



Research article

Melia dubia Cav. leaf fodder *in-vitro* ruminal fermentation and metabolizable energy vary with provenance and season

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Received: 28th February, 2025

Accepted: 04th February, 2026

Abstract

Cueing the shortage of green fodder, especially during lean periods and other inadequacies in the animal feed fodder sector and despite an array of tree foliage being used traditionally and advocated scientifically, there is a need to explore more multipurpose fodder species with higher digestibility for inclusion in livestock rations. The present study analyzed *Melia dubia* leaf fodder *in-vitro* dry matter digestibility (IVDMD), *in-vitro* organic matter digestibility (IVOMD), *in-vitro* gas production (IVGP) and metabolizable energy (ME@24 hours) and to ascertain variation across provenance and seasons (winter and summer). Study revealed significant variation ($p \leq 0.05$) in IVDMD (47.74–64.97%), IVOMD (86.97–89.88%), and ME (1.35–1.83 kcal) among selected provenances. Leaf fodder of Ahwa (P8) showed higher IVDMD, and higher IVOMD was of Kaprada (P4), whereas the highest IVGP, after 24 hours, was for Chichinagawtha (P6) provenance leaf fodder. IVGP ranged from 24.37 to 38.95 mL @ 24 hours. Further, IVDMD and IVOMD were significantly higher (63.47 and 90.13%, respectively) for winter season fodder compared to summer. Whereas summer season leaf fodder recorded significantly maximum IVGP (35.69 mL, at 24 hours) and ME (1.73 kcal @ 24 hours) and compared to winter fodder. Results revealed that IVGP gradually increased up to 24 hours and then decreased till 92 hours. The interaction effect due to provenance and season for IVDMD, IVOMD, IVGP and ME was also significantly different. Studied indicated that *M. dubia* leaf fodder digestibility attributes are comparable with many other family tree species, as corroborated by available published data, commonly fed to ruminants and livestock. Thus, the study illustrated that *M. dubia* leaf fodder has good digestibility and can be used as an alternate top feed fodder source to livestock during the winter season.

Keywords: Gas production, Leaf fodder, *Melia dubia*, Organic matter digestibility,

Introduction

The three major sources of fodder supply in India are crop residues, cultivated fodder and common property resources like forests, permanent pastures and grazing lands. Currently, India faces a net deficit of 35.60% green fodder, 10.95% dry crop residues and 44% concentrate feed ingredients (IGFRI, 2015). In livestock production, feed and fodder cost about 2/3rd of the total cost. Any attempt towards enhancing the availability of feeds and fodder, and economizing the feed cost, would result in better compensation to livestock farmers. It is essential to replace low-yielding annual grasses with high-yielding perennial grasses and introduction of multipurpose tree species adaptable to diverse conditions to increase pasture productivity (Thakur *et al.*, 2015; Bhusara *et al.*, 2016). Many studies have supported this view that grasses, legumes and trees in a silvipastoral system

provide effective land cover as well as produce nutritious fodder and other edible plant parts for livestock (Rai and Palsaniya 2015). Particularly, tree foliage is an important component of diets for ruminant animals such as goats, sheep, cattle and deer and plays an important role in the nutrition of herbivores in areas where few or no alternatives are available. Tree fodder contains high levels of crude protein, minerals and many show high levels of digestibility (Kamalak *et al.*, 2010).

Cueing these gaps and inadequacies in the animal feed sector, and despite an array of tree foliage being used traditionally and advocated scientifically, there is still a need to explore more multipurpose fodder species with higher nutritive value and digestibility for inclusion in livestock rations. *Melia dubia* Cav., of family Meliaceae, is a much advocated multipurpose industrially important species (Parthiban *et al.*, 2019; Thakur *et al.*, 2023) and an

amenable agroforestry ideotype (Mohanty *et al.*, 2019; Thakur *et al.*, 2018; Chavan *et al.*, 2022). The species is indigenous to the Western Ghats region of India and also found in Bangladesh, Myanmar, Thailand, Mexico, Sri Lanka, Malaysia, Indonesia, China, the Philippines, Australia and African countries (Sukhadiya *et al.*, 2021). Various studies confirmed the presence of different nutritional compositions, phytochemicals and minerals in *M. dubia* leaves (Jeyaleela *et al.*, 2015; Parmar *et al.*, 2019). Although few studies pertaining to *M. dubia* fodder nutritive value have been carried out, particularly in southern India, fodder quality assessment studies based on *in-vitro* dry matter and organic matter digestibility, gas production, and metabolizable energy have not been explored yet. Hence, the present study intended to elucidate this lacking information. Therefore, the present study was conducted with the hypothesis that existing *in-vitro* dry matter/organic matter digestibility, gas production and metabolizable energy variations among provenances and seasons could be helpful in obtaining quality leaf fodder and a suitable harvesting season. The findings would further help improve the fodder quality through selections of better genotypes from the selected provenances.

