

# Investigation of the Nutritional Potential of some Rangeland Plants Species by *in vitro* Gas Production and Fermentation Parameters in Torbat-e Jam, Iran: A Meta-Analysis and Meta-Regression

Meta Analysis

E. Ibrahimi Khoram Abadi<sup>1\*</sup> and M. Kazemi<sup>1</sup><sup>1</sup> Department of Animal Science, Faculty of Agriculture and Animal Science, University of Torbat-e Jam, Torbat-e Jam, Iran

Received on: 10 Mar 2022

Revised on: 1 Jun 2022

Accepted on: 10 Jun 2022

Online Published on: Dec 2023

\*Correspondence E-mail: [el.ebrahimi@tjamcaas.ac.ir](mailto:el.ebrahimi@tjamcaas.ac.ir)

© 2010 Copyright by Islamic Azad University, Rasht Branch, Rasht, Iran

Online version is available on: [www.ijas.ir](http://www.ijas.ir)

## ABSTRACT

The current study aimed to evaluate the nutritional potential of some rangeland plants by *in vitro* gas production kinetics and ruminal fermentation parameters in the Torbat-e Jam region using a meta-analysis approach. A comprehensive literature searches in the scientific databases was carried out to identify studies that investigated the variables of interest. The data analyzed were extracted from 13 peer-reviewed publications. The effect size of all outcomes was reported as a standardized means difference (SMD) with 95% confidence intervals (CI).  $Q$  test and  $I^2$  were calculated to determine the heterogeneity, and a meta-regression was also used to investigate sources of heterogeneity. A significant increase for 24 h *in vitro* gas production (IVGP) was observed ( $P < 0.01$ ). Among the estimated gas parameters, potential GP ( $b_{\text{gas}}$ ) and fractional rate of GP ( $c_{\text{gas}}$ ) were increased significantly ( $P < 0.05$ ). Also, short chain fatty acids (SCFA) ( $P < 0.05$ ), microbial protein yield (MPY) ( $P < 0.01$ ), and ammonia nitrogen ( $\text{NH}_3\text{-N}$ ) ( $P < 0.01$ ) of culture medium were increased. The values of  $I^2$  ( $> 50$ ) and  $Q$  ( $P < 0.05$ ) for 24 h IVGP,  $b_{\text{gas}}$ ,  $c_{\text{gas}}$ , SCFA, MPY and  $\text{NH}_3\text{-N}$  indicated high heterogeneity. No publication bias was found for 24 h IVGP,  $b_{\text{gas}}$ , and SCFA. With the evaluation of funnel plots, only one study on the right of each plot indicates publication bias for  $c_{\text{gas}}$ , MPY, and  $\text{NH}_3\text{-N}$ . A strong regression relation ( $P < 0.01$ ) between some chemical compositions (dry matter, crude protein, neutral detergent fiber, and acid detergent fiber) and MPY and  $\text{NH}_3\text{-N}$  was suggested from meta-regression analysis. In conclusion, the current meta-analysis confirmed that there were differences in fermentation parameters among all rangeland plants. However, all plants species had a relatively high potential nutritive value. According to the reported fermentation parameters, it seems that most of the Torbat-e Jam rangeland plants species have high nutritional value in small ruminant feeding. More *in vivo* and *in vitro* studies are needed to assess the other nutritive values and their effects on animal performance.

**KEY WORDS** gas production, *in vitro*, meta-analysis, rangeland plants.

## INTRODUCTION

Rangeland plants are the main source of forage for ruminants in Iran. In most countries, domestic production commonly depends on pastures (Arzani *et al.* 2017). In Iran, small ruminants feeding and rural economies formed based on the quality and quantity of plants growable in the pas-

tures (Valizadeh *et al.* 2011). The annual production of dry fodder for grazing small ruminants in Iran is estimated at about 10 million tons (Rahbar *et al.* 2008). There were different species of herbs, shrubs, grasses, and trees with suitable biodiversity in Torbat-e Jam mountainous rangelands, and small ruminants in this region often obtain their nutrient requirements through grazing with native pastures

(Kazemi *et al.* 2021). However, rangeland plants with different species contain unique vegetative characteristics and nutritional value. So, optimal use of pastures in ruminant nutrition depends on assessing the quantitative and the qualitative status of forage in rangelands (Arzani *et al.* 2017). The quality and quantity of rangeland plants can be affected by various parameters such as plant species, growth stage, and climate changes (Kazemi and Valizadeh, 2019). Forage quality indicates the amount of nutrients that may be absorbed by the animal as quickly as possible (Holchek *et al.* 2004). The ruminant performance and ruminal fermentation depend on rangeland forage chemical/mineral compounds and nutrient digestibility (Keim *et al.* 2018). Several studies have used the *in vitro* gas production (IVGP) technique to evaluate the nutritional value of some rangeland plants in the Torbat-e Jam region (Kazemi, 2019a; Kazemi and Valizadeh, 2019; Kazemi, 2021b; Kazemi and Ghasemi Bezdi, 2021). Also, dry matter and organic matter digestibility, and the effect of some range plants on ruminal fermentation parameters were determined by other researchers (Kazemi *et al.* 2012; Kazemi, 2019a; Kazemi, 2019b; Kazemi, 2019c; Kazemi, 2020; Kazemi, 2021a; Kazemi, 2021b). Combining the results of these studies allows for more accurate and reliable estimates about the effect of Torbat-e Jam rangeland plants on ruminal fermentation parameters. Meta-analysis is a statistical tool to combine the results of different studies and compile them statistically (Sutton and Higgins, 2008).

Obtaining new information about rangeland plants' fermentation conditions via Meta-analysis helps in the precise and better application of rangeland plants, ensuring that ruminants consume a balanced diet to meet their requirements. Thus, the purpose of the current study was to use a meta-analysis approach to investigate the nutritional value of some rangeland plants via IVGP and ruminal fermentation parameters in the Torbat-e Jam region, and to determine the heterogeneity of observed responses by using meta-regression.

## MATERIALS AND METHODS

### Literature search and study selection

A comprehensive literature searches in the scientific databases of Google Scholar, Web of Science, Scopus, and PubMed was carried out to identify studies that investigated the nutritional value of rangeland plants via IVGP and ruminal fermentation parameters. The keywords used to search relevant studies included rangeland, plant, gas production, rumen, fermentation, ruminants, *in vitro*, Iran, and Torbat-e Jam. A total of 74 scientific publications published between 2010 and 2021 were selected. The selected studies had to meet the following criteria: (1) plant samples should be randomly collected from natural rangeland of Torbat-

Jam; (2) data should be reported on IVGP and ruminal fermentation parameters; (3) the means, the number of repetitions, p-value, standard error of the mean, or the standard deviation of the mean for the measured variables should be reported, and (4) articles were discarded if they had been published as simulation studies, reviews, and also articles that did not measure the variables of interest. Based on the selection criteria, only 13 articles were included in the database for the final analysis (Krogstad *et al.* 2021; Orzuna-orzuna *et al.* 2021).

### Data extraction

The response variables extracted for the meta-analysis were included potential gas production ( $b_{\text{gas}}$ ), the fractional rate of GP ( $c_{\text{gas}}$ ), cumulative GP after 12, 24, 48, and 72 h incubation, metabolizable energy (ME), net energy for lactation ( $NE_L$ ), short chain fatty acids (SCFA), microbial protein yield (MPY),  $NH_3$ -N concentration and pH. The references of the articles included in the data set are listed in Table 1. Averages, standard deviation (SD), number of repetitions, and P-values of each treatment were derived from these articles. The SD value of each experimental group as presented in each article, were used directly in the meta-analysis. In cases where the SD was not reported, it was calculated by multiplying the standard error of the means (SEM) by the square root of the sample size, using the equation  $SD = SEM \times \sqrt{n}$ , where n was number of replicates (Orzuna-orzuna *et al.* 2021). Given the heterogeneity found among the reported parameters, a meta-regression was also used to determine its cause. The chemical compositions of plants dry matter (DM), crude protein (CP), neutral detergent fiber (NDF), and acid detergent fiber (ADF) were inserted in meta-regression analysis for heterogeneity determination.

### Statistical analysis

#### Effect size and Forest plots

Statistical analysis was performed using fixed and random effects models with Comprehensive Meta-Analysis (CMA) software version 3, and the effect size including  $b_{\text{gas}}$ ,  $c_{\text{gas}}$ , IVGP 12, 24, 48, and 72 h, ME,  $NE_L$ , SCFA, MPY,  $NH_3$ -N, and pH was calculated as the standardized means difference (SMD) with a confidence interval (CI) at 95% (Mahdavi *et al.* 2019). The SMD is the difference between treatment and control groups that has been standardized based on the standard deviation of treatment and control groups. The SMD provides the possibility to compare differences caused by different variables (Borenstein *et al.* 2009). The SMD of each production response parameter was the outcome of interest displayed in the forest plot. Information presented in forest plots also provides the means and 95% CI for primary studies.

