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Nutritional composition and *in vitro* digestibility of unconventional fodder trees for ruminant feeding systems

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Abstract

Feed scarcity during the dry season is the single greatest constraint on smallholder ruminant productivity in the Ethiopian highlands, yet many indigenous and naturalised tree species with promising nutritional profiles remain underexplored as livestock feed resources. Leading animal nutrition scientists have long argued that integrating multipurpose fodder trees into crop–livestock systems could simultaneously address dry-season feed gaps, improve soil fertility through nitrogen fixation, and reduce dependence on expensive purchased concentrates. This research evaluated the proximate nutritional composition, fibre fractions, anti-nutritional factor concentrations, and *in vitro* dry matter digestibility (IVDMD) of leaf material from five unconventional fodder tree species *Moringa oleifera*, *Leucaena leucocephala*, *Sesbania sesban*, *Ficus thonningii*, and *Grewia ferruginea* collected from smallholder farms in three agro-ecological zones around Addis Ababa, Ethiopia.

Leaf samples were harvested during the late wet season (September–October 2022), air-dried, and analysed for dry matter (DM), crude protein (CP), neutral detergent fibre (NDF), acid detergent fibre (ADF), acid detergent lignin (ADL), ether extract (EE), ash, condensed tannins (CT), and total phenolics. IVDMD was determined using the two-stage Tilley and Terry method at six incubation time points (6, 12, 24, 48, 72, and 96 hours). *Moringa oleifera* had the highest CP content ($27.3 \pm 1.4\%$ DM), the lowest NDF ($28.7 \pm 2.1\%$ DM), and the highest 96-hour IVDMD (76.8%). *Leucaena leucocephala* ranked second in CP (23.8%) and IVDMD (72.1%) but contained the highest condensed tannin concentration (4.7% DM). *Grewia ferruginea* showed the lowest CP (14.1%) and IVDMD (57.8%). A strong negative correlation was found between condensed tannin content and 48-hour IVDMD ($r = -0.81$; $P < 0.01$). These results identify *Moringa oleifera* and *Leucaena leucocephala* as the most nutritionally promising candidates for supplementing low-quality basal diets in Ethiopian smallholder ruminant systems, with the caveat that *Leucaena* inclusion should be limited to avoid mimosine toxicity.

Keywords: Fodder trees, *in vitro* digestibility, crude protein, ruminant nutrition, *Moringa oleifera*, *Leucaena leucocephala*, anti-nutritional factors, condensed tannins, smallholder farming, Ethiopia

Introduction

According to leading ruminant nutritionists, the single most effective strategy for improving smallholder livestock productivity in sub-Saharan Africa is to close the gap between what animals need and what they actually eat during the dry season ^[1]. In the Ethiopian highlands, natural pastures and crop residues the dominant basal feeds decline sharply in both quantity and quality from November through May, with crude protein levels often dropping below the 7% threshold required for rumen microbial maintenance ^[2]. Animals fed on such diets lose body condition, reduce milk output, and become more susceptible to disease, creating a cycle of low productivity that traps farming households in poverty ^[3].

Multipurpose fodder trees offer a biologically elegant solution to this problem. Many tropical and subtropical tree species produce green, protein-rich foliage throughout the year, including during periods when herbaceous pasture has senesced ^[4]. Their deep root systems access moisture and nutrients unavailable to shallow-rooted grasses, and several genera particularly *Leucaena*, *Sesbania*, and *Gliricidia* fix atmospheric nitrogen through symbiotic rhizobia, enriching the soil beneath their canopies ^[5]. When fed to ruminants as a supplement to low-

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quality roughages, tree foliage can increase voluntary dry matter intake by 15–30%, boost rumen ammonia-nitrogen concentration above the threshold for efficient fibre digestion, and improve overall diet digestibility [6]. Despite these advantages, adoption of fodder trees remains patchy across Ethiopia. Part of the explanation is agronomic establishment requires fencing, watering, and patience during the first two years before harvest can begin but a significant part is nutritional uncertainty [7]. Farmers and extension agents need reliable data on the feeding value of locally available species to make informed planting and supplementation decisions. While Moringa and Leucaena have been characterised in West and South-East African contexts, their nutritional profiles can vary by ecotype, altitude, and harvest season, making extrapolation to Ethiopian conditions unreliable [8, 9]. Indigenous species such as Ficus thonningii and Grewia ferruginea are used informally by pastoralist communities but have received almost no formal laboratory evaluation [10].