Materials and Methods

Provenances and geo-climatic conditions: We selected eight provenances (Table 1; Fig 1) in Gujarat, India. P₁ & P₂ fall in the Satpura mountain ranges and P₃ to P₈ fall in the northern Western Ghats or Sahyadri mountain ranges. Sagai-1 (P₁) and Sagai-2 (P₂) provenances characterized with black cotton and stony soil type, with annual normal rainfall of 924.8 mm and average annual maximum and minimum temperature of the region are 34.7 and 20.7°C, respectively (average over 30 years). Nanapondha (P₃) and Kaprada (P₄) provenances are in the northern extension of the Deccan Plateau of Central India, belonging to late Cretaceous-early Eocene

age and followed by Quaternary sediments having black cotton soils. The maximum and minimum temperatures were 41.2 and 9.9°C, respectively. The average annual normal rainfall of Nanaponda and Kaprada is 2399 and 2842 mm, respectively. Waghai and Chichinagavtha, and Mahal and Ahwa provenances (P₅, P₆, P₇, and P₈, respectively) fall under the south and north Dangs region. Soils of this region are black rock outcrops, shallow black, brown and alluvial soils of recent origin. The geological structure of this region is composed of the Deccan Trap. The average annual rainfall in the district ranges from 1500 to 3000 mm. Average annual temperature is about 26°C and maximum temperature may shoot up to 46°C and the winter temperatures are 22°C.

Sample collection and processing: Five trees (each tree representing one replication) were selected randomly in each provenance for leaf fodder sample collection. Tree growth parameters, *viz.* average tree height and diameter at breast height (DBH) and average canopy area (m²) were recorded (Table 1) in all provenances.

Leaf samples were collected from all directions (north-south and east-west) and lower, middle and upper tree canopy. The samples were washed with distilled water in the laboratory and air-dried in a hot air oven at 60 ± 5°C till a constant weight was achieved. The leaf fodder of these selected provenances (P₁-P₈) has been reported that moisture content (MC), dry matter (DM), crude protein (CP), crude fiber (CF), ether extract (EE), organic matter (OM), total ash (TA) and nitrogen free extract (NFE) ranged from 68.6 to 72.2%; 27.8 to 31.5%, 10.2 to 12.1%, 16.3 to 18.7%, 2.5 to 3.3%, 85.4 to 88.7%, 11.3 to 14.6% and 55.0 to 58.4%, respectively. DM, CP, CF, EE and OM (29.7, 12.0, 18.6, 3.3 and 87.6%, respectively) were higher in the summer. In contrast, MC, TA, AIA and NFE (70.9, 13.6, 1.0, and 58.4%, respectively) were higher in the winter season. neutral detergent fiber (NDF), acid detergent fiber (ADF), acid detergent lignin (ADL), hemicellulose and cellulose content ranged from 32.2 to 38.5%, 23.8 to 30.2%,

Table 1. Geographical location of *M. dubia* provenances (P₁-P₂ in Satpura mountain ranges and P₃ -P₈ in the northernmost Western Ghats or Sahyadri mountain ranges) in Gujarat, India, selected in the study

Provenance (P)	Geographical attributes			Average biometric attributes		
	Altitude (m)	Longitude (E)	Latitude (N)	Tree height (m)	DBH (cm)	Canopy area (m ²)
Sagai-1 (P1)	373.68	73°47'33.13"	21°40'19.28"	25.80	155.73	246.00
Sagai-2 (P2)	320.04	73°44'21.30"	21°40'47.32"	35.90	172.00	256.16
Nanapondha (P3)	63.09	73° 8'54.14"	20°26'5.45"	26.60	128.20	206.20
Kaprada (P4)	382.52	73°15'23.07"	20°20'15.71"	25.22	120.18	241.40
Waghai (P5)	127.41	73°29'4.63"	20°45'49.32"	33.76	156.99	221.20
Chichinagavtha (P6)	256.03	73°33'40.46"	20°47'19.06"	30.64	148.40	188.50
Mahal (P7)	262.13	73°36'6.52"	20°57'0.13"	42.52	149.00	157.40
Ahwa (P8)	319.13	73°39'24.02"	20°46'1.27"	36.60	117.20	126.60

11.8 to 20.8%, 6.9 to 9.7% and 9.3 to 12.3%, respectively, among provenances (Malek et al., 2024a, 2024b). Further, among the selected 8 provenances total alkaloid content (TAC), total flavonoids content (TFC), total saponin content (TSC), total tannin content (TTC), total terpenoids content (TTeC), total phenolic content (TPC) and total saponin content (TSC) ranged from 12.73 to 13.41, 2.27 to 2.78, 12.51 to 16.91, 12.34 to 12.64, 9.53 to 16.03, 7.13 to 7.76, and 12.34 to 12.64%, respectively. TSC content did not exhibit significant variation among provenances. Leaf fodder TAC, TFC, TSC, TTC and TTeC (14.57, 3.25, 13.01, 15.64 and 8.04%, respectively) were significantly higher in the winter season and TPC was significantly higher (19.44%) in the summer season (Malek et al., 2025). The dried samples were then finely ground (particle size <1 mm) using a mixture grinder and laboratory-grade pulverizer. Leaf fodder dry matter, and organic matter digestibility, gas production (all *in-vitro*) and metabolizable energy were estimated for winter and summer seasons of 2020 and 2021 (average of each season of two years) to ascertain the provenance and seasonal variations.

