) in dry matter and fibre digestibility. No significant differences were detected among grass cultivars for *in vitro* dry matter, neutral detergent fibre or acid detergent fibre digestibility, although numerical differences were observed. Similarly, digestibility did not differ significantly among the five *Leucaena* cultivars, whereas *Leucaena* cultivars had substantially higher crude protein concentrations than the grasses. Increasing Tarramba inclusion resulted in a linear increase in crude protein intake and apparent crude protein digestibility while apparent dry matter and organic matter digestibility remained unaffected. Dry matter and organic matter intake showed quadratic responses to increasing Tarramba inclusion, whereas ADF digestibility decreased linearly and NDF digestibility showed a numerical decline. Under the conditions of the short-term digestibility trial, inclusion of Tarramba leaves up to 30% of dietary dry matter did not adversely affect apparent dry matter and organic matter digestibility. These findings indicate that improved tropical grasses and *Leucaena* can provide complementary forage resources for improving the nutritional quality of grass-based diets for goats.

Keywords: Apparent digestibility, Crude protein, Goat nutrition, Tropical forages, *Leucaena leucocephala*.

INTRODUCTION

Ruminant production plays a vital role in food security, rural livelihoods and the sustainable utilisation of natural resources throughout the tropics. However, productivity is frequently constrained by an inadequate supply of high-quality forage, particularly during periods of seasonal feed shortage (Cooke et al., 2025; Mugoti et al., 2025). This challenge is especially evident in smallholder farming systems, where animal performance depends largely on locally available forage resources rather than purchased concentrates (Devendra and Thomas, 2002).

In Vietnam, increasing demand for meat and milk has driven the expansion of the ruminant sector over the past decade (MARD, 2020; FAO, 2021). Although the country has favourable climatic conditions for forage production, ruminant feeding systems continue to rely predominantly on native grasses, cultivated forages and crop residues, whose availability and nutritional quality vary markedly with season. In central Vietnam, seasonal drought and flooding can constrain forage availability and quality, contributing to fluctuations in the supply crude protein and digestible nutrients. These seasonal constraints can consequently limit feed intake, nutrient utilisation, and growth performance in goats and other ruminants (Dung et al., 2013; Nguyen et al., 2023).

Improving forage quality through the adoption of productive tropical grasses and protein-rich forage legumes has therefore become a key strategy for enhancing the productivity and sustainability of ruminant production systems (Da Silva et al., 2015; De Angelis et al., 2021). Improved cultivars of *Brachiaria* and *Panicum* are recognised for their high forage yield, persistence and adaptation to tropical environments (Da Silva et al., 2015). Among tropical forage legumes, *Leucaena leucocephala* is widely used as a protein-rich forage because of its high crude protein concentration, favourable digestibility, drought tolerance and ability to improve the nutritional balance of grass-based diets (Garcia et al., 1996; De Angelis et al., 2021). Numerous studies have reported that supplementation of grass-based diets with *Leucaena* can improve rumen nitrogen supply, nutrient digestibility, voluntary feed intake and animal performance.

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However, these responses vary among cultivars and dietary inclusion levels, and excessive inclusion may reduce feed utilisation because of anti-nutritional compounds, particularly mimosine (Phaikaew et al., 2012; Casanova-Lugo et al., 2014). Therefore, both cultivar selection and dietary inclusion level are critical for maximising the nutritional benefits of *Leucaena* while minimising potential adverse effects.

Several improved *Leucaena* cultivars have recently been introduced into Vietnam to enhance forage production under tropical conditions. Among these, the cultivar Tarramba exhibited superior adaptation, forage yield and nutritional characteristics compared with other introduced cultivars under local growing conditions (Liem et al., 2026). Previous studies also demonstrated that supplementation with Tarramba leaves improved growth performance, feed efficiency and carcass characteristics of Vietnamese goats (Liem et al., 2025), indicating its considerable potential as a protein supplement for grass-based feeding systems. However, information on the effects of Tarramba inclusion on nutrient intake and apparent nutrient digestibility in grass-based diets remains limited. Importantly, the selection of Tarramba for further feeding evaluation was based primarily on its agronomic performance and practical suitability under local conditions, rather than on superior *in vitro* digestibility relative to other *Leucaena* cultivars.

Despite recent advances in forage evaluation in Vietnam, comprehensive evaluations of the nutritive value and digestibility of improved tropical grass and *Leucaena* cultivars under local environmental conditions remain scarce. Furthermore, previous studies have generally focused on either forage quality or animal performance, whereas few have integrated laboratory evaluation of forage digestibility with *in vivo* assessment of nutrient intake and apparent digestibility. Linking these complementary approaches is essential for identifying forage cultivars with superior feeding value and developing evidence-based recommendations for incorporating protein-rich forage legumes into grass-based diets for small ruminants.

Therefore, this study integrated *in vitro* and *in vivo* approaches to evaluate the nutritive value and digestibility of improved tropical forages for goats in central Vietnam. Specifically, the objectives were to (i) compare the chemical composition and *in vitro* digestibility of five tropical grass cultivars and five *Leucaena leucocephala* cultivars, and (ii) determine the effects of increasing dietary inclusion of Tarramba *Leucaena leucocephala* leaves (0%, 10%, 20% and 30% of dietary dry matter) in a Mombasa grass-based diet on nutrient intake and apparent digestibility in goats. This study provides evidence to support forage cultivar selection and the development of nutritionally balanced and sustainable feeding strategies for goat production in Vietnam.

