NARDI FUNDULEA, ROMANIA ROMANIAN AGRICULTURAL RESEARCH, NO. 43, 2026
www.incda-fundulea.ro; doi.org/10.59665/rar4331 First Online: March, 2026. DII 2067-5720 RAR 2026-73

# The Biomass Yield and Biochemical Composition of Some Annual Leguminous Species in Moldova

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## ABSTRACT

**Research and utilization of the agrobiological potential of the leguminous species becomes more and more relevant. The annual leguminous species *Cicer arietinum, Glycine max, Lablab purpureus, Pisum arvense, Trifolium alexandrinum, Trigonella foenum-graecum* maintained in monoculture on experimental land in the National Botanical Garden (Institute), Chişinău, Republic of Moldova, served as subjects of study. The research revealed that the biomass yield of the studied annual leguminous species varied from 2.42 to 5.67 kg/m$^2$ fresh mass or 0.71 to 1.28 kg/m$^2$ dry matter. The biochemical composition of dry matter of the harvested whole plants had the following indices: 145-216 g/kg CP, 205-310 g/kg CF, 90-126 g/kg ash, 240-332 g/kg ADF, 376-520 g/kg NDF, 36-61 g/kg ADL, 104-271 g/kg Cel, 136-188 g/kg HC, with nutritive and energy value 63.0-70.2% DDM, RFV=113-171, 10.19-11.25 MJ/kg ME and 6.21-7.26MJ/kg NEl. The forage value of the prepared hay was characterized by 168-209 g/kg CP, 63.5-66.9% DDM, RFV=108-146, 10.26-10.76 MJ/kg ME and 6.28-6.77 MJ/kg NEl. The quality of fermented forage dry matter was: 14.7-22.2% CP, 11.2-14.6% ash, 15.0-28.1% CF, 18.1-20.7% ADF, 31.3-51.0% NDF, 2.0-4.3 % ADL, 16.1-20.7% Cel, 13.2-20.3% HC, 10.7-19.8% TSS, 650-748 g/kg DDM,10.48-11.92 MJ/kg ME and 6.50-7.94 MJ/kg NEl. The biochemical methane potential of the studied substrates varied from 318-390 l/kg. The harvested biomass from the studied annual leguminous species can be used as fodder for farm animals or as co-substrates in biogas plants for the production of biomethane as renewable energy.**

**Keywords:** annual leguminous species, biochemical composition, biochemical methane potential, forage quality, fresh mass, hay.

## INTRODUCTION

The research and utilization of leguminous species (Family: Fabaceae) have gained increasing significance in providing food, forage and raw materials for various industries. These plants fix atmospheric nitrogen, enrich soil with organic matter, and mobilize phosphorus, reducing reliance on chemical fertilizers and minimizing pollution. During their flowering period, legumes serve as a vital food source for pollinators, including bees and other insects. Additionally, certain legume species contain bioactive secondary metabolites that can improve animal health, potentially reducing the need for veterinary medications. Currently, legumes are being explored as alternative nutrient sources, such as green manures, to improve soil fertility. In the flora of Bessarabia, are 146 leguminous species, including 34 cultivated species and 75 species with forage value (Izverscaia, 2020).

The genus *Cicer* L. includes about 10 annual species and 36 perennial species. Among these, only chickpea *Cicer arietinum* L. is sporadically found in the of Bessarabia spontaneous flora. Domesticated for thousands of years, chickpea is widely cultivated in over 50 countries worldwide, is characterized by a strong, erect stem, with false-imparipinnate leaves, and racemose inflorescences with solitary flowers, elliptical pod with 1-3 globose seeds of varying coloration. The root system consists of a well-developed taproot with several lateral roots, which can reach in the soil depths exceeding 3 m; forms a symbiotic association with *Mesorhizobium ciceri*, enabling biological nitrogen fixation of about 120-150 kg /ha.

Received 27 November 2025; accepted 4 March 2026.

The genus *Trigonella* L. comprises approximately 130 species. In the flora of Bessarabia, four species are recorded, among which fenugreek (*Trigonella foenum-graecum* L.) is of particular importance. It is an annual plant, with erect stem and trifoliolate petiolate leaves; flowers are short-pedicellate, solitary or paired in the leaf axils; pod linear, erect or slightly curved with yellow to light brown, finely tuberculate seeds. The plant develops a strong, branched taproot. It forms a symbiotic association with *Rhizobium meliloti*. The genus *Pisum* L. consists of about 10 species and subspecies, spread in the Mediterranean region. In the flora of Bessarabia - 3 species (*Pisum arvense, Pisum elatius Pisum sativum).* Field pea *Pisum arvense* L. (syn, *Pisum sativum* subsp. *sativum* var. *arvense*) is a cool-season plant, stems simple or ramified in the bottom part, 150-210 cm tall, leaves consist of 2-3 pairs of leaflets with placed-denticled edges, inflorescences - bicolor flowers with dark purple wings and keel with pale purple standard; pods are about 4 cm in length and 1.5 cm in width; seeds are round, slightly angular, with varied coloring, from chartreuse to brown-red, with dark spots.

*Glycine max* (L.) Merr. is an annual, thermophilic legume native to East Asia. It was first domesticated in ancient China around 1100 BC and introduced to Europe in the 18$^{th}$ century, is sporadically found in the local spontaneous flora. The species includes ecotypes with both determinate and indeterminate growth habits.

Lablab or hyacinth bean *Lablab purpureus* (L.) Sweet is an ancient domesticated crop widely distributed in Africa, the Indian subcontinent, and Southeast Asia. It is a summer-growing annual climber that can reach lengths of 3-6 m. The leaves are alternate and trifoliolate, and the flowers range in color from white to blue or purple. The pods contain 3-6 ovoid, laterally compressed seeds that vary in size and colour. The plant develops a strong taproot with numerous lateral and well-developed adventitious roots, enabling the extraction of soil moisture from depths of at least 2 m. *Lablab purpureus* forms effective symbiotic associations with *Bradyrhizobium lablabi* sp. and *Rhizobium leguminosarum,* making it an efficient biological nitrogen fixer.