***In-vitro* digestibility:** *In-vitro* Dry Matter Digestibility was estimated as per the method described by Tilley and Terry (1963). A finely ground sample of feed was incubated for 48 hrs at 39° C with buffered rumen liquor (1:4) in flasks under anaerobic conditions. After 48 hrs of incubation added 2 ml 6N HCL and 0.1 g pepsin powder and further incubated for another 24 hrs. After complete incubation filtered content, dry the residue at 100° C overnight. The general formula for calculating DM digestibility is as follows-

In-vitro DM digestibility (%): (DM disappearance / Wt. of sample on DM base)*100

***In-vitro* gas production (IVGP %):** *In-vitro* gas production was estimated as per the method developed by Menke et al. (1979). To measure the *in-vitro* gas production, rumen liquor was collected from male Surti Goats at early morning before feeding and watering through a mouth tube. Strained rumen liquor diluted

with buffer at the ratio of 1: 2 (v/v). Four replicates of 200 mg DM of *M. dubia*. Leaves from all seasons and provenance were incubated in anaerobic conditions at 39°C with 30 mL rumen liquor fluid medium in 100 mL glass syringes. Gas production was recorded at 3, 6, 9, 12, 24, 48, 72 and 92 hours of incubation from scales on syringes. The formula for calculating gas production and metabolic energy is as follows:

Gas production (mL/200 mg DM) = (Reading after 34 hours of incubation-Initial reading-blank)/feed (mg)
Metabolic energy (KJ) = 1.24 + 0.146* gas (mL/200 mg DM) + 0.0057 *CP + 0.0029 *EE2

Metabolizable energy [ME (KJ)]: The following common equation was used to estimate metabolizable energy (ME) (KJ and KC):

ME(KJ) = 2.20 + 0.136 × G + 0.0057 × CP + 0.0029 × EE. Where, G = gas (m/200 mg DM), CP=crude protein (g/kg DM) and EE=ether extract (g/kg DM), hence ME(KC) = ME(KJ)/4.184

Statistical analysis: The data pertaining to leaf fodder proximate and cell wall composition were statistically analyzed following factorial completely randomized design (CRD) and F-test was done and ANOVA was constructed with following model $Y_{ijk} = \mu + \alpha_i + \beta_j + (\alpha\beta)_{ij} + \varepsilon_{ijk}$ where Y_{ijk} = the response from the k th experimental unit receiving the i th level of factor A and the j th level of factor B; μ = overall mean; α_i = an effect due to the i th level of factor A; β_j = an effect due to the j th level of factor B; $(\alpha\beta)_{ij}$ = an interaction effect of the i th level of factor A with j th level of factor B; ε_{ijk} = a random error have a normal distribution (0, σ^2). Further, treatment means were compared at $p < 0.05$. Further, Duncan's multiple range test (DMRT) was adopted to compare the sets of means of each factor (Sheoran et al., 1998; software OPSTATE).

Results and Discussion

***In-vitro* digestibility:** The results of our study deduced that *M. dubia* leaf fodder *in-vitro* dry matter digestibility (IVDMD) and organic matter digestibility (IVOMD) showed significant ($p \leq 0.05$) deviation among the different provenances and seasons (Fig 2; Table 2). Leaf fodder of Ahwa provenance (P_8) showed significantly higher (64.97%) IVDMD and the lowest (47.74%) was recorded in Waghai (P_5) provenance fodder. Significantly higher (89.88%) IVOMD was for P_4 (Kaprada) provenance and lowest (86.97%) for Sagai-2 (P_2) provenance fodder (Fig 2).

Further, IVDMD and IVOMD were significantly higher (63.47 and 90.13%, respectively) for winter season fodder (S_1) (Table 2). Results also elucidated that the interaction effect of season × provenance for *in-vitro* digestibility parameters (Table 2) was statistically significant ($p \leq 0.05$).