**Table 1** Summary of papers used for meta-analysis

Author	NC	Plant species	Parameters
		<i>Lucerne</i>	
		<i>Eruca sativa</i>	
Kazemi <i>et al.</i> (2012)	6	<i>Crocus sativus</i>	b <sub>gas</sub> , c <sub>gas</sub> , GP 12, 24, 48, 72, ME, NE <sub>L</sub> , SCFA, MPY
		<i>Cardaria draba</i>	
		<i>Setaria Spp.</i>	
		<i>Triticum aestivum</i>	
Kazemi and Valizadeh (2019)	4	<i>Arctium lappa, Verbascum Thapsus</i>	b <sub>gas</sub> , c <sub>gas</sub> , GP 12, 24, 48, ME, NE <sub>L</sub> , SCFA, NH <sub>3</sub> -N, pH
		<i>Althea officinalis</i>	
		<i>Ferula hermonis</i>	
Kazemi and Valizadeh (2018)	4	<i>Salvia hydrangea</i>	b <sub>gas</sub> , c <sub>gas</sub> , GP 12, 24, 48, ME, NE <sub>L</sub> , SCFA, NH <sub>3</sub> -N, pH
		<i>Sophora alopecur</i>	
Kazemi and Ibrahimi Khoram Abadi (2019), E1	4	<i>Cucumis melo</i>	b <sub>gas</sub> , c <sub>gas</sub> , GP 12, 24, 48, 72, SCFA, MPY, NH <sub>3</sub> -N, pH
Kazemi and Ibrahimi Khoram Abadi (2019), E2	4	<i>Teucrium Polium L.</i>	b <sub>gas</sub> , c <sub>gas</sub> , GP 12, 24, SCFA, MPY, N- NH <sub>3</sub> -N, pH
		<i>Malcolmia Africana</i>	
Kazemi, (2020)	4	<i>Plantago lanceolata</i>	b <sub>gas</sub> , c <sub>gas</sub> , GP 12, 24, 48, 72, ME, NE <sub>L</sub> , SCFA, NH <sub>3</sub> -N, pH
		<i>Phlomis cancellata</i>	
		<i>Klasea latifolia</i>	
		<i>Falcaria vulgaris</i>	
Kazemi (2019a)	4	<i>Malva neglecta</i>	b <sub>gas</sub> , c <sub>gas</sub> , GP 12, 24, 48, 72, ME, NE <sub>L</sub> , SCFA, NH <sub>3</sub> -N, pH
		<i>Chenopodium album</i>	
		<i>Polygonum aviculare</i>	
Kazemi (2019c)	4	<i>Lallemantia royleana</i>	b <sub>gas</sub> , c <sub>gas</sub> , GP 12, 24, 48, 72, MPY
Kazemi (2019b)	4	<i>Artemisia aucheri</i> Bioss	b <sub>gas</sub> , c <sub>gas</sub> , GP 12, 24, 48, 72, SCFA, MPY, NH <sub>3</sub> -N, pH
Kazemi <i>et al.</i> (2019)	8	<i>Crucis sativus L.</i>	b <sub>gas</sub> , c <sub>gas</sub> , GP 12, 24, 48, 72, SCFA, MPY
Kazemi (2021a)	4	<i>Portulaca oleracea L.</i>	b <sub>gas</sub> , c <sub>gas</sub> , GP 12, 24, 48, 72, MPY, SCFA, NH <sub>3</sub> -N, pH
		<i>Marrubium vulgare</i>	
Kazemi (2021b)	5	<i>Ceratocarpus arenarrius L.</i>	b <sub>gas</sub> , c <sub>gas</sub> , GP 12, 24, 48, 72, SCFA, NH <sub>3</sub> -N, pH
		<i>Gypsophila paniculata L.</i>	
		<i>Ferula gummosa</i> Bioss	
		<i>Centaurea virgata</i> Lam	
Kazemi and Ghasemi (2021)	3	<i>Alhagi maurorum</i>	b <sub>gas</sub> , c <sub>gas</sub> , GP 12, 24, 48, 72, ME, NE <sub>L</sub> , SCFA, NH <sub>3</sub> -N, pH

NC: number of comparisons; b<sub>gas</sub>: potential gas production; c<sub>gas</sub>: fractional rate of gas production; GP: gas production; ME: metabolizable energy; NE<sub>L</sub>: net energy for lactation; SCFA: short chain fatty acids; MPY: microbial protein yield and NH<sub>3</sub>-N: ammonia nitrogen.

The weight of each study is calculated from the inverse of the variance of the effect size (Mahdavi *et al.* 2019).

### Heterogeneity

The heterogeneity was determined using the  $Q$  test and the  $I^2$  (percentage of variation) statistic (Higgins and Thompson, 2002). Due to the relatively low power of the  $Q$  test to detect heterogeneity among a small number of treatment comparisons, an alpha level of 0.10 was used (Egger *et al.* 2001; Lean *et al.* 2009).  $I^2$  values ranged from 0 to 100%. Values close to 25% indicate low heterogeneity, close to 50% indicate moderate heterogeneity, and close to 75% indicate high heterogeneity among studies (Higgins and Thompson, 2002; Borenstein *et al.* 2009). Likewise,  $I^2$  values greater than 50% indicate significant heterogeneity (Lean *et al.* 2014).

### Publication bias

Existing publication bias was examined using the funnel plot, a simple scatter plot of the intervention effect estimates from individual studies (horizontal axis) plotted against study precision (vertical axis) (Lean *et al.* 2009). Large studies appear toward the top of the plot and generally cluster around the mean effect size. Smaller studies appear towards the bottom of the plot (Borenstein *et al.* 2009). Similar to meta-regression, funnel plot analysis was only performed for variables that were reported in at least 10 studies.

### Meta-regression

Meta-regression analysis was used to explore other sources of undetected heterogeneity parameters that show an  $I^2$  greater than 50%, and to better understand how the results

are affected by other parameters. The variables for meta-regression were chemical composition of rangeland plants including DM, CP, NDF, and ADF. Meta-regression was estimated through the method of moments (DerSimonian and Laird method). This method of estimating the variance between studies is well established (Borenstein *et al.* 2009; Mahdavi *et al.* 2019).

## RESULTS AND DISCUSSION

### Description of the database

The description of all variables included in the database is listed in Table 2. Mean  $b_{\text{gas}}$  (mL/200 mgDM) was  $49.43 \pm 14.32$ . Our database also included a  $c_{\text{gas}}$  (mL/h/200 mg DM) averaging  $0.093 \pm 0.1$ . The mean value of GP (mL/200 mg DM) after 12, 24, 48 and 72 h of incubation was,  $29.26 \pm 12.67$ ,  $38.92 \pm 13.39$ ,  $45.22 \pm 14.84$  and  $47.54 \pm 15.05$ , respectively. Metabolizable energy (MJ/kg DM),  $NE_L$  (MJ/kg DM), SCFA (mmol), MPY (mg/kg OMD),  $NH_3-N$  (mg/dL) and pH averaged  $7.92 \pm 1.77$ ,  $4.17 \pm 1.29$ ,  $31.17 \pm 15.53$ ,  $79.05 \pm 32.11$ ,  $21.24 \pm 7.50$  and  $6.68 \pm 0.22$ , respectively.

### The *in vitro* gas test parameters

The results of the meta-analysis for the *in vitro* gas test and estimated gas parameters are reported in Tables 3. A significant difference for 24 h IVGP was observed among the studied plants ( $P=0.012$ ) (Table 3) (Figure 1). However, the 12 ( $P=0.591$ ), 48 ( $P=0.805$ ) and 72 h ( $P=0.623$ ) IVGP were not affected by different rangeland plants incubated in the culture medium (Table 3). Among the estimated gas parameters,  $b_{\text{gas}}$  ( $P=0.044$ ) and  $c_{\text{gas}}$  ( $P=0.056$ ) were increased significantly in the culture medium (Table 3) (Figures 2 and 3).

The values of  $I^2$  and  $Q$  for 24 h IVGP,  $b_{\text{gas}}$  and  $c_{\text{gas}}$  effect size indicated high heterogeneity (Table 3). With the examination of funnel plots for publication bias, no bias in the publication for 24 h IVGP, and  $b_{\text{gas}}$  was observed except for  $c_{\text{gas}}$  (Figure 7). The meta-regression finding for heterogeneous variables are shown in Table 5. ADF ( $P=0.002$ ), CP ( $P=0.003$ ) and ADF ( $P=0.001$ ) showed a significant regression for  $b_{\text{gas}}$ ,  $c_{\text{gas}}$  and 24 h IVGP, respectively (Table 5).