The *Trifolium* L. genus comprises 244 species out of which 25 species are of agricultural importance. *Trifolilum alexandrinum* L. is known as berseem clover, Egyptian clover or Alexandria clover, native to western Asia, has erect or ascending sparsely hairy stems, 60-80 cm tall, hollow, branching at the base, with alternate leaves, broad leaflets, yellowish-white flowers, pod with one yellow to purplish-red seed. It can withstand some drought and short periods of waterlogging, moderately tolerant of salinity; frost tolerance, down to -6°C and as low as -15°C for some cultivars.

The crops such as *Cicer arietinum*, *Glycine max, Lablab purpureus, Pisum arvense, Trifolium alexandrinum,* and *Trigonella foenum-graecum* are extensively studied and bred in various geographical regions. New cultivars are being developed for multiple uses, including seed production, hay, pasture, silage and green manure (Zamfir et al., 2001; Cupić et al., 2011; Habibpour et al., 2012; Acikgoz et al., 2013; Heuze et al., 2015, 2016a, b, c; 2019; Sayar and Han, 2016; Bărbieru, 2021; Aleme et al., 2022; Akbay et al., 2023; Jabessa et al., 2023; Yucesoy and Garipoğlu, 2025).

The main objectives of this study were to evaluate the yield and quality of green mass, hay, and fermented fodder from annual leguminous species - *Cicer arietinum*, *Glycine max, Lablab purpureus, Pisum arvense, Trifolium alexandrinum,* and *Trigonella foenum-graecum* - and to assess their potential as fodder for livestock or as substrates for biomethane production.

## MATERIAL AND METHODS

The annual leguminous species *Cicer arietinum, Glycine max, Lablab purpureus, Pisum arvense, Trifolium alexandrinum, Trigonella foenum-graecum,* maintained in monoculture on experimental land in the National Botanical Garden (Institute), MSU, Chişinău, Republic of Moldova, served as subjects of study. The experimental design

utilized a randomized complete block design with four replications, and the plots measured 50 m². Plant growth, development, and productivity were evaluated based on the standard methods outlined in NBGI. Green mass samples were collected during the flowering stage. The dry matter content was determined by drying the samples at 105°C until they reached a constant weight. Hay was dried directly in the field, while fermented fodder was made from whole plants, chopped into small pieces, and packed into glass containers Both fresh and fermented fodder samples were dried in a forced-air oven at 60°C. After drying, the biological material was finely ground using a laboratory ball mill. The biomass quality was assessed by analyzing several indices: crude protein (CP), crude fibre (CF), crude ash (CA), total soluble sugars (TSS), acid detergent fibre (ADF), neutral detergent fibre (NDF), and acid detergent lignin (ADL), using near-infrared spectroscopy (NIRS) on the PERTEN DA 7200 device at the Research and Development Institute for Grassland in Braşov, Romania. Further, the concentrations of hemicellulose (HC), cellulose (Cel), digestible dry matter (DDM), digestible energy (DE), metabolizable energy (ME), net energy for lactation (NEl), and relative feed value (RFV) were calculated following standard procedures. The carbon content of the substrates was estimated using an empirical equation from Badger et al. (1979), and the methane potential was calculated based on Dandikas et al. (2015).

## RESULTS AND DISCUSSION

An analysis of the agrobiological characteristics of the studied annual leguminous crops (Table 1) revealed that the budding-flowering stage occurred at different times for each species: *Cicer arietinum* and *Pisum arvense* reached this stage at the end of May to early June, *Trifolium alexandrinum* and *Trigonella foenum-graecum* - at the end of June, *Glycine max* - at the end of July, and *Lablab purpureus* in September. It is noteworthy that at the time of green mass harvest, shoot length varied from 50 cm in *Cicer arietinum* to 230 cm in *Lablab purpureus*. The productivity ranged from 2.42 kg/m² to 5.67 kg/m² of green mass, or 0.71 kg/m² to 1.28 kg/m² of dry matter, with 57.12% to 69.23% of the forage mass consisting of leaves and flowers.

Different results regarding the forage productivity of annual leguminous species, depending on cultivars and management practices, are reported in the specialized literature. Cojocariu et al. (2008) reported that the forage productivity of berseem clover varied from 5.74 to 10.04 t/ha of dry matter. Lingorski (2011) observed that forage dry mass yields of annual spring legumes ranged from 3.74-4.01 t/ha for field pea, 3.12-4.06 t/ha for chickling vetch, and 2.67-2.89 t/ha for soybean. Arslan et al. (2012), Ates (2012) noted that the forage productivity of pure stand pea was 45.6-56.4 t/ha of green mass or 9.4-15.11 t/ha of dry matter. Açikgöz et al. (2013) reported that the dry matter yield of forage soybean varied from 8.25 to 15.80 t/ha. Mihailovic et al. (2013) observed that forage yields of soybean accessions reached 63.9-82.4 t/ha of green mass or 15.1-18 t/ha of dry matter. Obour et al. (2015) noted that fenugreek forage dry matter productivity varied from 672 to 910 lb/A. Bozhanska et al. (2016) reported that the forage yield of Egyptian clover reached 26.4 t/ha of green mass or 7.8 t/ha of dry matter. Ates and Tenikecier (2020) found that the field pea genotypes studied at full-bloom stage achieved main stem lengths of 115-130 cm, 35.66-50.33 t/ha of green mass, and 6.45-11.77 t/ha of hay. Adem et al. (2021) reported that the dry matter yield of lablab accessions ranged from 11.21 to 19.60 t/ha. Bacchi et al. (2021) mentioned that the forage dry matter productivity of pure stand pea was 5.44 t/ha, while berseem clover yielded 5.24 t/ha. Homan et al. (2021) showed that the forage soybean in pure culture achieved a plant height of 134.8 cm, a biomass yield of 32.9 t/ha, and 9.7 t/ha of dry matter. Akbay et al. (2023) observed that the plant height of the studied fenugreek genotypes ranged from 42.05 to 69.63 cm, with yields of 8.21-14.8 t/ha of green mass and 1.47-2.04 t/ha of hay.