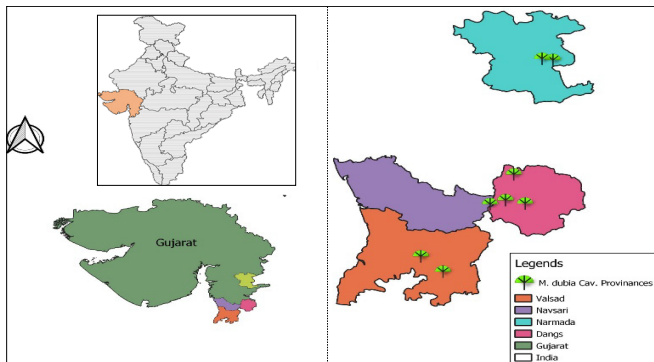


Fig 1. Geographic location of *M. dubia* provenances selected in the study

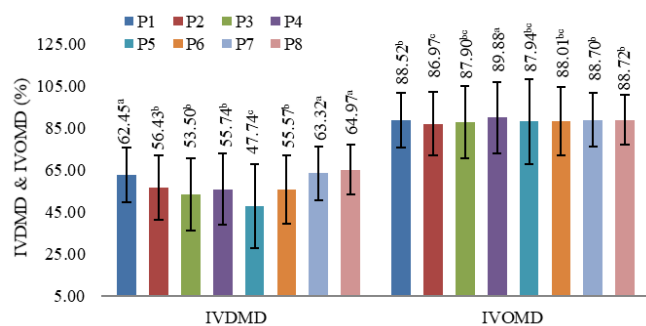


Fig 2: Variation in *M. dubia* leaf fodder *in vitro* dry matter digestibility (IVDMD; SEM=±0.77,) and *in vitro* organic matter digestibility (IVOMD; SEM=±0.21) among different provenances (P₁-Sagai-1, P₂-Sagai-2, P₃-Nana Pondha, P₄-Kaprada, P₅-Waghahi, P₆-Chichinagawtha, P₇-Mahal, P₈-Ahwa), different superscript letters with treatment means in each bar are significantly different according to Duncan’s multiple range test ($P \leq 0.05$)

Table 2. Seasonal variation in *M. dubia* leaf fodder *in-vitro* dry matter digestibility (IVDMD), *in-vitro* organic matter digestibility (IVOMD) in different provenances

Provenance	IVDMD (%)		IVOMD (%)	
	Season			
	S ₁	S ₂	S ₁	S ₂
Sagai-1 (P ₁)	63.58 ^b	61.32 ^{bc}	91.30 ^a	85.73 ^f
Sagai-2 (P ₂)	63.77 ^b	49.08 ^h	90.28 ^b	83.64 ^g
Nana Pondha (P ₃)	63.23 ^b	43.77 ⁱ	88.69 ^d	87.10 ^e
Kaprada (P ₄)	58.96 ^{cd}	52.51 ^{fg}	90.66 ^{ab}	89.10 ^{cd}
Waghahi (P ₅)	42.68 ⁱ	52.80 ^f	90.28 ^b	85.59 ^f
Chichinagavtha (P ₆)	55.99 ^{de}	55.15 ^{ef}	89.88 ^{bc}	86.14 ^f
Mahal (P ₇)	79.22 ^a	47.41 ^h	89.67 ^{bc}	87.73 ^e
Ahwa (P ₈)	80.31 ^a	49.64 ^{gh}	90.30 ^b	87.14 ^e
Mean	63.47 ^a	51.46 ^b	90.13 ^a	86.52 ^b
SEM(±)S	0.36		0.12	
SEM(±) S×P	1.03		0.35	

Winter season fodder of Ahwa provenance (P₈) showed the highest (80.31%) IVDMD, whereas the lowest (42.68%) was recorded in Waghahi provenance (P₅) during the same season. Maximum (91.30%) IVOMD was observed for P₁ provenance (Sagai-1) during the winter season, and was lowest (83.64%) for P₂ provenance (Sagai-2) in summer (Table 2).

Nutrient digestibility value is one of the determinants of forage quality. The higher the digestibility value of forage nutrients, the higher the nutrients used by the body to meet its nutritional needs (Suharlina *et al.*, 2016). In our study, IVDMD of *M. dubia* fodder varied from 43.77 to 80.31%. Rahim *et al.* (2011) recorded 62.60% IVDMD of *M. azedarach* fodder, a sister species from the same family

and comparatively, IVDMD of *M. dubia* fodder during the winter season was higher.

Kcal: Killo calories, S₁: Winter, S₂: Summer, SEM(±): Standard error, S: Season, P: Provenance (P₁-Sagai-1, P₂-Sagai-2, P₃-Nana Pondha, P₄-Kaprada, P₅-Waghahi, P₆-Chichinagawtha, P₇-Mahal, P₈-Ahwa); Different superscript letters with treatment means in same column and row are significantly different according to Duncan’s multiple range test ($p \leq 0.05$)

Commonly fed tree fodders have been reported to have 41.59 to 88.12% IVDMD (Datt *et al.*, 2008; Devasena and Adilaxmamma, 2016; Ramachandran *et al.*, 2014; Giridhar *et al.*, 2018) and *M. dubia* leaf fodder digestibility is in congruence with these studies. Further, in the present study, winter season fodder showed higher IVDMD. A similar trend has been reported by Damame *et al.* (2016) and Khuliso *et al.* (2022) in other common fodder tree species. IVDMD percentage, during winter, reported in the present study falls within the range reported by Elseed *et al* (2002) in many other species. Further, in the present investigation, *M. dubia* leaf fodder IVOMD ranged from 83.64 to 91.30%. IVOMD of common top feed sources has been reported 39.36 to 77.40% (Datt *et al.*, 2008; Kamalak *et al.*, 2010; Devasena and Adilaxmamma 2016). Hence, IVOMD of *M. dubia* fodder is higher compared to many other fodder tree species (Parmar *et al.*, 2022).