MATERIALS AND METHODS

Experiment 1: *In vitro* evaluation of tropical grasses and *Leucaena* cultivars

Five tropical grass cultivars, namely Ruzi (*Brachiaria ruziziensis*), Mulato II (*Brachiaria ruziziensis* × *B. decumbens* × *B. brizantha*), Purple (*Panicum maximum* cv. Purple), Mombasa (*Panicum maximum* cv. Mombasa) and Mun River (*Panicum maximum* cv. Mun River), were cultivated at the experimental farm of the University of Agriculture and Forestry, Hue University, Vietnam. The grasses were harvested at a regrowth age of 45 days in May 2024. Five *Leucaena leucocephala* cultivars (Tarramba, Peru, Cunningham, KU15 and KU56) were grown at the same site and harvested in March 2024 after a four-month regrowth period. Only the edible fractions (leaves and young stems) were collected for analysis.

For chemical composition analysis, three independent forage samples were collected from each grass cultivar and four independent forage samples were collected from each *Leucaena* cultivar at harvest. Thus, the chemical composition dataset comprised 15 grass samples and 20 *Leucaena* samples. Samples were collected from different locations within the harvested forage material and were treated as independent samples for statistical analysis.

For the *in vitro* digestibility assay, two independent forage samples were selected from each cultivar, giving a total of 20 forage samples across the five grass and five *Leucaena* cultivars. Each independent forage sample was assigned to one ANKOM F57 filter bag and constituted one experimental observation. The two independent samples from each cultivar were allocated to two digestion jars, with one sample from each cultivar placed in each jar. Thus, each jar contained 10 forage samples, representing one sample from each of the 10 cultivars, and 2 blank bags. All 20 sample-containing bags and 4 blank bags were incubated simultaneously in the two digestion jars during one incubation run in the ANKOM DAISYII system. The two digestion jars were considered incubation vessels within the same run and were not treated as independent biological replicates. All forage samples were oven-dried at 60 °C for 72 h, ground to pass through a 1-mm screen using a laboratory mill, sealed in airtight containers and stored at −20 °C until analysis.

Chemical analysis

Samples of all forage cultivars were analysed for dry matter (DM), organic matter (OM), crude protein (CP), neutral detergent fibre (NDF), acid detergent fibre (ADF), and acid detergent lignin (ADL) at the Laboratory of the University of Agriculture and Forestry, Hue University, Vietnam. The analytical procedures used for chemical composition are described in the following section.

***In vitro* digestibility**

In vitro digestibility was determined using the ANKOM DAISYII Incubator (ANKOM Technology Corporation, Fairport, NY, USA) at the Department of Animal Science, Faculty of Agriculture at Kamphaeng Saen, Kasetsart University, Thailand. Approximately 0.50 g of each dried forage sample was weighed into a pre-weighed ANKOM F57 filter bag that had been pre-rinsed with acetone and dried. Each filter bag contained one of the two independent forage samples collected for each cultivar; no additional filter bags were used as technical replicates of the same forage sample. Empty bags were included as blanks to correct for changes in bag weight during incubation. The 20 sample-containing bags and 4 blank bags were distributed between the two digestion jars, such that each jar contained 10 sample bags and 2 blank bags, representing one independent sample from each cultivar.

The artificial buffer consisted of two solutions. Buffer A contained KH_2PO_4 (10.0 g/L), $\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$ (0.5 g/L), NaCl (0.5 g/L), $\text{CaCl}_2 \cdot 2\text{H}_2\text{O}$ (0.1 g/L) and urea (0.5 g/L), whereas Buffer B contained Na_2CO_3 (15.0 g/L) and $\text{Na}_2\text{S} \cdot 9\text{H}_2\text{O}$ (1.0 g/L). Both buffers were prepared with distilled water according to the ANKOM protocol. Fresh rumen fluid was collected before the morning feeding from two rumen-fistulated beef steers weighing approximately 320–330 kg. The steers were fed rice straw *ad libitum* as roughage and 2 kg/day of a concentrate diet containing 12% crude protein. The diet was offered twice daily, and fresh water was available freely. Rumen fluid collected from the two steers was mixed and filtered through four layers of cheesecloth. The pooled rumen fluid was maintained under oxygen-free conditions and continuously flushed with CO_2 until preparation of the rumen inoculum according to the ANKOM DAISYII protocol. The pooled rumen fluid from the two donor steers was used as a common inoculum for all samples; therefore, donor animals were not considered independent experimental replicates.

For each digestion jar, 1330 mL of Buffer A and 266 mL of Buffer B were added to each digestion jar and equilibrated at 39 °C for approximately 30 min before adding 400 mL of rumen fluid. The jars were then incubated in the DAISYII incubator at 39 °C for 48 h. Both digestion jars were incubated simultaneously during the same incubation run. At the end of incubation, the filter bags were thoroughly rinsed with cold water to terminate microbial fermentation and remove residual digesta before being dried to constant weight. *In vitro* dry matter digestibility (IVDMD) was calculated after correcting for blank bag weight changes. Residues remaining after incubation were subsequently analysed for NDF and ADF using the ANKOM fibre analyser to determine *in vitro* neutral detergent fibre digestibility (IVNDFD) and *in vitro* acid detergent fibre digestibility (IVADFD), respectively. For statistical analysis, the two independent forage samples per cultivar were considered the experimental observations. Filter bags and digestion jars were not treated as additional experimental replicates.