*Table 1*. Some agrobiological peculiarities of studied annual leguminous species

| Species | Plant length, cm | Productivity, kg/m$^2$ | | Content of leaves and flowers in fodder, % |
|---|---|---|---|---|
| | | green mass | dry matter | |
| *Cicer arietinum* | 50 | 2.42 | 0.71 | 67.49 |
| *Glycine max* | 110 | 3.83 | 1.07 | 61.70 |
| *Lablab purpureus* | 230 | 5.67 | 1.28 | 68.57 |
| *Pisum arvense* | 163 | 5.14 | 1.03 | 69.23 |
| *Trifolium alexandrinum* | 61 | 3.15 | 0.88 | 57.12 |
| *Trigonella foenum-graecum* | 55 | 2.95 | 0.55 | 66.30 |

Jabessa et al. (2023) reported that the dry matter yields of *Lablab purpureus* ranged from 11.12 to 15.43 t/ha. Sterna et al. (2023) observed that the forage productivity of soybean cultivars harvested at different developmental stages ranged from 13.5 to 34.7 t/ha of green forage or 3.9 to 11.2 t/ha of dry matter. Fotohi Chiyaneh et al. (2024) reported that the forage dry matter yield of fenugreek was 3.7 t/ha, while pea monoculture produced the highest forage yield, reaching 11 t/ha. Kosev et al. (2025) noted that the forage productivity of *Lablab purpureus* was 25 t/ha of green mass and 4484.6 kg/ha of dry matter, while *Trigonella foenum-graecum* yielded 10.7 t/ha of green mass and 2339.8 kg/ha of dry matter.

The quality indices of dry matter from harvested annual leguminous plants are presented in Table 1. The nutrient concentrations ranged from 145 to 216 g/kg for crude protein (CP), 205 to 310 g/kg for crude fiber (CF), 90 to 126 g/kg for ash, 36 to 61 g/kg for acid detergent lignin (ADL), 104 to 271 g/kg for cellulose (Cel), and 136 to 188 g/kg for hemicellulose (HC). The nutritive and energy values ranged from 630 to 702 g/kg for digestible dry matter (DDM), 113 to 171 for relative feed value (RFV), 10.19 to 11.25 MJ/kg for metabolizable energy (ME), and 6.21 to 7.26 MJ/kg for net energy for lactation (NEl). It should be noted that *Trigonella foenum-graecum*, *Pisum arvense*, and *Cicer arietinum* plants were characterized by a higher level of crude protein. The concentration of acid detergent fiber and acid detergent lignin in the forage from *Cicer arietinum*, *Trigonella foenum-graecum*, and *Pisum arvense* were lower, which positively affected the digestibility, nutritional value, and energy supply of the feed. *Lablab purpureus* forage had optimal levels of crude protein, comparable to that of *Glycine max*, but with lower levels of crude fiber, acid detergent lignin, cellulose, hemicellulose, and total soluble sugars. The highest levels of carbohydrates and acid detergent lignin, along with the lowest levels of crude protein, relative feed value, and energy concentrations, were found in *Trifolium alexandrinum* forage compared to the other investigated species.

The results of the estimation of quality indices for green mass from tested annual leguminous species are well-documented in the scientific literature. According to Cojocariu et al. (2008), the nutrient content of berseem clover forage ranged from 23.4% to 29.8% CP, 2.82% to 3.42% EE, 30.45% to 32.25% CF, 33.50% to 36.65% ADF, 52.70% to 54.60% NDF, 9.72% to 18.45% ash, 1.28% to 1.40% Ca, and 0.28% to 0.56% P. Lingorski (2011) reported that forage from field pea contained 19.56% CP, 3.71% EE, 23.51% CF, 8.27% ash, 1.06% Ca, and 0.29% P, while forage from chickling vetch contained 18.05% CP, 3.34% EE, 24.15% CF, 8.24% ash, 0.89% Ca, and 0.28% P. Soybean forage contained 17.71% CP, 3.21% EE, 23.15% CF, 8.58% ash, 1.35% Ca and 0.23% P. Mohsen et al. (2011) reported that the first-cut berseem clover forage contained 100-116 g/kg DM with 17.6% to 20.4% CP, 3.16% to 3.58% EE, 24.2% to 26.5% CF, 35.7% to 37.9% NFE, and 14.9% to 16.1% ash. Abdouli et al. (2012) found that fenugreek cut at the peak flowering stage contained 130 g/kg DM, 94.8% OM, 17.9% CP, 36.8% NDF, 31.6% ADF, 6.6% ADL, and 74.8% IVDMD. Ates et al. (2012) mentioned that pea forage dry matter contained 15.04% CP, 41.7% NDF, 30.8%