***In-vitro* gas production (ml):** The results pertaining to *in-vitro* gas production (IVGP) of *M. dubia* leaf fodder from different provenances and seasons, at 03, 06, 09, 12, 24, 48, 72, 92 hours, revealed significant ($p \leq 0.05$) variation across selected provenances (Fig. 3a-h) and seasons (Table 3). Significantly highest IVGP, at 3, 6 and 9 hours (9.73, 15.71 and 17.53 mL, respectively), was of leaf from Mahal (P₇) provenance (Fig 3a, b, c). The IVGP after 12 hours was maximum (24.25 mL) from leaf fodder of Sagai-1 provenance and lowest (17.66 mL) was for Kaprada provenance leaf fodder (Fig. 3d). The peak IVGP, in all the provenances, was recorded after 24 hours compared to other time intervals (3–92 hours) (Fig. 3e). Significantly higher (38.95 mL) IVGP, at 24 hours, was for Chichinagavtha (P₆) provenance leaf fodder and lowest (24.37 mL) at same time interval was P₈ (Ahwa) provenance. From 48 to 92 hours, IVGP decreased gradually for all provenance fodder samples (Fig 3f-h).

There was significant seasonal variation in IVGP among selected provenances (Table 3). The summer (S₂) season IVGP was significantly ($p \leq 0.05$) maximum (15.29, 16.32, 23.78, 35.69 and 28.45 ml, at 6, 9, 12, 24 and 48 hours, respectively), and lowest (10.80, 13.13, 18.30 28.61 and 24.81 ml, respectively) was for winter season (S₁) leaf fodder at same time intervals. The IVGP increased up to 24 hours (35.69 and 28.61 mL for summer (S₂) and winter (S₁) season, respectively), after which decreased steadily till 92 hours (Fig. 4). The interaction (Table 3) between season

Table 3. Seasonal variation in *M. dubia* leaf fodder *in-vitro* gas production (IVGP; mL) after 3, 6, 9, 12, 24, 48, 72 and 92 hours in different provenances

Provenance	Time interval (Hours)							
	3		6		9		12	
	Season		Season		Season		Season	
	S ₁	S ₂	S ₁	S ₂	S ₁	S ₂	S ₁	S ₂
P ₁	8.52 ^{bc}	7.94 ^{bcd}	12.15 ^{de}	17.60 ^a	14.42 ^{cde}	18.17 ^a	18.61 ^{def}	29.89 ^a
P ₂	4.92 ^{fg}	8.00 ^{bcd}	6.46 ^h	15.95 ^{ab}	10.13 ^g	18.90 ^a	13.39 ^g	24.34 ^b
P ₃	4.58 ^{fg}	6.56 ^{cdef}	9.90 ^{fg}	15.60 ^{ab}	14.15 ^{cde}	15.80 ^{bc}	22.64 ^{bc}	21.40 ^{bcd}
P ₄	5.32 ^{fg}	6.62 ^{cdef}	8.57 ^{gh}	16.73 ^a	9.32 ^g	12.46 ^{ef}	13.13 ^g	22.18 ^{bc}
P ₅	7.76 ^{bcd}	5.55 ^{efg}	12.69 ^{cde}	14.54 ^{bc}	14.41 ^{cde}	17.23 ^{ab}	18.20 ^{ef}	23.17 ^{bc}
P ₆	9.53 ^b	5.85 ^{defg}	14.09 ^{bcd}	13.26 ^{cde}	15.83 ^{bc}	13.46 ^{de}	22.51 ^{bc}	23.40 ^{bc}
P ₇	12.88 ^a	6.59 ^{cdef}	14.39 ^{bc}	17.03 ^a	15.70 ^{bcd}	19.36 ^a	21.00 ^{cde}	23.04 ^{bc}
P ₈	4.08 ^g	4.72 ^{fg}	8.18 ^{gh}	11.60 ^{ef}	11.09 ^{fg}	15.14 ^{bcd}	16.92 ^f	22.81 ^{bc}
S mean	7.20 ^a	6.48 ^b	10.80 ^b	15.29 ^a	13.13 ^b	16.32 ^a	18.30 ^b	23.78 ^a
SEM (±) S	0.26		0.22		0.24		0.34	
SEM (±) S×P	0.72		0.63		0.69		0.97	
Provenance	Time interval (Hours)							
	24		48		72		92	
	Season		Season		Season		Season	
	S ₁	S ₂	S ₁	S ₂	S ₁	S ₂	S ₁	S ₂
P ₁	30.78 ^d	42.77 ^a	26.62 ^{de}	32.53 ^a	26.63 ^c	29.12 ^{ab}	29.47 ^b	31.95 ^a
P ₂	19.36 ^f	37.78 ^b	16.49 ^g	33.10 ^a	17.44 ^h	27.83 ^{bc}	21.26 ^{def}	28.99 ^b
P ₃	29.61 ^d	36.62 ^b	27.86 ^{cd}	28.73 ^{cd}	27.33 ^{bc}	24.10 ^{ef}	29.08 ^b	25.83 ^c
P ₄	25.70 ^e	30.76 ^d	20.19 ^f	25.11 ^e	18.47 ^{gh}	20.24 ^g	21.53 ^{def}	22.96 ^{de}
P ₅	33.32 ^c	37.89 ^b	31.80 ^{ab}	29.66 ^{bc}	30.69 ^a	26.22 ^{cd}	31.99 ^a	28.72 ^b
P ₆	36.76 ^b	41.15 ^a	33.33 ^a	29.64 ^{bc}	30.44 ^a	24.57 ^{de}	31.78 ^a	26.71 ^c
P ₇	33.87 ^c	29.24 ^d	28.16 ^{cd}	21.69 ^f	26.85 ^c	18.39 ^{gh}	29.14 ^b	21.10 ^{ef}
P ₈	19.45 ^f	29.29 ^d	14.00 ^g	27.14 ^{de}	14.41 ⁱ	22.42 ^f	19.65 ^f	23.40 ^d
S mean	28.61 ^b	35.69 ^a	24.81 ^b	28.45 ^a	24.03 ^b	24.11 ^a	26.74 ^a	26.21 ^b
SEM (±) S	0.30		0.27		0.24		0.25	
SEM (±) S×P	0.83		0.78		0.68		0.71	