In vitro digestibility variables were calculated as follows: $\text{IVDMD (\%)} = [1 - ((W_3 - W_1 \times C_1)/W_2)] \times 100$

where W_1 is the initial weight of the empty filter bag (g), W_2 is the sample weight (g), W_3 is the final weight of the filter bag after incubation and drying (g), and C_1 is the blank bag correction factor.

$\text{IVNDFD (\%)} = [1 - (\text{NDF residue after incubation} / \text{initial NDF content})] \times 100$

$\text{IVADFD (\%)} = [1 - (\text{ADF residue after incubation} / \text{initial ADF content})] \times 100$

Experiment 2: Effects of Increasing Dietary Inclusion of Tarramba *Leucaena leucocephala* on Feed Intake and Apparent Nutrient Digestibility in Goats

Animals and experimental design

Sixteen male Co goats, a local Vietnamese breed, with an initial body weight of 12.4 ± 1.3 kg (mean $\pm$ SD), were used in this experiment. Prior to the experiment, all goats were vaccinated against pasteurellosis, haemorrhagic septicaemia and foot-and-mouth disease and were treated with anthelmintics to ensure uniform health status. The goats were randomly allocated to four dietary treatments in a completely randomized design, with four goats per treatment. Each goat was housed individually in a separate pen equipped with an individual feeder and continuous access to clean drinking water throughout the experimental period.

Experimental diets and feeding management

Guinea grass (*Panicum maximum* cv. Mombasa) and *Leucaena leucocephala* cv. Tarramba used in the feeding trial were cultivated at the Institute for Development Studies, University of Agriculture and Forestry, Hue University, Vietnam. Tarramba was selected for Experiment 2 primarily because of its favourable agronomic adaptation and forage yield under the environmental conditions of central Vietnam, as previously reported by Liem et al. (2026), rather than because of superior *in vitro* digestibility among the *Leucaena* cultivars evaluated in Experiment 1. Guinea grass was harvested at a 45-day regrowth period, whereas Tarramba was harvested after a four-month regrowth period. Only the edible portions were collected daily, chopped and thoroughly mixed before being offered fresh to the animals.

Four experimental diets were formulated by replacing Guinea grass with increasing proportions of Tarramba leaves (0%, 10%, 20% and 30% of dietary DM). This resulted in Guinea grass: *Leucaena* ratios of 100:0, 90:10, 80:20 and 70:30 on a DM basis, respectively. The chemical composition of the feed ingredients and the experimental diets is presented in Tables 1 and 2, respectively.

Goats were fed twice daily at 09:00 and 15:00 h. During the 20-day adaptation period, the daily feed allowance was initially estimated at approximately 4% of body weight on a DM basis and was adjusted according to each goat's observed feed intake. During the subsequent 6-day measurement period, the daily amount offered to each goat was set at 90% of its individual average daily DM intake during the adaptation period to minimise feed refusals and facilitate accurate measurement of nutrient intake. Any feed refusals were collected before the morning feeding, weighed, and recorded daily. The quantities of fresh Guinea grass and *Leucaena* offered were adjusted according to their measured DM concentrations to maintain the intended dietary inclusion levels on a DM basis. Fresh drinking water was available ad libitum throughout the experiment.

Table 1 - Chemical composition of forage ingredients used in the feeding trial

Ingredients	DM (%)	Chemical composition (g/kg DM)					
		CP	EE	NDF	ADF	ADL	Ash
Guinea grass (<i>Panicum maximum</i> cv. Mombasa)	24.3	82	21	722	413	79	74
<i>Leucaena</i> leaves (<i>Leucaena leucocephala</i> cv. Tarramba)	33.2	254	72	330	192	87	70

Note: Values are expressed on a dry matter basis unless otherwise indicated. DM, dry matter; CP, crude protein; EE, ether extract; NDF, neutral detergent fibre; ADF, acid detergent fibre; ADL, acid detergent lignin.

Table 2 - Ingredient composition and calculated chemical composition of the experimental diets

Variables	<i>Leucaena</i> inclusion level (% of dietary DM)			
	0	10	20	30
Ingredient composition (kg/100 kg DM)				
Guinea grass	100	90	80	70
<i>Leucaena</i> leaves	0	10	20	30
Total	100	100	100	100
Calculated chemical composition (g/kg DM)				
OM	926	926	927	927
CP	82	99	116	134
EE	21	26	31	36
NDF	722	683	644	604
ADF	413	391	369	347
ADL	79	80	81	81
ME (MJ/kg DM)	9.10	9.32	9.54	9.76

Note: Values were calculated from the analysed chemical composition of the individual forage ingredients. DM, dry matter; OM, organic matter; CP, crude protein; EE, ether extract; NDF, neutral detergent fibre; ADF, acid detergent fibre; ADL, acid detergent lignin; ME, metabolisable energy.

Digestibility trial and sample collection

The digestibility trial lasted 26 days, including a 20-day adaptation period followed by a 6-day measurement period. Goats were adapted to the experimental diets and housing conditions during the first 20 days. Feed intake was measured from days 21 to 26, while quantitative faecal collection was performed from days 22 to 26. During the measurement period, the amounts of feed offered and refused were recorded daily for each goat. Samples of offered feeds and refusals were collected daily, weighed and oven-dried at 65 °C for 24 h to determine DM intake. Faeces were collected through the mesh-floor faecal collection system of individual metabolism cages and recovered at 08:00, 14:00 and 18:00 h each day. The total faecal output from each goat was weighed and recorded daily, thoroughly mixed, and a representative subsample corresponding to 20% of the daily faecal output was collected and stored at –20 °C until chemical analysis.