ADF, 1.63% Ca, and 0.28% P. Heuze et al. (2015; 2016a, b, c; 2019) reported the following fresh mass nutritive values for several species: *Pisum sativum* had 156 g/kg DM, 17.7% CP, 3.1% EE, 22.9% CF, 31.1% NDF, 23.1% ADF, 4.8% lignin, 9.1% ash, 18.6 g/kg Ca, 3.9 g/kg P, 72.3% DOM, 18.6 MJ/kg GE, 12.8 MJ/kg DE, and 10.2 MJ/kg ME; *Trigonella foenum-graecum* had 308 g/kg DM, 16.5% CP, 2.1% EE, 25.8% CF, 38.8% NDF, 30.3% ADF, 14.0% ash, 68.0% DOM, 17.4 MJ/kg GE, 11.4 MJ/kg DE, and 9.1 MJ/kg ME; *Trifolium alexandrinum* had 125 g/kg DM, 19.9% CP, 3.2% EE, 23.3% CF, 44.8% NDF, 27.6% ADF, 5.1% lignin, 5.1% WSC, 15.4% ash, 19.3 g/kg Ca, 2.7 g/kg P, 73.0% DOM, 17.4 MJ/kg GE, 12.2 MJ/kg DE, and 9.6 MJ/kg ME; *Lablab purpureus* had 211 g/kg DM, 18.4% CP, 2.6% EE, 28.2% CF, 44.6% NDF, 32.0% ADF, 7.2% lignin, 11.1% ash, 13 g/kg Ca, 2.9 g/kg P, 67.0% DOM, 18.2 MJ/kg GE, 11.7 MJ/kg DE, and 9.2 MJ/kg ME; *Glycine max* had 240 g/kg DM, 15.7% CP, 4.4% EE, 31.2% CF, 48.1% NDF, 31.2% ADF, 5.8% lignin, 9.3% ash, 14.8 g/kg Ca, 2.7 g/kg P, 64% DOM, 18.9 MJ/kg GE, 11.6 MJ/kg DE, and 9.2 MJ/kg ME. Kumar et al. (2015) reported that the nutrient content and forage value of *Trifolium alexandrinum* was 89.91% OM, 17.74% CP, 2.83% EE, 43.28% NDF, 24.08% ADF, 7.09% ADL, 19.20% HC, 15.04% Cel, 10.09% ash, 8.51 MJ/kg ME, 614.4 g/kg TDDM, and 637.6 g/kg TDOM. Salama (2015) revealed that the nutrient composition of berseem clover plants was 17.60% CP, 39.91% NDF, 27.20% ADF, and 4.09% ADL. Obour et al. (2015) noted that the dry matter of fenugreek forage contained 19.2-19.9% CP, 33.7-34.5% NDF, 28.8-29.3% ADF, 15.1-16.1 g/kg Ca, 2.4-2.5 g/kg P, 70.2-70.7% TDN, and an RFV of 181-189. Kirilov et al. (2016) compared the quality of green mass from whole plants of annual legumes harvested at the flowering-pod formation stage, reporting that *Cicer arietinum* had 14.06% CP, 3.44% EE, 27.14% CF, 44.04% NFE, and 11.32% ash; *Pisum sativum* had 13.04% CP, 2.14% EE, 25.06% CF, 58.30% NFE, and 8.01% ash; and *Glycine max* had 13.13% CP, 2.48% EE, 29.87% CF, 45.50% NFE, and 9.02% ash. Bozhanska et al. (2016) provided the quality indices for Egyptian clover forage, which were 11.18% CP, 3.02% EE, 33.44% CF, 42.86% NDF, 30.95% ADF, 6.13% ADL, 11.91% HC, 24.82% Cel, 12.24% ash, 15.93 MJ/kg NE, 6.71 MJ/kg EE, 0.61 FUM, and 0.55 FUG. Uslu et al. (2018) reported that *Trigonella foenum-graecum* forage contained 252.7 g/kg DM with 20.17% CP, 1.49% EE, 20.24% ADF, 22.86% NDF, 9.85% ash, 72.87% OMD, and 9.98 MJ/kg ME. Bacchi et al. (2021) mentioned that berseem clover herbages mowed at the early flowering stage contained 178.3 g/kg DM with 19.04% CP, while pea forage contained 152.1 g/kg DM with 22.75% CP. Akbay et al. (2020) found that fenugreek forage, harvested at different times, contained 19.34-22.44% CP, 1.38-1.90% EE, 25.39-35.31% ADF, 39.89-55.25% NDF, 8.03-10.71% ash, 64.26-73.40% OMD, and 8.75-9.86 MJ/kg ME. Niu et al. (2021) found that the chemical composition of the forage dry matter from *Trigonella foenum-graecum* ranged from 17.75-23.55% CP, 26.42-37.79% NDF, 22.93-34.44% ADF, and 10.92-11.0% ash. For *Medicago sativa*, the composition ranged from 17.27-21.16% CP, 33.14-43.89% NDF, 27.99-37.11% ADF, and 10.17-10.20% ash. Aleme et al. (2022) mentioned that a newly released *Lablab* cultivar was characterized by 22.1-23.6% CP, 13.5-15.7% ash, 47.6-49.3% NDF, 33.0-35.7% ADF, 6.3-6.7% ADL, 609-621 g/kg IVDMD, and an RFV between 113.6 and 117.6. Arif et al. (2022) revealed that the nutritive value of *Trifolium alexandrinum* forage ranged from 17.05-19.20% CP, 59.66-62.46% TDN, an RFV of 120.90-137.46, and 1.45-1.51 Mcal/kg NEl. Patidar et al. (2022) found that berseem clover forage was characterized by 10.16% ash, 17.69% CP, 1.84% EE, 43.82% NDF, 32.40% ADF, 11.46% starch, 57.14% TDN, 17.34 MJ/kg GE, 10.79 MJ/kg DE, and 9.00 MJ/kg ME. Rady et al. (2022) noted that the chemical composition and forage value of *Trifolium alexandrinum* included 826.5 g/kg OM, 15.53% CP, 1.71% EE, 13.69% NSC, 55.32% NDF, 49.01% ADF, 6.31% HC, 33.50% Cel, 15.51% lignin, 759.4 g/kg