S₁: Winter, S₂: Summer, SEM(±): Standard error, S: Season, P: Provenance (P₁-Sagai-1, P₂-Sagai-2, P₃-Nana Pondha, P₄-Kaprada, P₅-Waghai, P₆-Chichinagawtha, P₇-Mahal, P₈-Ahwa); Different superscript letters with treatment means in same column and row are significantly different according to Duncan's multiple range test ($p \leq 0.05$)

(S) × provenance (P) for IVGP at different time interval was statistically significant ($p \leq 0.05$) at 24 hours' time interval, maximum (42.77 ml) IVGP for summer season fodder from Sagai-1 (P₁) provenance (Table 3).

The total gas production value describes the effectiveness of the fermentation process. The estimation of gas production can be known based on the fermentation balance or feed characteristics (Johnson and Johnson, 1995). *In-vitro* fermentation techniques for gas production help evaluate and predict feed quality (Kurniawati, 2007).

It is crucial to choose a vegetation source for animal feed that is eco-friendly and has good nutrient content for animals. The present study found that IVGP at 24 hours was between 19.36 and 42.77 mL, which is in congruence with the findings of Girdhar *et al.* (2018) in this species (42.47 mL IVGP) and other species. In general, the higher the OM degradation more the gas production (Sahoo *et al.*, 2010). In the present study, initially IVGP was slow and it increased with time and peaked at 24 hours. The present findings are in congruity with Khan *et al.* (2008).