All of the *in vitro* gas production parameters including IVGP at different times of incubation,  $b_{\text{gas}}$ , and  $c_{\text{gas}}$  were changed when *Alhagi maurorum* incubated at three growth stages (vegetative, flowering and seeding) (Kazemi and Ghasemi Bezdi, 2021). The GP kinetic was significantly different among the *Falcaria vulgaris*, *Malva neglecta*, *Chenopodium album*, and *Polygonum aviculare* (Kazemi, 2019a). Kazemi (2020) reported a significant difference in IVGP parameters among the incubated plants including *Plantago lanceolata*, *Malcolmia africana*, *Phlomis cancellata*, and *Klasea latifolia*.

In another report, a significant difference in GP kinetic was observed when *Marrubium vulgare*, *Ceratocarpus arenarius*, *Gypsophila paniculate*, *Ferula gummosa* Boiss, and *Centaurea virgate* Lam were incubated in a culture medium (Kazemi, 2021b). The IVGP technique is widely used to evaluate the effect of various rangeland plants and forages on fermentation parameters (Makkar, 2004). This technique was simulated from the ruminal fermentation, and IVGP as a method was used simply to determine volatile fatty acid (VFA) contents as a main energy source of ruminants. When rumen microorganisms digest the feed particles, part of the produced gas is produced directly and another part of the gas is produced as a result of VFA buffering in the culture medium (Menke and Steingass, 1988). In addition, the amount of GP is also affected by external factors such as animal donor of rumen fluid, the temperature of incubation, pH, sufficient buffering capacity, and movement of GP bottles (Getachew *et al.* 1998). A positive correlation between 24 h IVGP and dry matter digestibility (DMD) was reported by Kazemi (2019a), Kazemi (2019b) and Kazemi (2019c). The increase in GP 24 could be due to a strong positive correlation between  $b_{\text{gas}}$  and total volatile fatty acids (TVFA), as was observed an increase in TVFA among the incubated rangeland plants (Kazemi, 2019a; Kazemi, 2019b; Kazemi, 2019c; Kazemi and Valizadeh, 2019).

### Effect of rangeland plants incubation on ruminal fermentation parameters

The meta-analysis findings for the ruminal fermentation parameters are reported in Table 4. SCFA changed when different rangeland plants were incubated in the culture medium ( $P=0.056$ ) (Table 4) (Figure 4). In the *in vitro* testing of rangeland plants, MPY increased significantly ( $P=0.003$ ) (Table 4) (Figure 5). Also, a significant increase in  $NH_3-N$  concentration was observed ( $P=0.019$ ) (Table 4) (Figure 6). In contrast, ME,  $NE_L$ , and pH were not affected by *in vitro* incubation of rangeland plants. We found a significant effect of the heterogeneity for SCFA, MPY, and  $NH_3-N$  concentration with high amounts (Table 4). No publication bias was found for SCFA (Figure 7). With the evaluation of funnel plots, only one study on the right of each plot indicates publication bias for  $c_{\text{gas}}$ , microbial protein yield, and  $NH_3-N$  (Figure 7). The meta-regression finding for heterogeneous variables is reported in Table 5. Chemical compositions including DM, CP, NDF, and ADF are the main parameters affecting MPY and  $NH_3-N$  concentration of culture medium after incubation of rangeland plants (Table 5). MPY and  $NH_3-N$  concentration of culture medium increased with increasing DM and CP contents of rangeland plants. However, the MPY and  $NH_3-N$  concentration decreased when NDF and ADF concentrations of rangeland plants decreased (Table 5).

**Table 2** Descriptive statistics of the complete data set

Parameter	Parameter estimates					
	NC	Mean	SD	Median	MIN	MAX
b <sub>gas</sub> (mL/200 mg DM)	54	49.43	14.32	51.11	21.35	77.98
c <sub>gas</sub> (mL/h/200 mg DM)	54	0.093	.10	0.07	0.03	0.80
GP 12h (mL/200 mg DM)	45	29.26	12.67	28.60	8.10	52.53
GP 24h (mL/200 mg DM)	54	38.92	13.39	39.98	13.56	62.67
GP 48h (mL/200 mg DM)	54	45.22	14.84	47.07	18.41	74.42
GP 72h (mL/200 mg DM)	43	47.54	15.05	46.52	21.19	79.67
ME (MJ/kg DM)	30	7.92	1.77	8.32	4.12	10.70
NEI (MJ/kg DM)	30	4.17	1.29	4	1.89	6.56
SCFA (mmol)	54	31.17	15.53	33.72	0.87	65.72
MPY (mg/kg OMD)	30	79.085	32.11	82.04	6.63	125.06
NH <sub>3</sub> -N (mg/dL)	41	21.24	7.50	16.53	11.80	39.48
pH	41	6.68	0.22	6.69	6.35	7.07

NC: number of comparisons; b<sub>gas</sub>: potential gas production; c<sub>gas</sub>: fractional rate of gas production; GP: gas production; ME: metabolizable energy; NE<sub>L</sub>: net energy for lactation; SCFA: short chain fatty acids; MPY: microbial protein yield; NH<sub>3</sub>-N: ammonia nitrogen and OMD: organic matter digestibility.

**Table 3** Effect size and heterogeneity of the *in vitro* gas production parameters measured for different rangeland plants incubated in a culture medium

Outcome	Model	NC	SMD	95% CI	P-value	Q	P-value	I <sup>2</sup>
b <sub>gas</sub>	Fixed	54	0.436	0.234, 0.639	0.001	38.19	0.001	86.69
	Random	54	0.278	-0.290, 0.846	0.044			
c <sub>gas</sub>	Fixed	54	0.427	0.230, 0.623	0.001	36.35	0.001	85.29
	Random	54	0.427	-0.100, 0.954	0.056			
GP 12 h	Fixed	45	0.160	-0.396, 0.076	0.185	37.38	0.001	88.37
	Random	45	0.194	-0.904, 0.515	0.591			
GP 24 h	Fixed	54	0.069	-0.129, 0.266	0.001	38.11	0.001	86.13
	Random	54	0.050	-0.596, 0.495	0.012			
GP 48 h	Fixed	54	0.081	-0.115, 0.278	0.419	37.56	0.001	85.88
	Random	54	-0.068	-0.859, 0.515	0.805			
GP 72 h	Fixed	43	0.047	-0.006, 0.474	0.691	34.44	0.001	87.98
	Random	43	-0.172	-0.428, 0.751	0.623			

b<sub>gas</sub>: potential gas production; c<sub>gas</sub>: fractional rate of gas production; GP12, 24, 48, and 72 h: the *in vitro* gas production after 12, 24, 48 and 72 h incubation and NC: number of comparisons.

SMD: standardized mean difference.