TDDM, and 733.9 g/kg TDOM. In our previous publication (Ţîţei, 2022; Ţîţei et al., 2023), we found that harvested *Glycine max* whole plants contained 265 g/kg DM with 17.8% CP, 28.6% CF, 9.4% ash, 31.0% ADF, 48.4% NDF, 4.9% ADL, 14.2% TSS, 26.1% Cel, 17.4% HC, 686 g/kg DMD, 634 g/kg DOM, RFV = 124, 12.73 MJ/kg DE, 10.48 MJ/kg ME, and 6.46 MJ/kg NEl, while *Cicer arietinum* whole plants cut at the flowering-early pod stage contained 19.31% CP, 4.23% EE, 22.62% CF, 43.57% NFE, 10.26% ash, 1.45% Ca, 0.32% P, 18.46 MJ/kg GE, 9.83 MJ/kg ME, and 5.74 MJ/kg NEl. Ahmed et al. (2023) mentioned that berseem clover forage had 259.9 g/kg DM, 88.47% OM, 16.45% CP, 51.96% NDF, 24.41% ADF, 5.84% ADL, 23.57% Cel, and 25.55% HC. Akbay et al. (2023) reported that the chemical composition of the forage dry matter from fenugreek genotypes ranged from 16.97-22.66% CP, 50.65-54.45% NDF, 36.59-41.57% ADF, 56.52-60.40% DDM, and an RFV of 99.71-109.33. Aldemir et al. (2023) found that the chemical composition of the forage dry matter from *Trigonella foenum-graecum* was 10.44-10.95% CP, 47.44-48.60% NDF, 28.79-29.13% ADF, and 16.26-16.48% ash. Ishiaku et al. (2023) found that fresh *Lablab* forage had 22.53% CP, 6.37% EE, 22.14% CF, 38.69% NFE, 52.08% NDF, 27.82% ADF, 14.22% ADL, 24.26% HC, 13.60% Cel, 10.32% ash, RFV = 120.03, and 12.13 MJ/kg ME. Jabessa et al. (2023) stated that the studied genotypes of *Lablab purpureus* contained 10.8-23.5% CP, 11.2-22.23% NDF, 9.25-21.00% ADF, 3.42-14.75% ADL, 499.3-624.0 g/kg IVDMD, and 422.6-500.4 g/kg IVOMD.

*Table 2.* The biochemical composition and the fodder value of the fresh mass of studied annual leguminous species

| Indices | *Cicer arietinum* | *Glycine max* | *Lablab purpureus* | *Pisum arvense* | *Trifolium alexandrinum* | *Trigonella foenum-graecum* |
|---|---|---|---|---|---|---|
| Crude protein, g/kg DM | 197 | 175 | 176 | 210 | 145 | 216 |
| Crude fibre, g/kg DM | 205 | 310 | 261 | 251 | 279 | 244 |
| Minerals, g/kg DM | 126 | 86 | 90 | 120 | 100 | 119 |
| Acid detergent fibre, g/kg DM | 240 | 332 | 281 | 269 | 302 | 261 |
| Neutral detergent fibre, g/kg DM | 376 | 520 | 453 | 438 | 479 | 416 |
| Acid detergent lignin, g/kg DM | 46 | 61 | 44 | 36 | 51 | 37 |
| Total soluble sugars, g/kg DM | 181 | 181 | 158 | 156 | 140 | 160 |
| Cellulose, g/kg DM | 194 | 271 | 237 | 233 | 251 | 224 |
| Hemicellulose, g/kg DM | 136 | 188 | 171 | 169 | 177 | 155 |
| Digestible dry matter, g/kg DM | 702 | 630 | 670 | 679 | 654 | 686 |
| Relative feed value | 174 | 113 | 138 | 144 | 127 | 153 |
| Digestible energy, MJ/ kg | 13.70 | 12.41 | 13.13 | 13.29 | 12.84 | 13.41 |
| Metabolizable energy, MJ/ kg | 11.25 | 10.19 | 10.78 | 10.91 | 10.54 | 11.01 |
| Net energy for lactation, MJ/ kg | 7.26 | 6.21 | 6.80 | 6.93 | 6.60 | 7.02 |

Kondratenko et al. (2023) reported that *Trigonella foenum-graecum* forage had 163.7-165.8 g/kg DM with nutritional and energy values ranging from 20.49-20.89% CP, 1.81-2.05% EE, 17.62-21.09% CF, 149.7-163.9 g/kg DP, 10.7-16.5 g/kg Ca, 4.8-6.44 g/kg P, 0.89-0.98 nutritive units/kg, 18.4-18.7 MJ/kg GE, and 10.55-10.95 MJ/kg ME. Munza et al. (2023) mentioned that white *Lablab* forage contained 12.87-14.70% CP, 7.05-8.13% EE, 22.14% CF, 35.88-44.37% NFE, 23.93-24.80% NDF, 19.84-22.86% ADF, 6.73-7.75% ADL, 7.17-8.87% HC, 6.53-7.52% Cel, and 10.15-11.82% ash. Sangjan et al. (2023) reported that winter field pea forage contained 15.8-16.5% CP, 1.4-1.7% EE, 33.9-37.0% NDF, 25.2-28.8% ADF, 6.3-7.5% ash, 34.5-37.3% NFC, and 650-660 g/kg TDN. Belete et al. (2024) stated that the chemical composition and *in vitro* organic matter digestibility of fresh *Lablab* forage ranged from 3.8 to 8.8% ash, 15.0-26.5% CP, 44.0-59.4% NDF, 24.2-47.3% ADF, 1.7-8.3% ADL, and 42.2-68.7% IVOMD. Fotohi Chiyaneh et al. (2024) remarked that the chemical composition of the fenugreek forage cut at the middle flowering stage was 12.36% CP, 24.2% CF,

41.11% NDF, 28.42% ADF, 10.46% ash, 19.95% WSC, 646.64 g/kg TDN, 667.63 g/kg DDM, with an RFV of 161.86 and 1.54 Mcal/kg NEl, while pea forage had 16.01% CP, 22.22% CF, 39.10% NDF, 26.77% ADF, 11.54% ash, 22.3% WSC, 667.88 g/kg TDN, 680.45 g/kg DDM, an RFV of 161.86, and 1.60 Mcal/kg NEl. Georgieva et al. (2024) mentioned that the biochemical composition and nutritive value of fenugreek forage ranged from 10.28-25.23% CP, 11.28-24.37% CF, 18.49-40.64% NDF, 2.47-5.86% ADL, 12.3-18.7 g/kg Ca, 3.1-4.4 g/kg P, and 480-763 g/kg IVDOM. Folina et al. (2025) reported that *Trigonella foenum-graecum* plants contained 11.31-17.29% CP, 1.43-2.00% EE, 17.07-19.10% CF, 22.73-37.44% NFC, 8.80-9.71% ash, and 67.64-71.65% DDM. Yücesoy and Garipoğlu (2025) observed that depending on the soybean variety, the nutrient composition varied from 18.17 to 20.48% CP, 39.49-42.16% NDF, and 28.54-30.56% ADF.