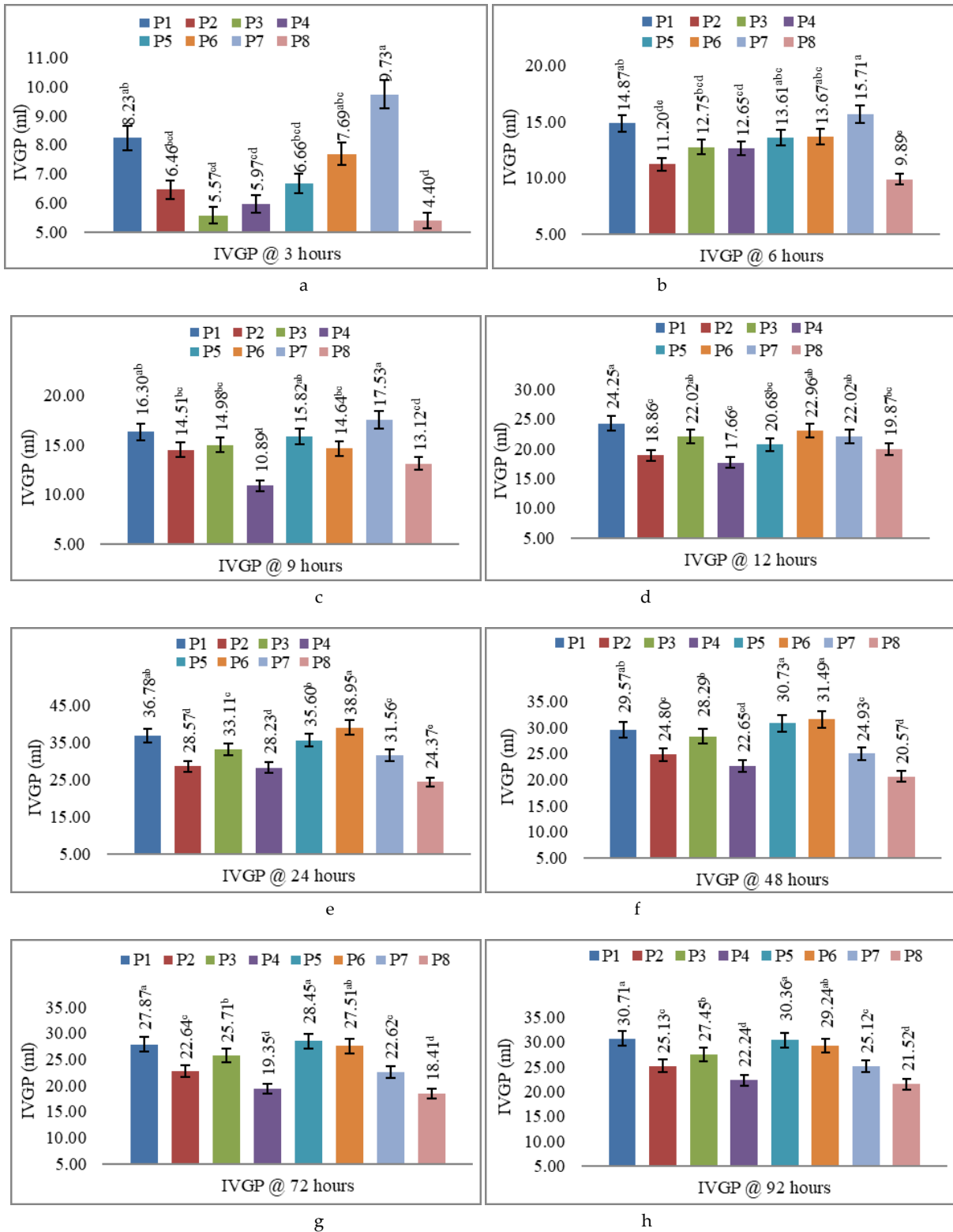


Fig. 3. Variation in *M. dubia* leaf fodder *in-vitro* gas production (IVGP ml) after a) 3, b) 6, c) 9, d) 12, e) 24, f) 48, g) 72 and h) 92 hours (with respective $SEM_{\pm} = 0.48, 0.53, 0.53, 0.71, 0.52, 0.56, 0.49$, and 0.49) among different provenances (P₁-Sagai-1, P₂-Sagai-2, P₃-Nana Pondha, P₄-Kaprada, P₅-Waghai, P₆-Chichinagawtha, P₇-Mahal, P₈-Ahwa), different superscript letters with treatment means in each bar are significantly different according to Duncan's multiple range test ($P \leq 0.05$)

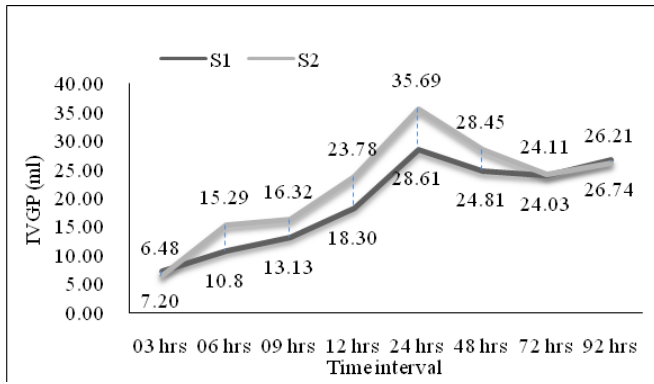


Fig 4. *M. dubia* leaf fodder in seasonal mean *in vitro* gas production (IVGP; ml) showing a peak at 24 hours

Gas production depends on the rumen fermentation and feed substrate degradability (Wahyono et al., 2019). However, all the tree leaves had an excellent digestibility range (above 75%) along with the high gas production potential, indicating their higher fermentative capacity as ruminant feeds (Parmar et al., 2022).

The higher asymptotic gas production found in the Mahal (P₇) provenance suggests a relatively better degradation of cell wall solubles and other degradable carbohydrates. Lag time has been reported to be influenced by the microbial capacity, such as the rate of hydration of feedstuff, microbial attachment to feed particles and nutrient limitations. It may further suggest that there was less limitation to microbial access and attachment, degradation and fermentation of leaves from the Mahal (P₇) provenance. The efficiency by which an animal utilizes feed nutrients has a significant impact on its

productive performance and waste production. The *in-vitro* gas production system helps to better quantify nutrient utilization and its accuracy in describing digestibility in animals. *In-vitro* gas production systems are regularly utilized to screen feed ingredients for inclusion in ruminant diets. The differences recorded in observations on *in-vitro* kinetics and substrate degradability parameters among the treatments are a reflection of the intrinsic chemical composition and anatomical structure of the fodder stuffs (Ansah et al., 2021).

Metabolizable energy (ME; kcal): The results indicated that the metabolizable energy (@24 hours) of *M. dubia* fodder varied significantly ($p \leq 0.05$) across different provenances and seasons. The highest ME (1.83 kcal) was recorded in leaf fodder of Chichinagawtha (P₆) provenance and the lowest (1.35 kcal) in fodder of Ahwa (P₈) provenance (Fig 5a).

The summer (S₂) season leaf fodder recorded significantly higher ME (1.73 kcal), whereas the lowest (1.49 kcal) was for winter (S₁) season fodder (Fig. 5b). The data further evinced that there was significant interaction between season and provenance (S×P) for ME of *M. dubia* fodder. Interaction between summer season fodder and Sagai-1 (P₁) provenance showed maximum ME (1.96 kcal); however, minimum (1.18 kcal) was due to interaction between winter season and Sagai-2 (P₂) provenance (Fig 5b).

ME of *M. dubia* fodder in our study ranged between 1.18 and 1.93 kcal. ME of common fodder trees has been reported in the range of 0.77 and 2.8 kcal (Gupta et al., 2016; Giridhar et al., 2018; Khuliso et al., 2022); hence, ME of *M. dubia* fodder is consistent with these reports.