Chemical analysis

Forage samples, feed ingredients, experimental diets and faecal samples were analysed for dry matter (DM), organic matter (OM), crude protein (CP), ether extract (EE) and ash according to AOAC (1990). Neutral detergent fibre (NDF), acid detergent fibre (ADF) and acid detergent lignin (ADL) were analysed according to Van Soest et al. (1991). All chemical analyses were conducted at the Laboratory at the University of Agriculture and Forestry, Hue University, Vietnam. For Experiment 2, apparent nutrient digestibility coefficients were calculated as follows:

$$\text{Apparent digestibility (\%)} = [(\text{nutrient intake} - \text{faecal nutrient output}) / \text{nutrient intake}] \times 100$$

Statistical analysis

Statistical analyses were performed using Minitab Statistical Software (Version 16; Minitab LLC, State College, PA, USA). Data from Experiments 1 and 2 were analysed separately using one-way analysis of variance (ANOVA).

For Experiment 1, the five tropical grass cultivars and five *Leucaena leucocephala* cultivars were analysed as two separate forage groups, with cultivar fitted as the fixed effect within each group. For chemical composition, three forage samples per grass cultivar ($n = 3$) and four forage samples per *Leucaena* cultivar ($n = 4$) were used as the experimental observations. For *in vitro* digestibility, two independently collected forage samples per cultivar ($n = 2$) were considered the experimental observations. Filter bags and digestion jars were not treated as additional experimental replicates.

For Experiment 2, dietary treatment was fitted as the fixed effect, and individual goats were considered the experimental units ($n = 4$ per treatment). The assumptions of ANOVA were assessed using the model residuals, including normality and homogeneity of variance, before interpretation of treatment effects. Initial body weight was considered as a potential covariate; however, because goats were randomly allocated to treatments and initial body weight did not differ significantly among treatments ($P = 0.447$), the primary analysis was conducted without covariate adjustment.

When a significant cultivar or treatment effect was detected ($P < 0.05$), means were separated using Tukey's honestly significant difference test. For Experiment 2, orthogonal polynomial contrasts were additionally used to assess linear and quadratic responses to increasing dietary *Leucaena* inclusion (0%, 10%, 20%, and 30% of dietary DM). Statistical significance was declared at $P < 0.05$, while P-values for linear and quadratic responses were reported separately. Data are presented as means $\pm$ standard deviation (SD). For Experiment 1, the standard error of the mean (SEM) values were calculated separately for the grass and *Leucaena* groups and are reported in Table 3 and 4. The SEM is also reported for Experiment 2.

RESULTS AND DISCUSSION

Chemical composition of tropical grass and *Leucaena* cultivars

The chemical composition of the five tropical grass cultivars and five *Leucaena leucocephala* cultivars is presented in Table 3. Among the tropical grasses, OM, CP, EE and ADL concentrations did not differ significantly ($P > 0.05$), whereas NDF and ADF varied markedly among cultivars ($P < 0.05$). Ruzi and Mulato II contained lower NDF (63.3 and 66.3% DM, respectively) and ADF (31.7 and 32.9% DM, respectively) concentrations than the three *Panicum maximum* cultivars (Purple, Mombasa and Mun River), which contained approximately 72% NDF and 41% ADF. These lower fibre concentrations are consistent with a greater potential for fibre degradation and may contribute to higher forage digestibility compared with the *Panicum* cultivars. In contrast, no significant differences were detected among the five *Leucaena* cultivars for any measured chemical component ($P > 0.05$). This suggests that the measured chemical characteristics were relatively consistent among the introduced *Leucaena* cultivars under the conditions of the present study. Although CP concentration ranged numerically from 23.3 to 27.9% DM, all cultivars consistently exhibited high protein concentrations together with substantially lower NDF and ADF concentrations than the grasses. This indicates that the introduced *Leucaena* cultivars showed relatively consistent nutritive characteristics under the environmental conditions of central Vietnam and have considerable potential as protein supplements in forage-based diets.

Table 3 - Chemical composition of five tropical grass cultivars and five *Leucaena leucocephala* cultivars (% DM)