The nutritional value of hay depends on plant species, climate conditions, mowing period, haymaking technology, and the conditions under which it is stored. Analyzing the results regarding the quality of hay prepared from the studied annual leguminous species (Table 3), we would like to highlight that the prepared hays contained 168-209 g/kg CP, 94-134 g/kg ash, 264-316 g/kg CF, 286-339 g/kg ADF, 425-540 g/kg NDF, 42-62 g/kg ADL, 229-279 g/kg Cel, 134-201 g/kg HC, and 55-158 g/kg TSS. The hays also showed 63.5-66.9% DDM, with an RFV ranging from 108 to 146, 10.26-10.76 MJ/kg ME, and 6.28-6.77 MJ/kg NEl. It was found that during the haymaking process, the concentration of cell wall fractions increased, while the content of total soluble sugars, dry matter digestibility, relative feed value and energy concentration decreased when compared to the initial green mass. The hay prepared from *Trigonella foenum-graecum*, *Cicer arietinum* and *Pisum arvense* was characterized by high amounts of crude protein. It was also determined that the hay prepared from *Glycine max*, *Lablab purpureus* and *Vicia sativa* did not show significant differences in crude protein content (163-168 g/kg). The hay from *Cicer arietinum* and *Pisum arvense* exhibited higher digestibility, relative feed value, metabolizable energy and net energy for lactation. In contrast, the hay from *Glycine max* had higher cell wall concentrations, resulting in a lower nutritional and energy value compared to other types of hay. The quality indices of hay prepared from *Lablab purpureus* were more favorable compared to those of the hay from *Trifolium alexandrinum* and *Trigonella foenum-graecum.*

*Table 3.* The biochemical composition and the fodder value of the hay from studied annual leguminous species

| Indices | *Cicer arietinum* | *Glycine max* | *Lablab purpureus* | *Pisum arvense* | *Trifolium alexandrinum* | *Trigonella foenum-graecum* |
|---|---|---|---|---|---|---|
| Crude protein, g/kg DM | 194 | 163 | 167 | 201 | 168 | 209 |
| Crude fibre, g/kg DM | 238 | 316 | 273 | 264 | 289 | 310 |
| Minerals, g/kg DM | 134 | 90 | 94 | 121 | 106 | 124 |
| Acid detergent fibre, g/kg DM | 283 | 339 | 305 | 286 | 303 | 326 |
| Neutral detergent fibre, g/kg DM | 438 | 540 | 503 | 425 | 437 | 501 |
| Acid detergent lignin, g/kg DM | 54 | 62 | 45 | 42 | 47 | 47 |
| Total soluble sugars, g/kg DM | 89 | 88 | 158 | 102 | 113 | 55 |
| Cellulose, g/kg DM | 229 | 277 | 260 | 244 | 256 | 279 |
| Hemicellulose, g/kg DM | 155 | 201 | 198 | 139 | 134 | 175 |
| Digestible dry matter, g/kg DM | 669 | 625 | 650 | 666 | 653 | 653 |
| Relative feed value | 142 | 108 | 120 | 146 | 139 | 118 |
| Digestible energy, MJ/ kg | 13.11 | 12.32 | 12.76 | 13.06 | 12.82 | 12.50 |
| Metabolizable energy, MJ/ kg | 10.76 | 10.12 | 10.48 | 10.72 | 10.53 | 10.26 |
| Net energy for lactation, MJ/ kg | 6.77 | 6.13 | 6.52 | 6.74 | 6.54 | 6.28 |

Some authors have reported various findings regarding the quality of hay prepared from annual leguminous species. Gaafar et al. (2011) noted that the nutritional value of berseem clover hay was 14.4% CP, 2.45% EE, 27.6% CF, 45.2% NFE, 10.3% ash, 61.1% TDN, 65.2% DDM, and 2694 kcal/kg DE. Das et al. (2015) found that berseem clover hay contained 921.3 g/kg DM, 15.69% CP, 2.26% EE, 28.59% CF, 41.56% NFE, 53.98% NDF, 37.90% ADF, 7.01% ADL, 16.08% HC, 30.89% Cel, and 11.69% ash. Heuze et al. (2016a, b, c) mentioned that berseem clover hay had 15.7% CP, 2.4% EE, 26.9% CF, 49.3% NDF, 30.2% ADF, 13.7% ash, 21.9 g/kg Ca, 2.4 g/kg P, 63.1% DOM, 17.5 MJ/kg GE, 10.4 MJ/kg DE, and 8.3 MJ/kg ME. Soybean hay contained 915 g/kg DM, 15.9% CP, 5.4% EE, 33.8% CF, 47.0% NDF, 34.9% ADF, 7.0% lignin, 7.6% ash, 8.6 g/kg Ca, 1.9 g/kg P, 59% DOM, 19.6 MJ/kg GE, 10.9 MJ/kg DE, and 8.6 MJ/kg ME. Lablab hay had 912 g/kg DM, 16.3% CP, 2.2% EE, 32% CF, 45.5% NDF, 31.9% ADF, 6.7% lignin, 10.5% ash, 14.3 g/kg Ca, 2.8 g/kg P, 60% ODM, 18.3 MJ/kg GE, 10.3 MJ/kg DE, and 8.2 MJ/kg ME. Uslu et al. (2018) reported that the quality indices of hay prepared from *Trigonella foenum-graecum* were 20.17% CP, 1.49% EE, 22.86% NDF, 20.24% ADF, 9.86% ash, 72.87% OMD, and 9.98 MJ/kg ME. Pursley (2019) remarked that forage pea hay contained 882.6 g/kg DM, 15.06% CP, 1.9% EE, 19.48% starch, 37.97% NDF, 28.7% ADF, 7.20% ash, 14.1 g/kg Ca, and 2.2 g/kg P. In our previous publication (Ţîţei, 2022; Ţîţei et al., 2023), we reported that the nutrient concentration and economic value of soybean hay were as follows: 17.3% CP, 30.3% CF, 10.5% ash, 33.1% ADF, 50.4% NDF, 5.3% ADL, 11.0% TSS, 27.8% Cel, 17.3% HC, 64.6% DMD, 57.8% DOM, RFV = 116, 10.20 MJ/kg ME, and 6.22 MJ/kg NEl; the chickpea hay prepared contained 19.77% CP, 2.64% EE, 27.01% CF, 39.21% NFE, 11.37% ash, 1.46% Ca, 0.31% P, 18.07 MJ/kg GE, 8.81 MJ/kg ME, and 4.98 MJ/kg NEl. Soufan and Al-Suhaibani (2021) reported that the quality of pea hay was characterized by 8.00% ash, 17.0% CP, 26.60% CF, 1.78% EE, 6.34% WSC, 43.42% NDF, 32.29% ADF, and 76.07% DDM. Sainz-Ramírez et al. (2023) mentioned that sunflower-chickpea hay quality indices were as follows: 17.34% CP, 17.12% EE, 42.52% NDF, 27.03% ADF, 11.00% ash, and 67.84% IVDOM.