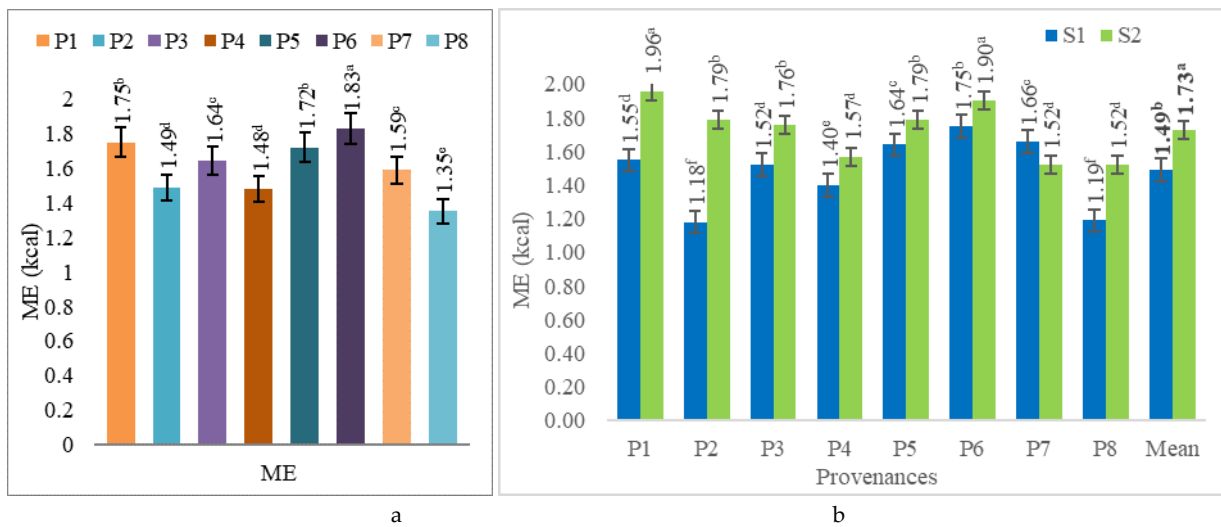


Fig. 5. Variation in *M. dubia* leaf fodder metabolizable energy (@24 hrs) a) among different provenances (SEm=±0.02) and b) seasons (SEm= ± 0.01 and 0.03 for season and S×P, respectively) (ME-metabolizable energy, P-provenance (P₁-Sagai-1, P₂-Sagai-2, P₃-Nana Pondha, P₄-Kaprada, P₅-Waghahi, P₆-Chichinagawtha, P₇-Mahal, P₈-Ahwa), different superscript letters with treatment means in each bar are significantly different according to Duncan’s multiple range test ($P \leq 0.05$)

Furthermore, in the present study, ME of summer season fodder of *M. dubia* was higher compared to winter. Similar findings have been reported by Khuliso *et al.* (2022). Location and season have a significant effect on the ME of plants (Mir *et al.*, 2018). Gas-measuring techniques have been routine in feed evaluation since the early 1980s, when a high correlation was found between metabolizable energy (ME) measured in live animals and that predicted from gas production.

Gas measurement is a direct measure of microbial activity and can be a better index of forage ME content than an indirect *in-vitro* measure based on nutrients fermented. The volume of gas produced in 24 hours from incubating 200 milligrams (mg) of feed, together with the concentration of crude protein and crude fat, is used to predict ME.

Conclusion

Study revealed significant variation in *Melia dubia* leaf fodder *in-vitro* dry matter digestibility (IVDMD), *in-vitro* organic matter digestibility (IVOMD), *in-vitro* gas production (IVGP) and metabolizable energy (ME@24 hours) among selected provenances. Leaf fodder of Ahwa (P8) provenance showed higher IVDMD, and higher IVOMD was of Kaprada (P4) provenance, whereas the highest IVGP, after 24 hours, was for Chichinagawtha (P6) provenance leaf fodder. IVDMD and IVOMD were significantly higher for winter season leaf fodder compared to summer leaf fodder. Conversely, IVGP and ME were higher in the summer season leaf fodder. Results also revealed that IVGP gradually increased up to 24 hours and then decreased till 92 hours. The studied *M. dubia* leaf fodder digestibility attributes are comparable with those of many other family and non-family tree species commonly fed to ruminants and livestock. Thus, the study illustrated that *M. dubia* leaf fodder has good digestibility and can be fed to livestock. The results suggested that provenances with better digestibility attributes could be further selected and used for improvement, which would result in better quality fodder.

Acknowledgment

The authors extend sincere thanks to the Education Department, Gujarat State Government for providing SHODH (Scheme of Developing High-quality research) fellowship to Mr. S. S. Malek for his Doctoral study. Authors are grateful to Deans, College of Forestry, Navsari Agricultural University, Navsari, and College of Veterinary Science & Animal Husbandry (Kamdhenu University, Gandhinagar), Navsari Campus, Gujarat, India for providing necessary support to accomplish this study.

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