Component	Ruzi	Mulato II	Purple	Mombasa	Mun River	SEM	P
OM	93.35 $\pm$ 0.87	92.78 $\pm$ 0.82	92.62 $\pm$ 0.90	92.60 $\pm$ 1.14	93.13 $\pm$ 1.19	0.57	0.852
CP	7.43 $\pm$ 0.61	8.22 $\pm$ 0.81	8.08 $\pm$ 0.30	8.15 $\pm$ 0.19	8.11 $\pm$ 0.24	0.28	0.350
EE	2.03 $\pm$ 0.06	2.26 $\pm$ 0.12	2.10 $\pm$ 0.20	2.09 $\pm$ 0.32	1.89 $\pm$ 0.38	0.14	0.523
NDF	63.30 $\pm$ 2.36 ^b	66.30 $\pm$ 0.60 ^b	72.70 $\pm$ 0.89 ^a	72.10 $\pm$ 0.76 ^a	72.50 $\pm$ 0.51 ^a	0.71	0.001
ADF	31.70 $\pm$ 1.64 ^b	32.90 $\pm$ 0.63 ^b	41.10 $\pm$ 0.82 ^a	41.29 $\pm$ 0.60 ^a	41.20 $\pm$ 1.06 ^a	0.59	0.001
ADL	7.11 $\pm$ 0.39	8.00 $\pm$ 0.54	7.59 $\pm$ 0.47	7.93 $\pm$ 0.37	7.27 $\pm$ 1.17	0.38	0.430
Component	Tarramba	Peru	Cunningham	KU15	KU56	SEM	P
OM	93.00 $\pm$ 0.58	93.10 $\pm$ 0.36	92.90 $\pm$ 0.68	93.00 $\pm$ 0.25	93.30 $\pm$ 0.61	0.26	0.778
CP	25.40 $\pm$ 3.35	26.90 $\pm$ 4.22	26.40 $\pm$ 3.44	23.30 $\pm$ 4.65	27.90 $\pm$ 1.38	1.79	0.454
EE	7.17 $\pm$ 0.81	6.33 $\pm$ 1.59	6.30 $\pm$ 1.33	6.35 $\pm$ 1.19	6.43 $\pm$ 1.08	0.61	0.836
NDF	33.02 $\pm$ 3.10	33.89 $\pm$ 1.40	33.61 $\pm$ 1.89	35.22 $\pm$ 1.63	36.50 $\pm$ 1.23	1.07	0.198
ADF	19.20 $\pm$ 1.55	19.87 $\pm$ 2.03	18.88 $\pm$ 1.17	20.08 $\pm$ 2.36	19.80 $\pm$ 2.44	0.98	0.898
ADL	8.72 $\pm$ 0.61	9.16 $\pm$ 0.67	9.36 $\pm$ 0.75	9.49 $\pm$ 0.57	9.53 $\pm$ 0.59	0.32	0.410

Note: Values are means $\pm$ SD ($n=3$ for tropical grass cultivars and $n=4$ for *Leucaena leucocephala* cultivars). SD, standard deviation; SEM, standard error of the mean. Means within a row followed by different superscript letters differ significantly according to Tukey's multiple comparison test ($P < 0.05$).

The contrasting chemical composition of grasses and *Leucaena* reflects their complementary roles in tropical ruminant feeding systems. Tropical grasses primarily provide structural carbohydrates required for normal rumen function, whereas *Leucaena* provides a concentrated source of crude protein that can supply nitrogen for microbial protein synthesis and ruminal fermentation. Similar compositional characteristics have been reported previously, with *Leucaena* consistently containing two- to three-fold greater CP concentrations and considerably lower fibre concentrations than tropical grasses (Takawale et al., 2016; Álvarez Ríos et al., 2018; Okoruwa, 2020).

Differences in fibre concentration among the grass cultivars are also consistent with their botanical characteristics. The lower NDF and ADF concentrations observed in Ruzi and Mulato II are consistent with their superior forage quality. Reduced cell wall content generally facilitates microbial degradation in the rumen and is closely associated with improved forage digestibility (Bacorro et al., 2018; Rojas García et al., 2020). These compositional differences therefore provide a biological explanation for the digestibility responses observed in the subsequent *in vitro* evaluation.

In addition to forage nutritive value, biomass productivity is another important criterion for cultivar selection and practical application in smallholder production systems. Under the same environmental conditions, our previous field evaluation showed that the grass cultivars produced approximately 15–24 t DM ha⁻¹ year⁻¹, whereas edible *Leucaena* biomass ranged from 4.0 to 5.3 t DM ha⁻¹ year⁻¹ (Liem et al., 2026). Despite producing less biomass than the tropical grasses, the markedly higher crude protein concentration of *Leucaena* enhances its value as a complementary forage rather than a replacement for grasses. Integrating productive grasses with protein-rich forage legumes has been widely recognised as an effective strategy for improving forage quality and reducing reliance on commercial protein supplements in tropical ruminant production systems (Casanova-Lugo et al., 2014; Cowley and Roschinsky, 2019).

Overall, Ruzi and Mulato II were characterised by lower fibre concentrations than the *Panicum* cultivars, indicating greater potential digestibility, whereas all *Leucaena* cultivars consistently provided high crude protein concentrations suitable for protein supplementation. Together, these compositional characteristics establish the nutritional basis for interpreting the *in vitro* digestibility results presented in the following section.

***In vitro* digestibility of tropical grasses and *Leucaena* cultivars**

The *in vitro* digestibility of the five tropical grass cultivars and five *Leucaena leucocephala* cultivars is presented in Table 4. Among the tropical grasses, cultivar effects were not statistically significant for IVDMD ($P = 0.063$), IVNDFD ($P = 0.052$) or IVADFD ($P = 0.091$), although numerical differences were observed among cultivars. Similarly, none of the digestibility indices differed significantly among the five *Leucaena* cultivars (IVDMD, $P = 0.490$; IVNDFD, $P = 0.141$; IVADFD, $P = 0.231$). Significant differences among grass cultivars were observed for IVDMD, IVNDFD and IVADFD ($P < 0.05$), whereas none of these digestibility indices differed significantly among the *Leucaena* cultivars ($P > 0.05$). The numerical variation in digestibility among the grasses was broadly consistent with differences in fibre composition (Table 3), suggesting that variation in cell wall characteristics may contribute to differences in forage digestibility.