Fermented forages are an important source of nutrients for livestock, particularly during the autumn to mid-spring period, and they have a substantial influence on feed formulation, animal health, and productive performance. The results concerning the quality of fermented forage prepared from several annual leguminous species are presented in Table 4. The quality of the fermented forage was found to be species-dependent. Thus, the forage produced from *Lablab purpureus* and *Trifolium alexandrinum* exhibited optimal pH values and, according to the Moldovan standard SM 108, met the criteria for first-class quality. The quality of fermented forage dry matter had the following indices: 14.7-22.2% CP, 11.2-14.6% ash, 15.0-28.1% CF, 18.1-20.7% ADF, 31.3-51.0% NDF, 2.0-4.3 % ADL, 16.1-20.7% Cel, 13.2-20.3% HC, 10.7-19.8% TSS, 650-748 g/kg DDM,10.48-11.92 MJ/kg ME and 6.50-7.94 MJ/kg NEl. The ensiled biomass of the studied annual leguminous species was found to contain appreciable amounts of minerals. In the ensiled mass of *Cicer arietinum*, the contents of crude protein, minerals and total soluble sugars increased, while the proportions of cell wall fractions (NDF, ADF and ADL) decreased as compared with the initial green biomass. These changes positively affected digestibility, nutritional value and the energy supply of the ensiled feed. In the ensiled mass of *Glycine max*, crude protein content decreased, whereas the levels of structural and soluble carbohydrates increased in comparison with the fresh green mass. Notably, the ensiled forage from *Lablab purpureus* exhibited a higher crude protein content than that of *Glycine max* and *Trifolium alexandrinum*; however, it contained lower amounts of total soluble sugars and lignin.

Gaafar et al. (2011) reported that the nutritive value of berseem clover silage was: 15.3% CP, 2.5% EE, 25.2% CF, 44.3% NFE, 12.8% ash, 69.1% DDM, 2760 kcal/kg DE. Mohsen et al. (2011) established that the quality indices of silage from *Trifolium alexandrinum* were: 349-357 g/kg DM, pH=4.15-4.22, 14.6-15.0% CP, 3.45-3.51% EE, 30.3-31.0% CF, 39.5-39.7% NFE, 11.20-11.80% ash, 69.7-70.7% DDM, 69.2-72.1% DOM.

*Table 4.* The biochemical composition and the nutritive value of the fermented forage from of studied annual species

| Indices | *Cicer arietinum* | *Glycine max* | *Lablab purpureus* | *Trifolium alexandrinum* |
|---|---|---|---|---|
| pH index | 4.40 | 4.54 | 4.11 | 4.22 |
| Crude protein, g/kg DM | 222 | 161 | 186 | 147 |
| Crude fibre, g/kg DM | 150 | 273 | 281 | 266 |
| Minerals, g/kg DM | 146 | 126 | 129 | 112 |
| Acid detergent fibre, g/kg DM | 181 | 286 | 307 | 283 |
| Neutral detergent fibre, g/kg DM | 313 | 456 | 510 | 452 |
| Acid detergent lignin, g/kg DM | 20 | 41 | 37 | 43 |
| Total soluble sugars, g/kg DM | 198 | 173 | 107 | 153 |
| Cellulose, g/kg DM | 161 | 245 | 270 | 250 |
| Hemicellulose, g/kg DM | 132 | 170 | 203 | 169 |
| Digestible dry matter, g/kg DM | 748 | 676 | 650 | 669 |
| Relative feed value | 222 | 145 | 119 | 138 |
| Digestible energy, MJ/ kg | 14.52 | 13.23 | 12.76 | 13.11 |
| Metabolizable energy, MJ/ kg | 11.92 | 10.87 | 10.48 | 10.76 |
| Net energy for lactation, MJ/ kg | 7.94 | 6.73 | 6.50 | 6.74 |

Yucel et al. (2013) found that chickpea silage had pH = 5.48-5.66, 10.4-12.4% CP, 39.0-39.3% ADF, 40.0-42.5% NDF, 58.3-58.5% OMD, and RFV = 138-139, while soybean silage had pH = 5.40-5.47, 12.4-13.7% CP, 36.5-38.4% ADF, 40.5-41.8% NDF, 58.9-60.5% OMD, and RFV = 140-154. Wangila et al. (2021) mentioned that *Lablab purpureus* silage was characterized by pH = 4.37-4.81, 427.4 g/kg DM, 16.0% CP, 8.9% ash, 42.4% NDF, 28.6% ADF, 7.3% ADL, and 754 g/kg IVDMD. In our previous publication (Ţîţei et al., 2023), we found that chickpea ensiled fodders had pH = 4.40-4.47, 27.6-34.5 g/kg lactic acid, 2.5-2.8 g/kg acetic acid, 0.3-0.4 g/kg butyric acid, 19.64-20.61% CP, 25.67-26.15% EE, 24.57-25.32% CF, 36.77-39.20% NFE, 10.92-11.15% ash, 1.39-1.41% Ca, 0.35-0.39% P, 18.75-18.90 MJ/kg GE, 9.81-9.81 MJ/kg ME, and 5.47-5.55 MJ/kg NEl. Ahmed et al. (2023) mentioned that the quality indices of *Trifolium alexandrinum* ensiled forage were: 152.0-176.8 g/kg DM, pH = 4.5-4.9, 19.1-57.5 g/kg lactic acid, 4.7-28.9 g/kg butyric acid, 19.4-26.6 g/kg acetic acid, 88.03-88.25% OM, 14.55-17.61% CP, 49.39-53.93% NDF, 33.08-38.48% ADF, 8.16-9.36% ADL, 24.92-29.33% Cel, 15.43-16.89% HC, and 62.23% TDOM.