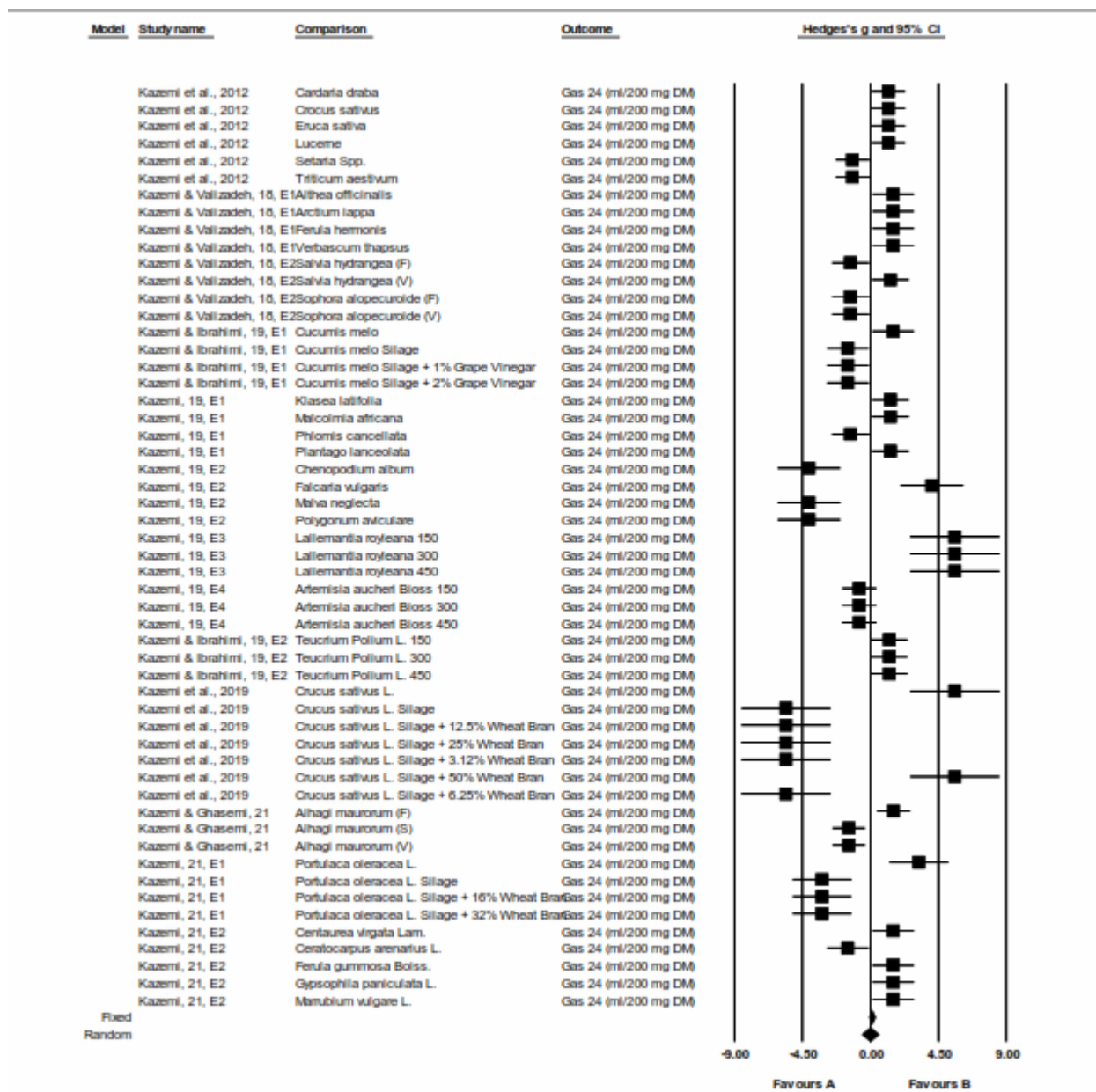
It has been reported that the TVFA and NH<sub>3</sub>-N concentration can be affected by the *in vitro* incubation of rangeland plants (*Falcaria vulgaris*, *Malva neglecta*, *Chenopodium album*, and *Polygonum aviculare*) (Kazemi, 2019a).

Different concentrations of TVFA and NH<sub>3</sub>-N have been reported among some rangeland plants (*Plantago lanceolata*, *Malcolmia africana*, *Phlomis cancellata* and *Klasea latifolia*) (Kazemi, 2020). Similarly, significant differences for TVFA and NH<sub>3</sub>-N concentrations were found among the five rangeland plant species (*Marrubium vulgare* L., *Ceratocarpus arenarius* L., *Gypsophila paniculate* L., *Ferula gummosa* Boiss and *Centaurea virgate* Lam) (Kazemi, 2021b).

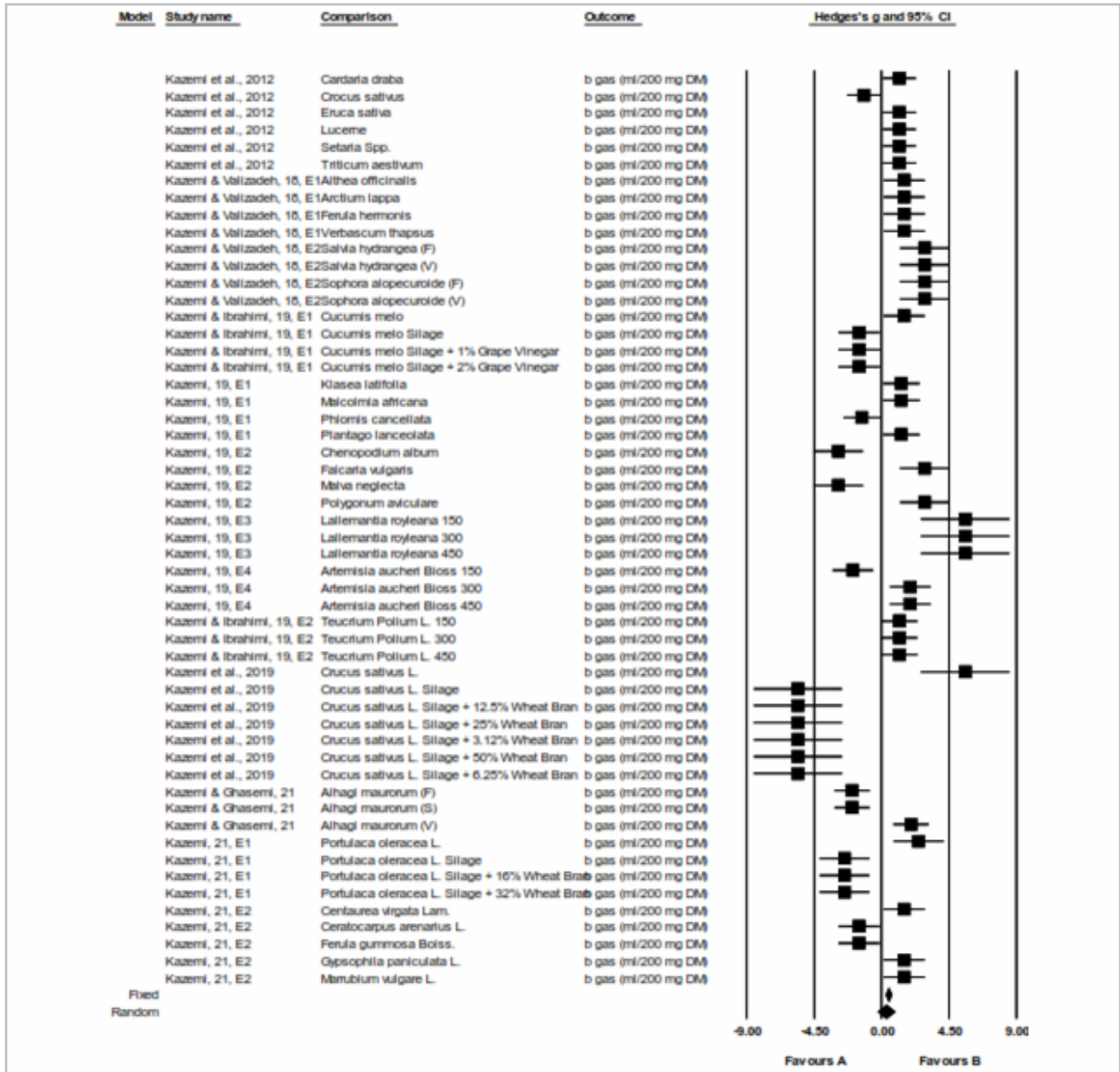
TVFA and MPY were changed when *Lucerne* (*Medicago sativa*), *Eruca sativa*, *Crocus sativus*, *Cardaria draba*, *Setaria* spp., and *Triticum aestivum* were incubated at the early growth stage (Kazemi *et al.* 2012).

There have been several reports of changes in the MPY by *in vitro* testing of different rangeland plants (*Lallemania royleana*, *Artemisia aucheri* Boiss, and *Teucrium polium* L.) (Kazemi, 2019c; Kazemi and Ibrahimi Khoram Abadi, 2019). In contrast, no significant difference between the two growth stages of *Alhagi maurorum* (vegetative and flowering) for ME and NE<sub>L</sub> was reported by Kazemi and Ghasemi Bezdi (2021).