Among the tropical grasses, Ruzi exhibited the numerically highest IVDMD, IVNDFD, whereas Purple had the highest numerical IVADFD. For IVDMD, Ruzi (60.79%) showed the highest value and was significantly higher than Mulato II, Mombasa and Mun River, while Purple had an intermediate value. For IVNDFD, Ruzi (52.39%) was significantly higher than Mombasa (39.06%) and Mun River (42.30%),

Although these differences were not statistically significant, the relatively higher digestibility values observed for Ruzi and Mulato II were consistent with their lower NDF and ADF concentrations. Lower concentrations of structural fibre generally increase the accessibility of plant tissues to rumen microorganisms and are associated with improved forage digestibility (Jung and Allen, 1995). Similar relationships between fibre concentration and digestibility have been reported for tropical *Brachiaria* cultivars (Maranatha et al., 2019; Lemus et al., 2021), supporting the potential feeding value of Ruzi and Mulato II as forage resources for ruminant production.

The five *Leucaena* cultivars showed numerical variation in *in vitro* digestibility, but none of the differences was statistically significant. Peru exhibited the highest IVDMD (56.73%), whereas KU15 had the lowest value (47.33%). Peru also showed the highest numerical IVNDFD (39.16%) and IVADFD (31.87%), while Tarramba had the lowest numerical values for IVNDFD (23.16%) and IVADFD (13.69%). However, the overall effects of cultivar were not significant for IVDMD ($P = 0.490$), IVNDFD ($P = 0.141$) or IVADFD ($P = 0.231$). These numerical differences should therefore not be interpreted as evidence of superior digestibility of any particular *Leucaena* cultivar under the conditions of the present study.

The numerical variation in *Leucaena* digestibility may reflect differences in plant maturity, tissue characteristics or secondary metabolites. One possible contributing factor was the relatively long regrowth interval (four months) used before harvest. As forage legumes mature, increasing cell wall lignification and changes in tissue structure generally reduce microbial accessibility and fibre degradation. Previous studies have likewise reported greater digestibility in younger *Leucaena* harvested at shorter regrowth intervals (Rengsirikul et al., 2011). In addition, secondary metabolites such as mimosine and condensed tannins may modify ruminal fermentation and nutrient utilisation by influencing microbial activity and protein degradation (Barros-Rodríguez et al., 2014; Yanuartono et al., 2019; Lakshmi et al., 2020). However, these compounds were not determined in the present study, and their contribution to the observed differences among cultivars therefore remains speculative.

The feeding value of *Leucaena*, however, should not be assessed solely on the basis of fibre digestibility. All cultivars contained approximately threefold higher crude protein concentrations than the tropical grasses (Table 3), indicating that their principal nutritional contribution is the supply of crude protein and nitrogen for ruminal microbial protein synthesis rather than structural carbohydrates. Supplementation with *Leucaena* can therefore improve rumen nitrogen availability, support microbial protein synthesis and potentially enhance the utilisation of low-protein tropical grasses. This complementary nutritional role has been widely demonstrated in grass-based feeding systems (Takawale et al., 2016; Álvarez Ríos et al., 2018; Okoruwa, 2020).

Although Peru exhibited numerically higher IVDMD, IVNDFD and IVADFD than Tarramba, these differences were not statistically significant. Digestibility alone was therefore not the determining criterion for cultivar selection in the subsequent feeding trial. Under the same environmental conditions in central Vietnam, our previous field evaluation demonstrated that Tarramba combined favourable nutritive value with good adaptation, high forage yield and persistence compared with other introduced cultivars (Liem et al., 2026). These agronomic characteristics are essential for sustainable forage production systems, where reliable biomass supply and field performance are as important as nutritive value. Consequently, Tarramba was selected as the cultivar for evaluating the effects of increasing *Leucaena* inclusion in practical goat feeding systems.

Overall, the present findings demonstrate the complementary nutritional functions of tropical grasses and *Leucaena* in forage-based diets. Ruzi and Mulato II exhibited relatively high fibre digestibility among the grass cultivars, with significant cultivar effects detected for IVDMD, IVNDFD and IVADFD, whereas all *Leucaena* cultivars showed no significant differences in any of the three digestibility indices. Nevertheless, all *Leucaena* cultivars consistently provided high crude protein concentrations despite the variation in their numerical fibre digestibility values. Together, these characteristics provide the nutritional basis the effects of increasing dietary inclusion of Tarramba on nutrient intake and apparent nutrient digestibility in goats under practical feeding conditions.

Table 4 - *In vitro* dry matter and fibre digestibility of five tropical grass cultivars and five *Leucaena leucocephala* cultivars after 48 h incubation using the DAISYII system

Cultivar	IVDMD (%)	IVNDFD (%)	IVADFD (%)
Grasses			
Ruzi	60.79	52.39	51.19
Mulato II	57.97	50.00	50.87
Purple	54.52	48.34	51.53
Mombasa	50.20	39.06	40.27
Mun River	50.99	42.30	44.06
SEM	0.81	1.37	1.45
P-value	0.063	0.052	0.091
<i>Leucaena</i> cultivars			
Tarramba	51.77	23.16	13.69
Peru	56.73	39.16	31.87
Cunningham	51.22	41.67	35.57
KU15	47.33	35.97	19.27
KU56	55.03	35.55	27.98
SEM	1.93	2.55	3.39
P-value	0.490	0.141	0.231

Values are means based on two independent forage samples per cultivar (n=2). IVDMD, *in vitro* dry matter digestibility. IVNDFD, *in vitro* neutral detergent fibre digestibility. IVADFD, *in vitro* acid detergent fibre digestibility. SEM, standard error of the mean.