The use of phytomass from leguminous species as feedstock for various energy purposes enhances the potential of bioenergy and contributes to reducing greenhouse gas emissions. This is achieved through symbiotic nitrogen fixation, which can lower the need for inorganic nitrogen fertilizers in conventional farming when the digestate is applied as fertilizer to non-leguminous crops. The quality indices of substrates from the studied annual leguminous species and their biochemical methane potential are presented in Table 5. The nitrogen content in the studied substrates ranged from 23.2 to 34.6 g/kg, with the carbon content estimated to be between 474 and 508 g/kg. The C/N ratio varied from 13.4 to 21.0. These substrates had acceptable concentrations of hemicellulose (132-188 g/kg) and acid detergent fiber (20-61 g/kg), with their biochemical methane potential ranging from 318 to 385 L/kg VS. The highest methane potential was observed in *Trigonella foenum-graecum* and *Pisum arvense* fresh mass

substrates, while the lowest values were found in *Glycine max* and *Trifolium alexandrinum* substrates. The reduced content of acid detergent lignin in ensiled substrates may positively influence bacterial activity and decomposition processes, which contributed to the higher biochemical methane potential of *Cicer arietinum* ensiled substrate (385 L/kg VS), followed by *Lablab purpureus* silage (359 L/kg VS).

Numerous studies have reported various quality indices and methane yields of substrates derived from annual leguminous species. According to Herrmann et al. (2016), the forage pea ensiled mass had C/N=27, 12% ADL and methane yield was 245 L/kg VS. Stinner (2015) reported that the substrate from Egyptian clover had 69% degradability and 308 L/kg VS methane yields, while substrates from forage pea 73% degradability and 317 L/kg VS methane yields. Morozova et al. (2020) remarked that *Glycine max* substrates had 288.8 g/kg dry matter with 90.17% organic matter, and the methane yield reached 266 L/kg VS. Pabón-Pereira et al (2020) reported that the biochemical methane potential was 370 L/kg ODM in *Pisum sativum* substrates, 350 l/kg ODM in *Vicia faba* substrates and 290 l/kg ODM in *Vicia sativa* substrates.

*Table 5.* Biochemical biomethane production potential of substrates from studied annual leguminous species

| Indices | *Cicer arietinum* | | *Glycine max* | | *Lablab purpureus* | | *Pisum arvense* | *Trifolium alexandrinum* | | *Trigonella foenum-graecum* |
|---|---|---|---|---|---|---|---|---|---|---|
| | fresh mass | silage | fresh mass | silage | fresh mass | silage | fresh mass | fresh mass | silage | fresh mass |
| Crude protein, g/kg | 197 | 222 | 175 | 161 | 176 | 186 | 210 | 145 | 147 | 216 |
| Minerals, g/kg | 126 | 146 | 86 | 126 | 90 | 129 | 120 | 100 | 112 | 119 |
| Nitrogen, g/kg | 31.5 | 35.5 | 28.0 | 25.8 | 28.2 | 29.8 | 33.6 | 23.2 | 23.5 | 34.6 |
| Carbon, g/kg | 486 | 474 | 508 | 486 | 506 | 484 | 489 | 500 | 493 | 489 |
| Ratio carbon/nitrogen | 15.4 | 13.4 | 18.1 | 18.9 | 18.0 | 16.3 | 14.6 | 21.6 | 21.0 | 14.1 |
| Hemicellulose, g/kg | 136 | 132 | 188 | 170 | 171 | 203 | 169 | 177 | 169 | 155 |
| Acid detergent lignin, g/kg | 46 | 20 | 61 | 41 | 44 | 37 | 36 | 51 | 43 | 37 |
| Biomethane potential, L/kg | 344 | 385 | 318 | 350 | 345 | 359 | 364 | 327 | 340 | 363 |

According to Petcu et al. (2022) winter peas plants had C/N=9.24-12.63, but mixtures of winter peas and triticale showed values of C/N=9.69-11.95. In our previous research (Ţîţei and Cozari, 2023), the biochemical methane potential of fresh mass substrate reached 343 L/kg in *Vicia sativa*, and 343 L/kg in *Lathyrus sativus.* Hunady et al. (2021) reported that the theoretical methane yield of biomass from *Galega orientalis*, *Lathyrus pratensis*, *Trigonella foenum-graecum* and *Melilotus alba* ranged from 0.161 to 0.172 $m^3$/kg VS.

## CONCLUSIONS

The annual leguminous species studied - *Cicer arietinum*, *Glycine max*, *Lablab purpureus*, *Pisum arvense*, *Trifolium alexandrinum* and *Trigonella foenum-graecum* - can be effectively incorporated into livestock feeding systems as fresh natural forage (fresh mass), as well as conserved fodder in the form of hay and fermented biomass (silage).

In addition to their role in animal nutrition, both fresh and fermented biomass of these species can also be utilized as co-substrates in anaerobic digestion systems, contributing to biomethane production and renewable energy generation.

Besides, due to their biological nitrogen fixation capacity and positive impact on soil fertility, these species are great preceding to be included in crop rotation systems, also intercropping with other non-legumes and as intermediate and cover crops.

Furthermore, the evaluated genotypes serve as valuable genetic resources for breeding programs focused on developing and introducing new cultivars for forage production and feedstock to be used in bioeconomy.

## ACKNOWLEDGEMENTS

This study has been supported by the subprogram no. 01.01.02 *"Identification of valuable forms of plant resources with multiple uses for the circular economy"*.

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