Anti-nutritional factors (ANFs) add a layer of complexity. Condensed tannins, present at varying concentrations across tree species, bind dietary protein in the rumen, reducing its availability to microbes and the host animal. At moderate levels (2–4% DM), this binding can actually be beneficial by protecting protein from excessive rumen degradation and increasing the flow of amino acids to the small intestine. At higher levels (> 5% DM), however, tannins depress voluntary intake and overall digestibility [11]. Quantifying ANFs alongside proximate composition and digestibility is therefore essential for setting safe inclusion rates. This research aimed to characterise the nutritional composition, fibre fractions, anti-nutritional factor content, and *in vitro* dry matter digestibility of five fodder tree species available in the central Ethiopian highlands, and to rank them by overall feeding value for ruminant supplementation.

Material and Methods

Study area description

Leaf samples were collected from smallholder farms and roadside plantations in three agro-ecological zones within 80 km of Addis Ababa, Ethiopia: the mid-altitude sub-humid zone (1,800–2,200 m above sea level; Oromia Special Zone), the mid-altitude semi-arid zone (1,500–1,800 m; East Shewa Zone), and the highland humid zone (2,200–2,600 m; North Shewa Zone). These zones represent the range of altitudes and moisture regimes where fodder trees are commonly planted or naturally occurring in the central highlands [7].

Climatic and environmental conditions

Sampling was conducted during the late wet season (September–October 2022), when leaf biomass was at its peak and foliage quality was expected to be highest. Mean monthly temperatures during the collection period ranged from 14 °C at highland sites to 24 °C at the mid-altitude semi-arid sites. Cumulative rainfall for June–September 2022 ranged from 620 mm in the semi-arid zone to 1,140 mm in the highland humid zone, all within 10% of the 30-year average [3].

Material

Mature leaf material (including petioles but excluding woody stems > 5 mm diameter) was harvested from five tree species: Moringa oleifera, Leucaena leucocephala, Sesbania sesban, Ficus thonningii, and Grewia ferruginea. For each species, samples were collected from at least six individual trees across all three agro-ecological zones (minimum 18 trees per species). Leaves were air-dried under shade for five days, ground through a 1-mm sieve using a Wiley mill, and stored in airtight containers at room temperature until analysis at the Addis Ababa Institute of Animal Sciences nutrition laboratory [12].

Methods

Proximate analysis followed AOAC (2005) standard methods: dry matter (method 930.15), crude protein by Kjeldahl (method 984.13; $N \times 6.25$), ether extract by Soxhlet extraction (method 920.39), and ash by muffle furnace at 550 °C for 6 hours (method 942.05). Fibre fractions (NDF, ADF, ADL) were determined using the Van Soest detergent system with an ANKOM²⁰⁰ fibre analyser (ANKOM Technology, Macedon, NY, USA) [12]. Condensed tannins were quantified by the butanol–HCl method using purified quebracho tannin as the standard, and total phenolics by the Folin–Ciocalteu method expressed as tannic acid equivalents [11].

In vitro dry matter digestibility was measured by the two-stage procedure of Tilley and Terry (1963), modified for the ANKOM DaisyII incubator system. Rumen fluid was collected from two ruminally cannulated Horro sheep maintained on a mixed hay–concentrate diet. Samples (0.5 g) were incubated in buffered rumen fluid at 39 °C for 6, 12, 24, 48, 72, and 96 hours (stage 1), followed by pepsin–HCl digestion for 48 hours (stage 2). IVDMD was calculated as the percentage of DM disappearing after both stages [13]. Each species × time-point combination was run in triplicate. Data were analysed by one-way ANOVA with Tukey's post-hoc test; Pearson correlations were computed between chemical composition and IVDMD values. Significance was set at $P < 0.05$.