Effects of increasing *Leucaena leucocephala* leaf inclusion on feed intake and apparent nutrient digestibility in goats

The effects of increasing dietary inclusion of *Leucaena leucocephala* cv. Tarramba leaves on feed intake and apparent nutrient digestibility are presented in Table 5. Increasing dietary inclusion of *Leucaena leucocephala* cv. Tarramba affected several nutrient intake variables, with the most pronounced response observed for crude protein intake, whereas apparent dry matter (DM) and organic matter (OM) digestibility were not affected by dietary treatment. The polynomial contrasts further identified linear or quadratic responses for several nutrient intake and digestibility variables. Dry matter and OM intake differed among dietary treatments (ANOVA; $P < 0.05$), and showed significant quadratic responses to increasing *Leucaena* inclusion (both $P < 0.05$), whereas the linear responses were not significant.

Table 5 - Feed intake and apparent nutrient digestibility of goats fed Mombasa grass-based diets containing increasing levels of *Leucaena leucocephala* leaves

Variables	Leucaena Inclusion levels (% of dietary DM)				SEM	ANOVA P	Linear P	Quadratic P
	0%	10%	20%	30%				
Initial body weight (kg)	12.35 ± 0.50	12.58 ± 0.75	12.65 ± 0.50	12.20 ± 0.43	0.26	0.447		
Feed intake (g/head/day)								
DM	316.7 ± 37.8 ^b	377.6 ± 29.5 ^a	375.2 ± 17.6 ^a	358.9 ± 13.4 ^{ab}	13.2	0.024	0.057	0.013
OM	296.0 ± 35.3 ^b	352.6 ± 27.6 ^a	350.0 ± 16.4 ^a	334.5 ± 12.5 ^{ab}	12.3	0.025	0.063	0.013
CP	30.5 ± 3.67 ^c	42.5 ± 3.4 ^b	48.5 ± 2.3 ^{ab}	52.3 ± 1.9 ^a	1.45	<0.001	<0.001	0.016
NDF	217.9 ± 25.9 ^{ab}	240.4 ± 18.7 ^a	219.1 ± 10.3 ^{ab}	191.0 ± 7.2 ^b	8.59	0.013	0.021	0.012
ADF	122.1 ± 14.5 ^{ab}	135.9 ± 10.5 ^a	125.0 ± 5.9 ^{ab}	110.6 ± 4.0 ^b	4.84	0.023	0.061	0.012
Apparent digestibility (%)								
DM	70.5 ± 6.5	72.8 ± 3.7	72.1 ± 2.9	72.7 ± 2.2	2.09	0.857	0.546	0.692
OM	71.2 ± 6.5	73.4 ± 3.7	72.5 ± 2.8	73.0 ± 2.3	2.07	0.884	0.622	0.692
CP	72.4 ± 6.3 ^b	75.8 ± 3.6 ^{ab}	78.7 ± 2.1 ^{ab}	80.7 ± 1.4 ^a	1.93	0.049	0.007	0.725
NDF	72.8 ± 6.9	72.9 ± 4.4	69.8 ± 3.4	67.5 ± 2.2	2.29	0.336	0.091	0.638
ADF	65.4 ± 7.7	64.7 ± 4.4	59.4 ± 3.8	56.4 ± 4.0	2.62	0.090	0.017	0.658

Note: Values are presented as means ± SD (n=4 per treatment). Different superscript letters within the same row indicate significant differences according to Tukey's multiple comparison test (P < 0.05). Linear and quadratic P-values were obtained using orthogonal polynomial contrasts for the quantitative inclusion levels (0, 10, 20, and 30% of dietary DM). DM, dry matter; OM, organic matter; CP, crude protein; NDF, neutral detergent fibre; ADF, acid detergent fibre; SEM, standard error of the mean.

Goats receiving diets containing 10% and 20% *Leucaena* consumed more DM and OM than those fed the grass-only diet, whereas intake at the 30% inclusion level was intermediate and did not differ significantly from the other treatments. The quadratic pattern indicates that intake increased at moderate inclusion levels and then tended to decline at the highest inclusion level, rather than increasing progressively with *Leucaena* inclusion. This response may reflect changes in the nutritional balance and physical characteristics of the diets following partial replacement of Mombasa grass with *Leucaena*. Comparable increases in feed intake following *Leucaena* supplementation have been reported in tropical small ruminants (Aregheore, 2002; Rusdy et al., 2020). The absence of a significant reduction in DM or OM intake at the 30% inclusion level indicates that feed intake was not adversely affected at the highest inclusion level tested under the conditions of the present study.

CP intake increased markedly with increasing *Leucaena* inclusion, from 30.5 g/head/day in the control diet to 52.3 g/head/day at 30% inclusion, and showed significant linear ($P < 0.001$) and quadratic ($P = 0.016$) responses. This response was consistent with the substantially higher CP concentration of *Leucaena* leaves than Mombasa grass observed in Experiment 1. NDF intake was affected by dietary treatment (ANOVA; $P = 0.013$) and showed both significant linear ($P = 0.021$) and quadratic ($P = 0.012$) responses. ADF intake also differed among treatments (ANOVA; $P = 0.023$) and showed a significant quadratic response ($P = 0.012$). For both NDF and ADF, intake was highest at 10% inclusion and declined at 30% inclusion, consistent with the lower fibre concentration of *Leucaena* relative to Mombasa grass.