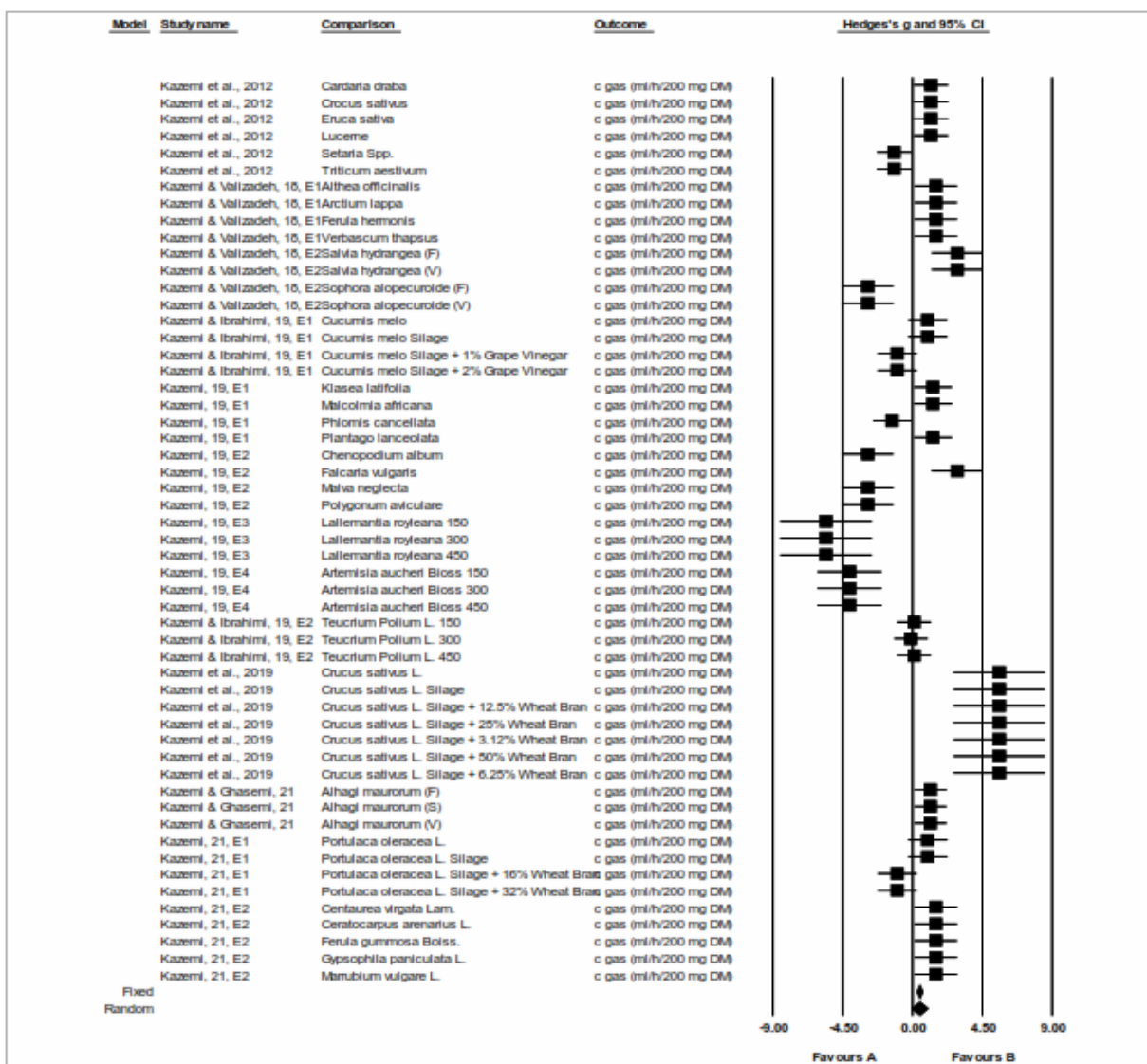
Makkar (2005) observed a positive correlation between IVGP and SCFA production. This finding has been used to estimate the SCFA production from gas values as an indicator of energy available to the animals. SCFA could supply up to 80% of the energy requirement in ruminants (Bergman, 1990). It is reported that higher content of non-fiber carbohydrate (NFC) and higher DM digestibility can lead to more SCFA production (Gilaverte *et al.* 2011; Kazemi and Ghasemi Bezdi, 2021).



**Figure 1** Forest plot of 24 h *in vitro* gas production of different rangeland plants (the size of the squares illustrated the weight of each study relative to the mean effect size, which is indicated by the diamond at the bottom)



**Figure 2** Forest plot of potential gas production ( $b_{gas}$ ) measured for different rangeland plants (the size of the squares illustrated the weight of each study relative to the mean effect size, which is indicated by the diamond at the bottom)



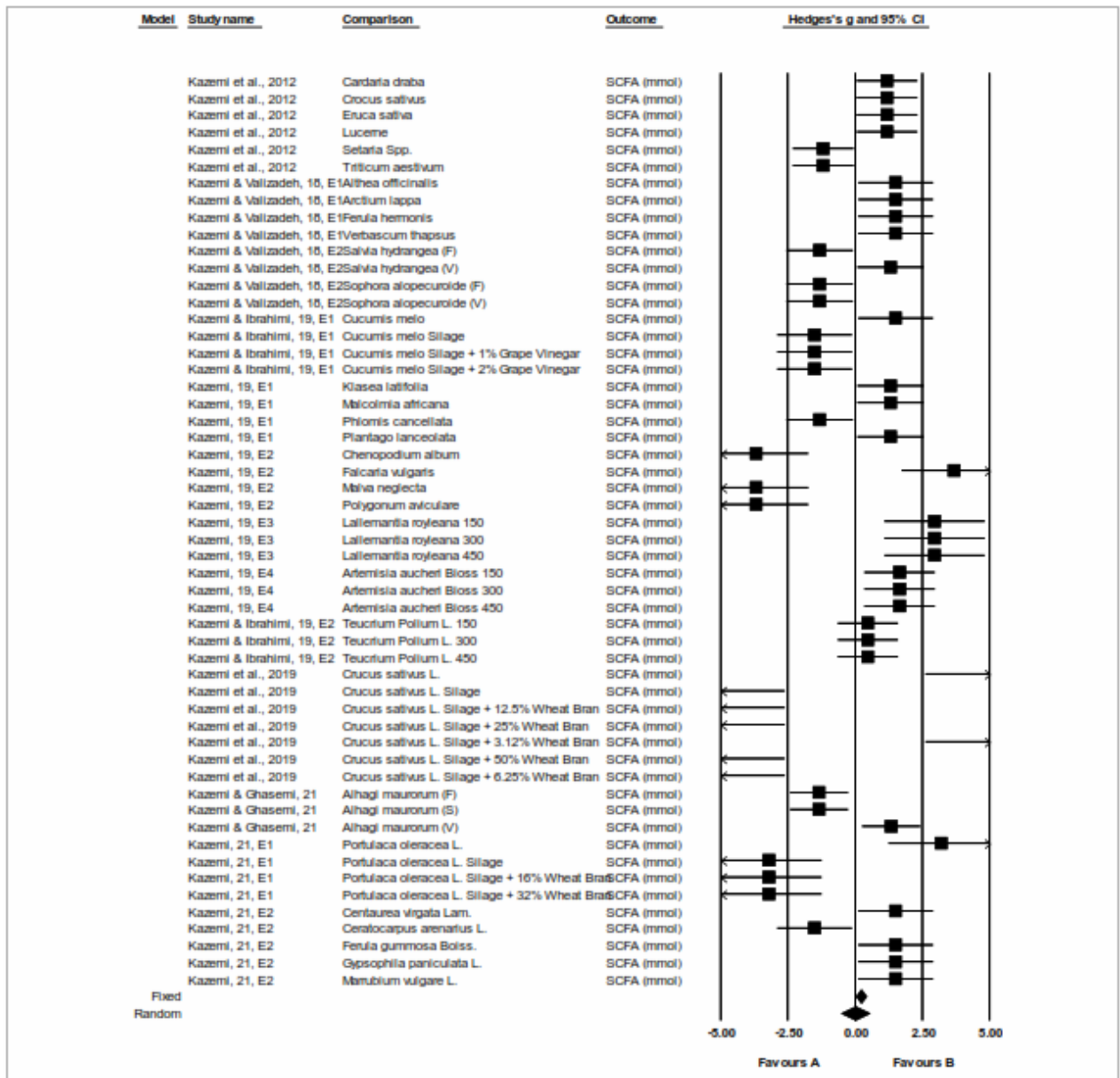
**Figure 3** Forest plot of fractional rate of gas production ( $c_{gas}$ ) measured for different rangeland plants (the size of the squares illustrated the weight of each study relative to the mean effect size, which is indicated by the diamond at the bottom)

**Table 4** Effect size and heterogeneity of some ruminal fermentation parameters measured by an *in vitro* technique for different rangeland plants

Outcome	Model	NC	SMD	95% CI	P-value	Q	P-value	I <sup>2</sup>
ME	Fixed	30	0.234	-0.006, 0.474	0.056	49.18	0.001	83.05
	Random	30	0.162	-0.428, 0.751	0.591			
NEI	Fixed	30	0.234	-0.006, 0.474	0.056	49.18	0.001	83.05
	Random	30	0.162	-0.428, 0.751	0.591			
SCFA	Fixed	54	0.207	-0.012, 0.402	0.018	36.88	0.001	85.43
	Random	54	0.005	-0.529, 0.519	0.056			
MPY	Fixed	30	1.508	0.338, 0.955	0.001	27.48	0.001	86.38
	Random	30	0.050	0.520, 2.494	0.003			
NH <sub>3</sub> -N	Fixed	41	0.282	0.066, 0.498	0.419	23.63	0.001	82.80
	Random	41	0.635	0.105, 1.165	0.019			
pH	Fixed	41	0.099	-0.092, 0.289	1.015	32.12	0.001	55.06
	Random	41	0.090	-0.196, 0.376	0.614			

NC: number of comparisons; ME: metabolizable energy; NE: net energy for lactation; SCFA: short chain fatty acids; MPY: microbial protein yield and NH<sub>3</sub>-N: ammonia nitrogen.  
SMD: standardized mean difference.