Ethical considerations

The use of ruminally cannulated sheep for rumen fluid collection was approved by the Addis Ababa Institute of Animal Sciences Ethical Review Committee (Protocol AAIS-ERC-2022-008, approved 4 August 2022). Animals were maintained under standard institutional guidelines with ad libitum access to water and a nutritionally adequate diet. No animals were euthanised for this research.

Results

All five species produced sufficient leaf biomass for triplicate analyses across the three agro-ecological zones. No significant zone × species interaction was detected for any chemical parameter ($P > 0.10$ for all), so data were pooled across zones for presentation. Species differences were significant ($P < 0.01$) for every measured variable.

Table 1: Proximate chemical composition (% DM, mean ± SD) of five fodder tree species from central Ethiopia

Species	CP	NDF	ADF	EE	Ash
<i>M. oleifera</i>	27.3 ± 1.4 ^a	28.7 ± 2.1 ^c	17.4 ± 1.6 ^c	6.8 ± 0.7 ^a	11.2 ± 0.9 ^a
<i>L. leucocephala</i>	23.8 ± 1.2 ^b	34.2 ± 2.8 ^d	21.6 ± 1.9 ^d	4.3 ± 0.5 ^b	8.7 ± 0.8 ^b
<i>S. sesban</i>	21.6 ± 1.3 ^{bc}	36.8 ± 3.1 ^d	24.3 ± 2.2 ^{bc}	3.7 ± 0.4 ^{bc}	9.4 ± 1.1 ^{ab}
<i>F. thonningii</i>	17.4 ± 1.1 ^c	41.3 ± 3.4 ^b	28.7 ± 2.6 ^b	3.1 ± 0.3 ^c	10.8 ± 1.3 ^a
<i>G. ferruginea</i>	14.1 ± 0.9 ^d	47.6 ± 4.1 ^a	33.2 ± 2.9 ^a	2.4 ± 0.3 ^c	7.3 ± 0.6 ^b

CP = crude protein; NDF = neutral detergent fibre; ADF = acid detergent fibre; EE = ether extract. Means in the same column with different superscript letters differ significantly ($P < 0.05$). *M. oleifera* had the highest CP and lowest fibre fractions; *G. ferruginea* had the lowest CP and highest fibre.