Apparent DM and OM digestibility remained relatively high across all treatments (approximately 70–73%) and were not affected by dietary treatment ($P > 0.05$). Neither the linear nor quadratic contrasts were significant. These findings indicate that replacing up to 30% of Mombasa grass with Tarramba leaves did not alter overall DM or OM digestibility.

In contrast, apparent CP digestibility increased significantly from 72.4% in the control diet to 80.7% at 30% *Leucaena* inclusion and was affected by dietary treatment (ANOVA; $P = 0.049$). The significant linear contrast ($P = 0.007$), together with the non-significant quadratic contrast ($P = 0.725$), indicated a linear response in apparent CP digestibility across the inclusion levels. This response is consistent with the greater protein contribution of *Leucaena* leaves and may reflect improved apparent digestion of dietary CP as the proportion of *Leucaena* increased. Similar improvements in protein utilisation have been reported following the incorporation of forage legumes into low-protein tropical grass diets for goats and sheep (Garcia et al., 1996; Takawale et al., 2016; Álvarez Ríos et al., 2018; Okoruwa, 2020).

Although apparent NDF and ADF digestibility showed a numerical decline as *Leucaena* inclusion increased, the treatment ANOVA was not significant ($P > 0.05$). However, the orthogonal polynomial contrasts revealed a significant linear decline for ADF digestibility ($P = 0.017$), whereas NDF digestibility only showed a tendency toward a linear decrease ($P = 0.091$), with no significant quadratic responses ($P = 0.638$ and 0.658 , respectively). Thus, the decline in ADF digestibility was detected as a significant linear response across the inclusion gradient, despite the non-significant overall treatment effect in the ANOVA. NDF digestibility showed a numerical decline, with a non-significant tendency toward a linear response ($P = 0.091$). This pattern may be related to differences in the fibre characteristics of the dietary ingredients observed in Experiment 1. Nevertheless, the absence of a significant effect on DM and OM digestibility indicates that the observed reduction in fibre digestibility did not translate into a measurable reduction in overall organic nutrient digestion.

Overall, increasing Tarramba inclusion modified dietary nutrient intake and several digestibility responses. CP intake and apparent CP digestibility increased linearly with increasing *Leucaena* inclusion, whereas DM and OM intake showed quadratic responses. NDF and ADF intake also changed with increasing *Leucaena* inclusion, accompanied by linear declines in their apparent digestibility. Importantly, DM and OM digestibility remained unaffected across the inclusion levels tested. Under the conditions of the present short-term digestibility trial, inclusion of Tarramba leaves up to 30% of dietary DM did not adversely affect DM intake or apparent DM and OM digestibility.

CONCLUSION

This study demonstrated the complementary nutritional roles of improved tropical grasses and *Leucaena leucocephala* for goat feeding under the conditions of central Vietnam. The evaluated *Leucaena* cultivars contained substantially higher crude protein concentrations, highlighting their potential as protein-rich forage resources for grass-based diets. Although numerical differences in *in vitro* digestibility were observed among cultivars, no statistically significant differences were detected within either the grass or *Leucaena* groups.

In the feeding trial, increasing Tarramba *Leucaena* inclusion increased crude protein intake and apparent crude protein digestibility, with a significant linear response in the latter. Dry matter and organic matter intake showed quadratic responses to increasing Tarramba inclusion, whereas apparent dry matter and organic matter digestibility were not affected by dietary treatment. Apparent acid detergent fibre digestibility showed a significant linear decline, while apparent neutral detergent fibre digestibility showed only a numerical decline without a significant linear or quadratic response.

Overall, the findings provide a basis for considering improved forage cultivars and indicate the potential of Tarramba as a protein-rich, locally available forage resource for grass-based diets in smallholder production systems. Under the conditions of this short-term digestibility trial, inclusion of Tarramba leaves at up to 30% of dietary dry matter did not

adversely affect apparent dry matter or organic matter digestibility. However, the present study does not establish an optimum inclusion level or provide evidence on growth performance, nitrogen utilisation, health responses, or long-term production outcomes. Further studies assessing these responses under longer-term and practical feeding conditions are warranted to determine appropriate inclusion levels of Tarramba in goat diets.

DECLARATIONS

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Data availability

The datasets generated and analysed during the current study are available from the corresponding author upon reasonable request.

Author's contribution

T.N.Liem: Conceptualization, Investigation, Data curation, Formal analysis, Writing – original draft. N.M.Dung: Conceptualization, Investigation, Data curation, Writing – original draft. L.M.Duc: Conceptualization, Investigation, Data curation. D.T.Hai: Methodology, Formal analysis. L.V.An: Project administration, Conceptualization, Resources, Supervision, Validation, Writing – original draft, Writing – review & editing. All authors read and approved the final manuscript.

Ethical considerations

All animal procedures were reviewed and approved by the Animal Ethics Committee of Hue University, Vietnam (Approval No. HUVN0055). All animal procedures complied with the committee's guidelines for the ethical use of animals in research and were reported in accordance with the ARRIVE guidelines.

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Competing interests

The authors declare no competing interests in this research and publication.

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