**Figure 4** Forest plot of short chain fatty acids (SCFA) measured for different rangeland plants (the size of the squares illustrated the weight of each study relative to the mean effect size, which is indicated by the diamond at the bottom)

However, the different SCFA produced among the different rangeland plants can be attributed to their different chemical compositions, and also their different DM and OM digestibility at different growth stages (Kazemi and Ghasemi Bezdi, 2021). In addition, proximal analysis of rangeland plant species revealed that IVGP were affected by the extent of lignification of NDF and confirmed reducing effects of NDF and ADF on IVGP. Several studies have been conducted to evaluate the effect of NDF and ADF contents of different rangeland plants on IVGP (Khazaal et al. 1994; Tolera et al. 1997; Abdulrazak et al. 2000; Haddi et al. 2003; Kazemi et al. 2012).

All of them have reported a negative effect of NDF and ADF contents on  $c_{gas}$  and IVGP. The negative correlation between NDF and ADF contents of plants with IVGP can be attributed to the microbial activity reducing in the culture medium after their incubation (Kazemi et al. 2012). The results of Bach et al. (2005) indicated that the degradation of true protein of feed can increase  $NH_3-N$  concentration in the rumen.

Most of the ruminal ammonia is absorbed across the rumen wall. However, there is a mechanism in ruminants to recycle 40 to 80% of hepatic Urea-N output to the gastrointestinal tract.

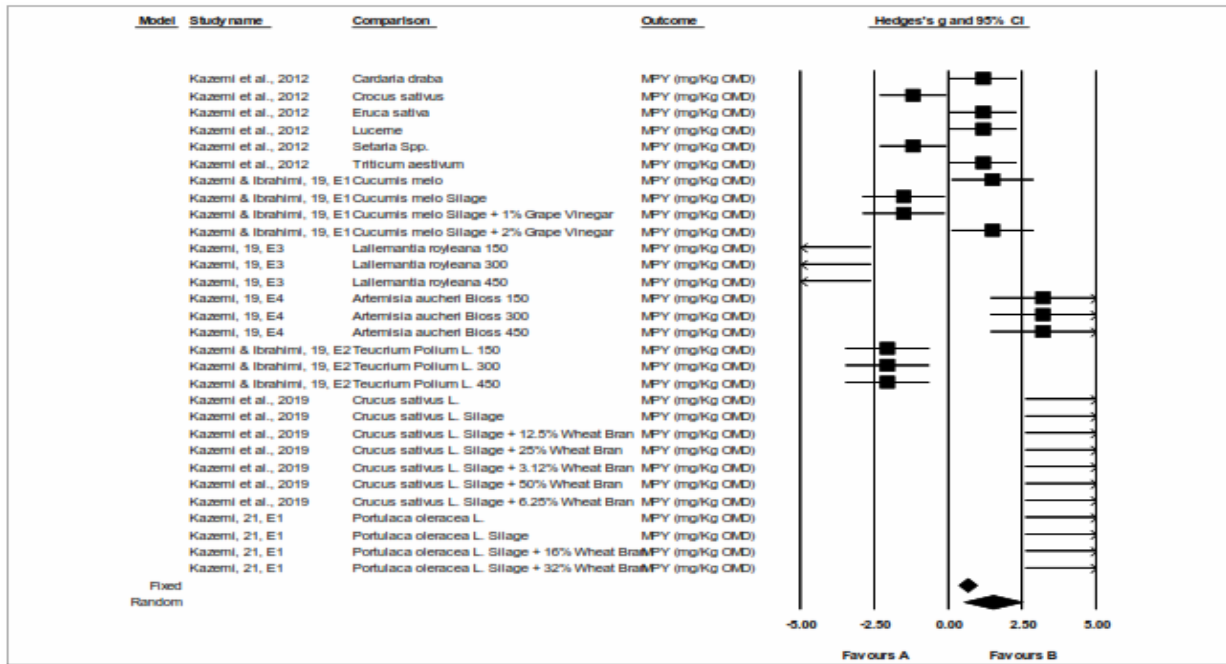


Figure 5 Forest plot of microbial protein yield (MPY) measured for different rangeland plants (the size of the squares illustrated the weight of each study relative to the mean effect size, which is indicated by the diamond at the bottom)

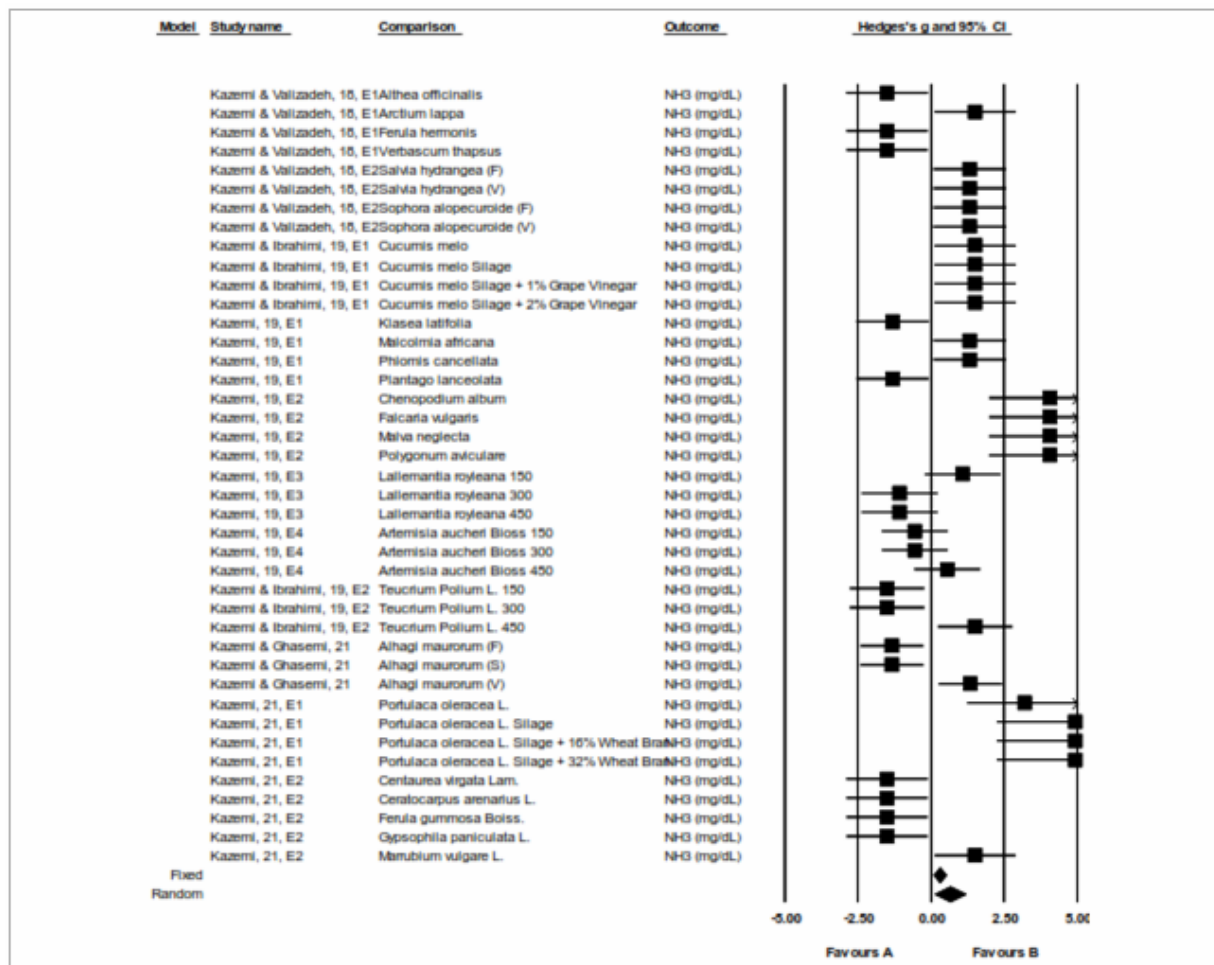
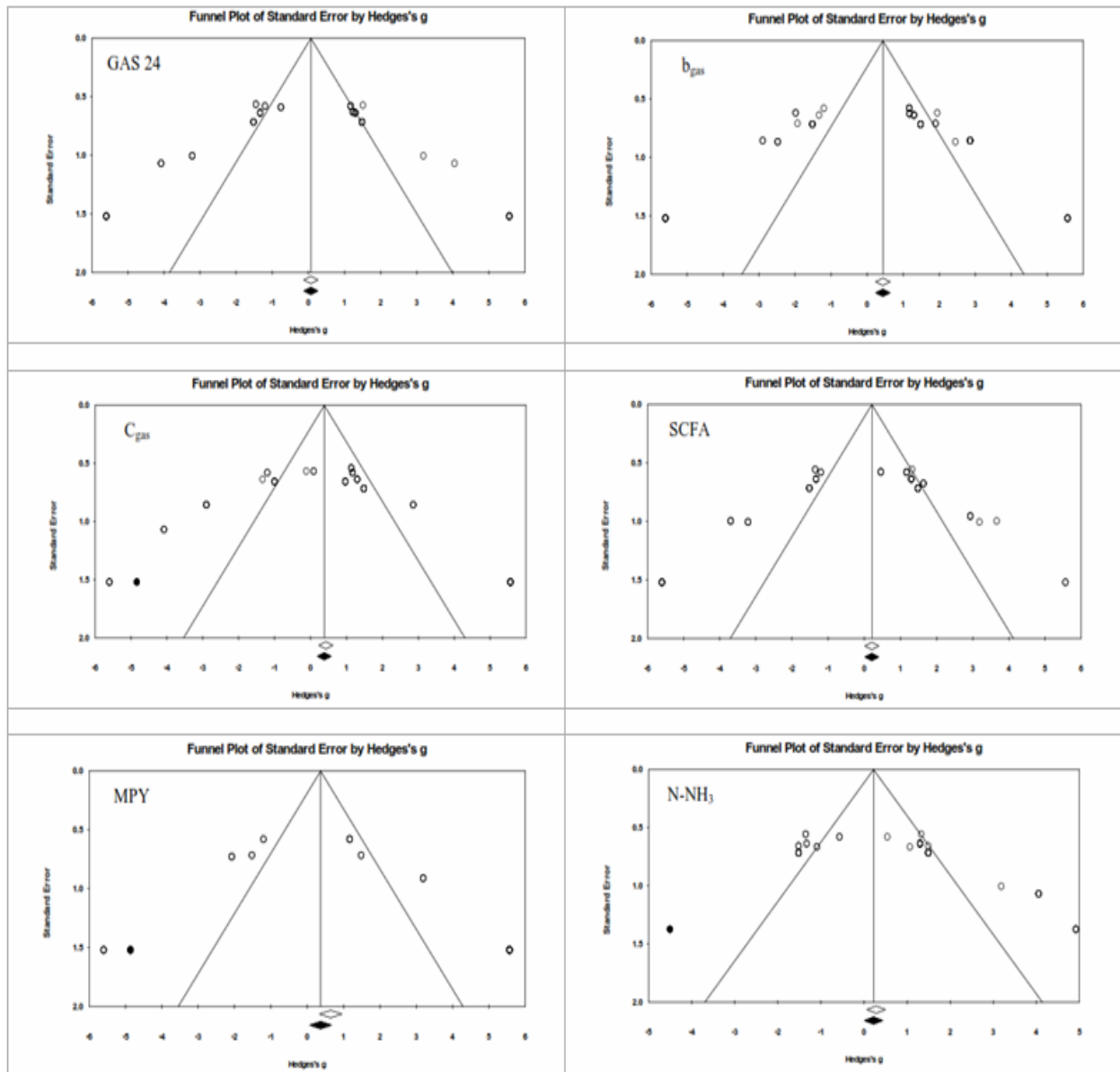