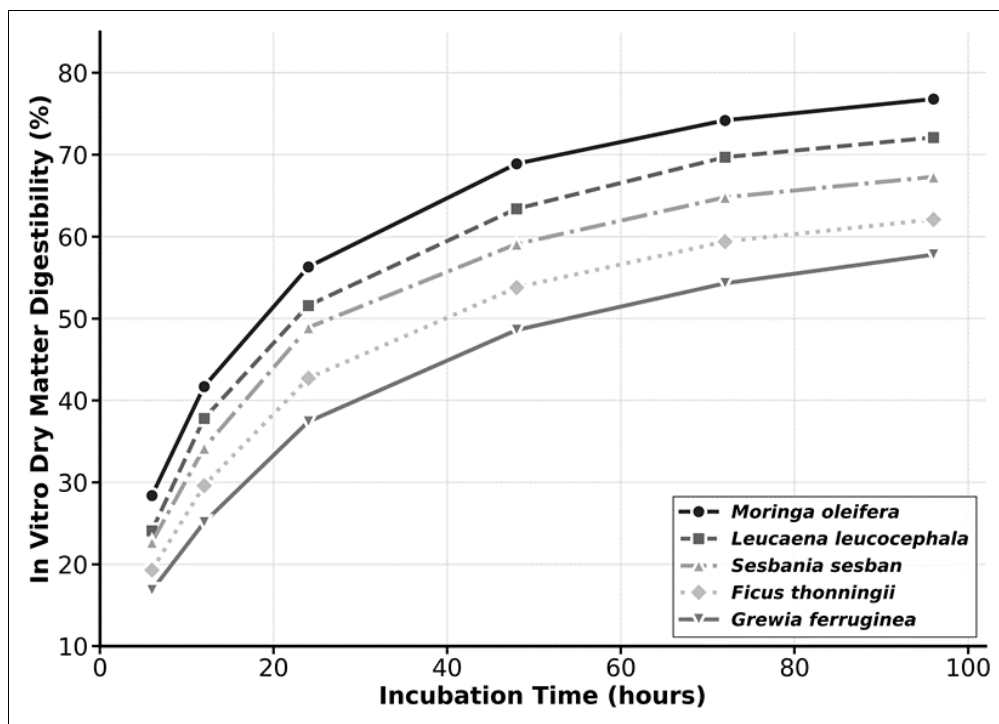


Fig 1: *In vitro* dry matter digestibility (%) of five fodder tree species at six incubation time points (6, 12, 24, 48, 72, and 96 hours). *Moringa oleifera* achieved the highest digestibility at all-time points, while *Grewia ferruginea* was consistently the lowest

Digestibility curves followed an asymptotic pattern for all species, with rapid increases between 6 and 48 hours and progressively smaller gains thereafter. By 96 hours, the

species ranking from highest to lowest IVDMD was: *M. oleifera* (76.8%) > *L. leucocephala* (72.1%) > *S. sesban* (67.3%) > *F. thonningii* (62.1%) > *G. ferruginea* (57.8%).

Table 2: Ranking of five fodder tree species by key nutritional parameters and anti-nutritional factor content

Rank	Species	CP (%)	IVDMD 96 h (%)	CT (% DM)	Phenolics (% DM)	Overall Rating
1	<i>M. oleifera</i>	27.3	76.8	1.2	2.8	Excellent
2	<i>L. leucocephala</i>	23.8	72.1	4.7	5.3	Good (with limits)
3	<i>S. sesban</i>	21.6	67.3	2.8	3.6	Good
4	<i>F. thonningii</i>	17.4	62.1	3.4	4.1	Moderate
5	<i>G. ferruginea</i>	14.1	57.8	5.1	6.7	Limited

CT = condensed tannins. Overall rating integrates CP, IVDMD, and ANF levels. *L. leucocephala* carries a “with limits” caveat because of its mimosine content (not measured here but well documented) and moderately high tannins

Table 3: Summary statistics for 48-hour *in vitro* dry matter digestibility and Pearson correlations with chemical composition variables (n = 5 species)

Variable	Mean	Sd	Range	R with ivdmd ₄₈
IVDMD 48 h (%)	58.8	7.9	48.6–68.9	
Crude protein (%)	20.8	5.1	14.1–27.3	0.94**
NDF (%)	37.7	7.1	28.7–47.6	−0.96**
Condensed tannins (%)	3.4	1.5	1.2–5.1	−0.81**
Total phenolics (%)	4.5	1.5	2.8–6.7	−0.88**

** $P < 0.01$. Crude protein showed the strongest positive correlation with IVDMD ($r = 0.94$), while NDF had the strongest negative association ($r = -0.96$). Both tannin measures were negatively correlated, confirming their inhibitory effect on digestibility

Biodiversity assessment

A brief floristic survey of the collection sites recorded between 8 and 14 tree species per farm, of which 3–5 were used informally as livestock feed. *M. oleifera* and *S. sesban* were found predominantly in the mid-altitude sub-humid zone, where they had been intentionally planted within homestead gardens or along field boundaries. *F. thonningii* and *G. ferruginea* occurred naturally across all three zones, often growing along watercourses and at forest margins. *L.*

leucocephala was present only in the semi-arid zone, where it had been introduced as part of a soil conservation programme in the 1990s. The density of fodder-suitable trees averaged 12.7 ± 4.3 trees per hectare of farmland, suggesting that meaningful quantities of leaf biomass estimated at 0.8–1.4 tonnes DM per hectare per year for *M. oleifera* under managed coppicing could be harvested without threatening tree survival [4, 5]