Figure 6 Forest plot of NH<sub>3</sub>-N concentration measured for different rangeland plants (the size of the squares illustrated the weight of each study relative to the mean effect size, which is indicated by the diamond at the bottom)



**Figure 7** Funnel plot for analyzing publication bias of 24 h gas production, potential gas production ( $b_{gas}$ ), fractional rate of gas production ( $c_{gas}$ ), short chain fatty acids (SCFA), microbial protein yield (MPY), and NH<sub>3</sub>-N (empty circles indicate observed values, and full circles possible missing values)

**Table 5** Summary of meta-regression analysis

Outcomes	Covariate	Slope	P-value	Intercept	P-value
$b_{\text{gas}}$	DM	0.01	0.130	0.81	0.003
	CP	0.03	0.090	-0.09	0.770
	NDF	-0.06	0.940	0.45	0.143
	ADF	0.06	0.002	-1.10	0.003
$c_{\text{gas}}$	DM	0.03	0.270	0.34	0.190
	CP	0.05	0.003	1.29	0.004
	NDF	-0.01	0.250	1.07	0.004
	ADF	-0.01	0.260	0.83	0.020
GP 24 h	DM	0.04	0.920	0.09	0.740
	CP	0.02	0.220	0.44	0.750
	NDF	-0.01	0.850	0.11	0.680
	ADF	0.04	0.001	-1.00	0.004
SCFA	DM	0.01	0.083	0.46	0.179
	CP	0.03	0.838	0.26	0.402
	NDF	-0.01	0.422	0.79	0.005
	ADF	-0.01	0.411	0.49	0.174
MPY	DM	0.08	0.001	2.68	0.001
	CP	0.20	0.001	-2.68	0.001
	NDF	-0.08	0.001	3.36	0.001
	ADF	-0.20	0.001	5.30	0.001
$\text{NH}_3\text{-N}$	DM	0.06	0.001	2.03	0.001
	CP	0.11	0.001	-1.45	0.001
	NDF	-0.07	0.001	2.70	0.001
	ADF	-0.06	0.003	1.67	0.003

$b_{\text{gas}}$ : potential gas production;  $c_{\text{gas}}$ : fractional rate of gas production; GP 24 h: gas production after 24 h incubation; SCFA: short chain fatty acids; MPY: microbial protein yield;  $\text{NH}_3\text{-N}$ : ammonia nitrogen; DM: dry matter; CP: crude protein; NDF: neutral detergent fiber and ADF: acid detergent fiber.

The  $\text{NH}_3\text{-N}$  concentration in the culture medium reflects the balance between of degradability of protein sources and  $\text{NH}_3\text{-N}$  consumption for microbial protein synthesis (Dijkstra *et al.* 2005). Also,  $\text{NH}_3\text{-N}$  concentration can be affected by the quantity and type of fermented carbohydrates (Dijkstra *et al.* 2005). The increased  $\text{NH}_3\text{-N}$  of culture medium as a result of the *in vitro* incubation of rangeland plants can be attributed to their high protein content (Kazemi, 2020). Therefore, increasing the fermentation efficiency in the culture medium will be useful to make more nutrients available for microbial protein synthesis (Blümmel *et al.* 1997).

## CONCLUSION

Having knowledge about the ME, NE, chemical composition and other nutritional values of different plants can also help the nutritionists to provide a suitable diet for ruminants. The current meta-analysis confirmed that there were differences in fermentation parameters among all rangeland plants. However, all rangeland plants species had a relatively high potential nutritive value. Despite considerable differences among the studies, the current meta-analysis showed that the fermentation profile altered with the incubation of different rangeland plants in the culture medium. Also, the meta-analysis revealed that the chemical compositions of rangeland plants species including DM, CP, NDF, and ADF were an important source of the variation. According to the reported fermentation parameters, it seems that the rangeland plants of Torbat-e Jam have a high nutritional potential in small ruminants feeding. However, more *in vivo* and *in vitro* studies are needed to assess the other nutritive values and their effects on animal performance.

## ACKNOWLEDGEMENT

The authors would like to thank the University of Torbat-e Jam for financial support of this project.

## REFERENCES

- Abdulrazak S.A., Fujihara T., Ondilek J.K. and Ørskov E.R. (2000). Nutritive evaluation of some Acacia tree leaves from Kenya. *Anim. Feed Sci. Technol.* **85**, 89-98.
- Arzani H., Motamedi H., Aghajanlu H., Rashtvand R. and Zareii A. (2017). Forage quality of important rangeland species in Alamut Qazvin and Badamestan mountain rangelands of Zanjan. *J. Watershed Manag. Res.* **69(4)**, 805-818.
- Bach A., Calsamiglia S. and Stern M.D. (2005). Nitrogen metabolism in the rumen. *J. Dairy Sci.* **88**, 9-21.
- Bergman E.N. (1990). Energy contributions of volatile fatty acids from the gastrointestinal tract in various species. *Physiol. Rev.* **70**, 567-590.
- Blümmel M., Steinga H. and Becker K. (1997). The relationship between *in vitro* gas production, *in vitro* microbial biomass yield and 15N incorporation and its implications for prediction of voluntary feed intake of roughages. *British J. Nutr.* **77(6)**, 911-921.
- Borenstein M., Hedges L., Higgins J.P. and Rothstein H. (2009). Introduction to Meta-Analysis. John Wiley and Sons, Chichester, United Kingdom.
- Dijkstra J., Forbes J.M. and France J. (2005). Quantitative Aspects of Ruminant Digestion and Metabolism. CAB Int., Wallingford. United Kingdom.
- Egger M., Smith G.D. and Altman D.G. (2001). Systematic Reviews in Health Care. MJB Publishing Group, London, United Kingdom.
- Getachew G., Blümmel M., Makkar H.P.S. and Becker K. (1998). *In vitro* gas measuring techniques for assessment of nutritional quality of feeds: A review. *Anim. Feed Sci. Technol.* **72**, 261-281.
- Gilaverte S., Susin I., Pires A.V., Ferreira E.M., Mendes C.Q., Gentil R.S. and Rodrigues G.H. (2011). Diet digestibility, ruminal parameters and performance of Santa Ines sheep fed dried citrus pulp and wet brewer grain. *Rev. Bras. Zootec.* **40**, 639-647.
- Haddi M.L., Filacorda S., Meniai K., Rollin F. and Susmel P. (2003). *In vitro* fermentation kinetics of some halophyte shrubs sampled at three stages of maturity. *Anim. Feed Sci. Technol.* **104**, 215-225.
- Higgins J. and Thompson S.G. (2002). Quantifying heterogeneity in a meta-analysis. *Stat Med.* **21**, 1539-1558.
- Holchek J.I., Herbal C.H. and Pieper R.D. (2004). Range Management Principles and Practices. Prentice Hall Publisher, New Jersey, USA.
- Kazemi M. (2019a). Comparing mineral and chemical compounds, *in vitro* gas production and fermentation parameters of some range species in Torbat-e Jam, Iran. *J. Rangel. Sci.* **9(4)**, 351-363.
- Kazemi M. (2019b). Effects of hexane extract of medicinal plant *Artemisia aucheri* Boiss. on fermentation characteristics, gas production parameters, and degradability under *in vitro* conditions. *Iranian J. Med. Arom. Plants.* **35(6)**, 902-913.
- Kazemi M. (2019c). Laboratory study of Balangu (*Lallemantia royleana*) essential oil effects on gas production kinetic, degradability, protozoan population, and some fermentation parameters of a balanced diet. *J. Anim. Sci.* **30(1)**, 29-43.
- Kazemi M. (2020). Determination of nutritional value of four plant species (*Malcolmia africana*, *Plantago lanceolata*, *Phlomis cancellata*, and *Klasea latifolia*) in rangelands of Bala Jam, Torbat-e Jam. *J. Plant Ecol. Conserv.* **7(15)**, 155-179.
- Kazemi M. (2021a). Investigating the nutritional value and ensiling possibility of purslane as a weed plant (*Portulaca oleracea* L.) with or without wheat bran. *Anim. Prod. Res.* **10**, 25-36.
- Kazemi M. (2021b). Nutritional value of some rare forage plants fed to small ruminants. *Trop. Subtrop. Agroecosyst.* **24**, 1-11.
- Kazemi M. and Ghasemi Bezdi K. (2021). An investigation of the nutritional value of camelthorn (*Alhagi maurorum*) at three growth stages and its substitution with part of the forage in Afshari ewes' diets. *Anim. Feed Sci. Technol.* **271**, 1-12.
- Kazemi M. and Ibrahimi Khoram Abadi E. (2019). The effect of hexane-extracted *Teucrium polium* L. on the ruminal microorganism's fermentation activity using *in vitro* technique. *Vet. Res. Biol. Prod.* **127**, 110-120.
- Kazemi M., Ibrahimi Khoram Abadi E. and Mokhtarpour A. (2019). Evaluation of the nutritional value of Iranian melon (*Cucumis melo* cv. Khatooni) wastes before and after ensiling in sheep feeding. *J. Livest. Sci. Technol.* **7(2)**, 9-15.
- Kazemi M., Moheghi M.M. and Tohidi R. (2021). A study on the nutritional characteristics of some plants and their effects on ruminal microbial fermentation and protozoa population. *AMB Exp.* **11**, 174-182.
- Kazemi M., Tahmasbi A.M., Naserian A.A., Valizadeh R. and Moheghi M.M. (2012). Potential nutritive value of some forage species used as ruminants feed in Iran. *African J. Biotechnol.* **11(57)**, 12110-12117.
- Kazemi M. and Valizadeh R. (2018). Nutritional value of two plant species containing *Salvia hydrangea* and *Sophora alopecuroides* in two phenological stages. *Plant Ecophysiol.* **41**, 175-178.
- Kazemi M. and Valizadeh R. (2019). Nutritive value of some rangeland plants compared to medicago sativa. *J. Rangel. Sci.* **9(2)**, 136-150.
- Keim J.P., Cabanilla J., Balocchi O.A., Pulido R.G. and Bertrand A. (2018). *In vitro* fermentation and *in situ* rumen degradation kinetics of summer forage brassica plants. *Anim. Prod. Sci.* **59(7)**, 1271-1280.
- Khazaal K., Boza J. and Ørskov E.R. (1994). Assessment of phenolics-related anti-nutritive effects in Mediterranean browse. A comparison between the use of the *in vitro* gas production technique with or without insoluble polyvinyl polypyrrolidone or nylon bag. *Anim. Feed Sci. Technol.* **49**, 133-149.
- Krogstad K.C., Tempelman P.R.J., Abney-Schulte C. and Bradford B.J. (2021). Feeding a branded, modified wet corn gluten feed to lactating dairy cows: A meta-regression approach. *Appl. Anim. Sci.* **37**, 559-573.
- Lean I.J., Rabiee A.R., Duffield T.F. and Dohoo I.R. (2009). Invited review: Use of meta-analysis in animal health and reproduction. Methods and applications. *J. Dairy Sci.* **92**, 3545-3565.
- Lean I.J., Thompson J.M. and Dunshea F.R. (2014). A meta-analysis of zilpaterol and ractopamine effects on feedlot performance, carcass traits and shear strength of meat in cattle.

- PLoS One*. **9**(12), e115904.
- Mahdavi A., Mahdavi A. Darabighane B., Mead A. and Lee M.R.F. (2019). Effects of soybean oil supplement to diets of lactating dairy cows, on productive performance, and milk fat acids profile: A meta-analysis. *Italian J. Anim. Sci.* **18**(1), 809-819.
- Makkar H.P.S. (2004). Recent advances in the *in vitro* gas method for evaluation of nutritional quality of feed resources. Pp. 55-88 in *Assessing Quality and Safety of Animal Feeds*. FAO Animal Production and Health Series 160. FAO, Rome.
- Makkar H.P.S. (2005). *In vitro* gas methods for evaluation of feeds containing phytochemicals. *Anim. Feed Sci. Technol.* **123**, 291-302.
- Menke K.H. and Steingass H. (1988). Estimation of the energetic feed value obtained from chemical analysis and *in vitro* gas production using rumen fluid. *Anim. Res. Dev.* **28**, 7-55.
- Orzuna-Orzuna J.F., Dorantes-Iturbide G., Lara-Bueno A., Mendoza-Martínez G.D., Miranda-Romero L.A. and Hernández-García P.A. (2021). Effects of dietary tannins' supplementation on growth performance, rumen fermentation, and enteric methane emissions in beef cattle: A meta-analysis. *Sustainability*. **13**, 7410-7419.
- Rahbar A., Mir jalili A. and Baghestani Meybodi N. (2008). Comparison of forage quality of 2 range species, *Artemisia aucheri* and *Peteropyron aucheri* under water spreading and control condition in Herat station, Yazd province. *Iranian J. Rangel. Desert Res.* **14**(4), 579-588.
- Sutton A. and Higgins J. (2008). Recent developments in meta-analysis. *Stat Med.* **27**, 625-650.
- Tolera A., Khazaal K. and Ørskov E.R. (1997). Nutritive evaluation of some browses species. *Anim. Feed Sci. Technol.* **67**, 181-195.
- Valizadeh R., Ghadami Kohestani M. and Melati F. (2011). Chemical composition, *in situ* Degradability and *in vitro* gas production of winterfat plant (*Eurotia ceratoides*). *Iranian J. Anim. Sci. Res.* **3**(2), 159-165.