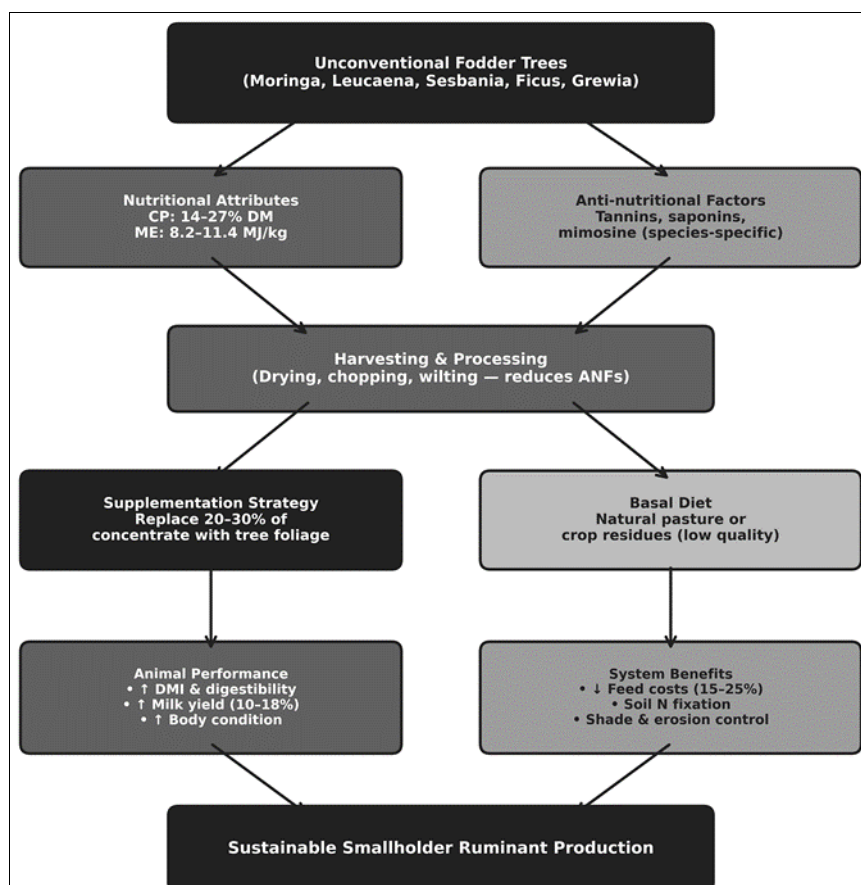


Fig 2: Conceptual model showing the pathway from unconventional fodder tree selection through nutritional characterisation, processing, supplementation strategy, and expected outcomes for animal performance and system sustainability in smallholder ruminant production

Comprehensive interpretation

The nutritional and digestibility data converge on a clear species hierarchy: *M. oleifera* stands out as the most promising fodder tree for ruminant supplementation in the Ethiopian highlands, combining the highest protein content, lowest fibre, lowest tannin levels, and highest *in vitro* digestibility. *L. leucocephala* ranks second but carries a caveat related to its mimosine content and moderately elevated tannins, both of which may limit safe inclusion rates. At the other end, *G. ferruginea*'s low protein, high fibre, and high tannin profile limit its value to situations where no better options are available. The strong correlations between chemical composition and IVDMD provide a practical shortcut: CP and NDF values alone can predict digestibility with reasonable accuracy, reducing the need for time-consuming *in vitro* assays in future screening exercises.

Discussion

The CP content of *M. oleifera* leaves in this research (27.3% DM) is consistent with the 25–30% range reported across multiple tropical countries [8, 9]. This positions Moringa as a protein supplement comparable to commercial concentrates such as noug cake (*Guizotia abyssinica*, ~29% CP) or cottonseed cake (~24% CP), but at a fraction of the cost for farmers who grow the trees on their own land [4]. The 96-hour IVDMD of 76.8% further supports this comparison, falling within the range typical of medium-quality legume hays rather than tree foliage [6].

Leucaena's strong nutritional profile (CP 23.8%, IVDMD 72.1%) is tempered by its condensed tannin content of 4.7% DM, which sits at the upper end of the “beneficial” window (2–4% DM) and approaches the level at which intake depression begins [11]. Additionally, *Leucaena* contains

mimosine, a non-protein amino acid that causes hair loss, poor growth, and thyroid suppression in ruminants fed more than 30% of their diet as *Leucaena* foliage [5]. For Ethiopian producers, a safe recommendation would be to cap *Leucaena* at 20–25% of the total ration dry matter, a level that captures the protein benefit while avoiding toxicity.

The poor performance of *G. ferruginea* (CP 14.1%, IVDMD 57.8%, CT 5.1%) is noteworthy because this species is used informally as emergency drought feed by pastoralist communities in the semi-arid lowlands. While its protein content technically exceeds the 7% minimum for rumen function, the high tannin burden likely reduces effective protein availability to the animal, and the elevated NDF (47.6%) slows passage rate and limits voluntary intake [2, 11]. Farmers relying on *Grewia* would benefit from simple processing techniques such as air-drying for 48 hours or mixing with wood ash that can reduce tannin bioactivity by 30–40% [14].

The strong negative correlation between condensed tannins and IVDMD ($r = -0.81$) across all five species reinforces the importance of quantifying ANFs alongside proximate composition in any feed evaluation programme. Relying on CP alone as a proxy for feed quality would overestimate the value of tannin-rich species like *Leucaena* and *Grewia*, potentially leading to inclusion rates that depress rather than improve animal performance.

Conclusion

Feed scarcity during the dry months continues to limit ruminant productivity across Ethiopia's mixed crop–livestock farming systems, and tapping the nutritional potential of locally available fodder trees is one of the most accessible strategies for closing the feed gap without incurring high

purchased-input costs. This research evaluated five tree species commonly found on or near smallholder farms in three agro-ecological zones around Addis Ababa and ranked them by a combination of crude protein content, fibre fractions, anti-nutritional factor levels, and *in vitro* dry matter digestibility. Several findings deserve emphasis. *Moringa oleifera* emerged as the top-ranked species across all measured parameters, with CP comparable to commercial protein concentrates, the lowest fibre and tannin levels, and the highest 96-hour IVDMD (76.8%). *Leucaena leucocephala* placed second with an excellent protein profile (23.8% CP) and good digestibility (72.1%) but requires careful inclusion rate management because of its mimosine content and moderately elevated condensed tannins. *Sesbania sesban* occupied a solid middle position, offering good protein (21.6%) with moderate tannins (2.8%), and may be particularly suited to wet-season supplementation when its fast-growing, coppiceable shoots provide abundant green biomass. *Ficus thonningii* and *Grewia ferruginea* showed lower overall potential, though they remain useful as emergency feeds when preferred species are unavailable.

We recommend that Ethiopian extension services prioritise *Moringa oleifera* for inclusion in smallholder agroforestry promotion programmes, targeting homestead garden boundaries and contour hedgerows where the tree can be coppiced at 60–90-day intervals to produce sustained leaf harvests. *Leucaena* should be promoted alongside *Moringa* but with explicit guidance on maximum dietary inclusion (20–25% of ration DM) and the availability of rumen inoculation with *Synergistes jonesii* bacteria to detoxify mimosine where *Leucaena* forms a larger share of the diet. *Sesbania* fits well in riparian and wetland-margin plantings where its fast growth and nitrogen-fixing capacity provide dual benefits for feed and soil improvement. Future research should address three gaps highlighted by this work. First, *in vivo* feeding trials are needed to confirm that the *in vitro* digestibility rankings translate into actual animal performance responses milk yield, weight gain, and reproductive efficiency under on-farm conditions. Second, seasonal sampling across wet and dry periods would reveal whether the nutritional profiles reported here (collected at peak quality in September–October) hold up during the critical dry months when supplementation is most needed. Third, economic analysis comparing the cost per kilogram of digestible protein from tree foliage versus purchased concentrates would provide farmers with the financial evidence needed to justify the labour investment required for tree establishment and management.

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