

THE BIOCHEMICAL COMPOSITION OF BIOMASS FROM INTERMEDIATE WHEATGRASS (*Agropyron intermedium*) AND POTENTIAL USES IN MOLDOVA

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Abstract

This research was conducted to determine biomass performances of intermediate wheatgrass (*Agropyron intermedium*) and its potential uses. We found that the dry matter content, biochemical composition and nutritive value of intermediate wheatgrass whole plants was characterized by the following indices: 13.1% CP, 39.0% CF, 67.2% NDF, 40.7% ADF, 3.6% ADL, 7.5% TSS, 9.1% ash with nutritive energy values 57.2% DMD, RFV = 79, 11.37MJ/kg DE, 9.34MJ/kg ME and 5.55MJ/kg NEL. The dry matter of prepared hay from intermediate wheatgrass contained 122g/kg CP, 94 g/kg ash, 398 g/kg CF, 412 g/kg ADF, 682g/kg NDF, 37 g/kg ADL with 55.8% DMD, 11.30 MJ/kg DE, 9.28 MJ/kg ME, 5.30 MJ/kg NEL. The obtained silage is characterized by pleasant smell and dark green colour, the nutrient content and feed energy values of dry matter were: 144 g/kg CP, 120g/kg ash, 395g/kg CF, 399 g/kg ADF, 654 g/kg NDF, 21 g/kg ADL, 57.8% DMD, 11.48MJ/kg DE, 9.43MJ/kg ME, 5.45MJ/kg NEL. The biochemical methane potential of the intermediate wheatgrass substrates ranged from 320 to 333 L/kg volatile solids.

Key words: *Agropyron intermedium*, biochemical composition, biomethane potential, green mass, hay, nutritive value, silage.

INTRODUCTION

The Poaceae family (grasses) comprises over 10,000 annual and perennial species, predominantly herbaceous, and largely distributed across tropical and warm-temperate regions. Numerous species within this family hold significant agricultural value, serving as sources of food, forage, and ornamental plants. Perennial grasses are hypothesized to provide a broader spectrum of supporting and regulating ecosystem services compared to annual species, including reduced soil disturbance, enhanced carbon sequestration over time, and the maintenance of biodiversity.

Intermediate wheatgrass, *Agropyron intermedium* (Host) Beauv. (syn. *Elymus hispidus* (Opiz) Melderis; *Elytrigia intermedia* (Host) Nevski; *Thinopyrum intermedium* [Host] Barkworth & D.R. Dewey; *Thinopyrum intermedium* spp. *barbulatum*), is a long-lived C₃ perennial grass native to Central Europe, the Balkans, and Asia Minor. It has been extensively introduced and cultivated as a forage species in the USA, Canada, Australia, and South America. The species exhibits weakly to

strongly creeping rootstocks that form a dense sod under moist conditions, with generally short rhizomes, although these traits vary among ecotypes and cultivars. The stems are robust, erect, and typically reach 90-150 cm in height. Foliage coloration ranges from bright green to dull blue-green, while leaf margins, inflorescences, and seeds can be glabrous or pubescent. The inflorescence is a narrow spike, 15-25 cm in length, with spikelets arranged alternately along the rachis. Each spikelet contains 2-6 florets, with glumes that are pointed and approximately half the length of the spikelet. Seeds mature by August, with an estimated 194,000 seeds per kilogram. Seedling vigor is rated from good to excellent, and establishment occurs relatively quickly compared to other meadow grasses. The species performs poorly in wet, poorly drained, or moderately saline to alkaline soils. It exhibits moderate cold tolerance, can withstand occasional spring flooding, and is highly fire-tolerant. The pubescent form is more resilient to low-fertility, alkaline soils, higher elevations, and drier conditions than the intermediate form. Intermediate wheatgrass is well-suited for the

stabilization of disturbed soils and can be employed in urban and critical environments with limited irrigation, including ditch banks, dikes, and roadsides.

Recently, it has been proposed as a dual-purpose perennial cereal, capable of providing both forage and grains for human consumption. Its integration into agricultural systems offers environmental benefits such as soil protection and water-quality regulation, attributable to its permanent soil cover and extensive belowground biomass. While domestication is at an early stage and grain yield remains low compared to annual cereals, the seeds can be utilized in baked products or beverages, potentially supplementing or substituting wheat, rye, or barley. Forage production presents an additional avenue for agricultural valorization. The species is compatible with other crops, notably alfalfa (*Medicago* spp.), with mixed stands often enhancing both longevity and productivity.

Intermediate wheatgrass has been the subject of extensive study and breeding across diverse geographical regions. New cultivars are being developed for multiple applications, including grain production, grazing pasture, hay, and silage (Wills et al., 1998; Ogle et al., 2011; Hybner & Jacobs, 2012; Culman et al., 2013, 2023; Jungers et al., 2017, 2018, 2022; De Oliveira et al., 2020; Lanker et al., 2020; Bowden, 2023; Bugayov et al., 2023; Locatelli et al., 2023; Rusch et al., 2025; Fagnant, 2024; Pan et al., 2025; Gutknecht et al., 2026; Pinto et al., 2026).

This research was conducted to evaluate the biomass performance of intermediate wheatgrass (*Agropyron intermedium*) and to assess its potential applications as livestock fodder, as well as a feedstock for biogas reactors in the production of biomethane as a renewable energy source.

MATERIALS AND METHODS

The study focused on a local ecotype of intermediate wheatgrass (*Agropyron intermedium* (Host) P. Beauv.) cultivated in a pure stand on the experimental plot of the “Alexandru Ciubotaru” National Botanical Garden (Institute), Moldova State University, Chişinău. Local ecotypes of tall oatgrass

(*Arrhenatherum elatius*), tall fescue (*Festuca arundinacea*), and meadow fescue (*Festuca pratensis*) were used as control species. Samples were collected at the first cut, consisting of whole plants harvested manually. The freshly cut biomass was chopped into pieces of approximately 1.5-2.0 cm using a laboratory forage chopper. The dry matter content was determined by drying the samples at 105°C until constant weight was achieved.

Silage was prepared from the chopped green biomass, which was compacted in tightly sealed glass containers and stored at ambient temperature (18-20°C). After 45 days of fermentation, the containers were opened and the sensory characteristics and fermentation parameters of the silage were evaluated according to standard laboratory procedures outlined in the Moldovan standard SM 108. Hay samples were obtained by drying the biomass directly in the field.

For biochemical analyses, plant samples were dried in a forced-air oven at 60°C and ground in a beater mill fitted with a 1 mm sieve. The main biochemical parameters - crude protein (CP), ash, acid detergent fibre (ADF), neutral detergent fibre (NDF), acid detergent lignin (ADL), and total soluble sugars (TSS) - were determined using near-infrared spectroscopy (NIRS) with a PERTEN DA 7200 analyser at the Research and Development Institute for Grasslands, Braşov, Romania. 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 using standard equations.

The carbon content of the substrates was estimated using the empirical equation proposed by Badger et al. (1979), while the biochemical methane potential was calculated according to the equations described by Dandikas et al. (2015).

RESULTS AND DISCUSSIONS

The analysis of biological characteristics of the studied grass species (Table 1) indicated that at the time of the first cut, the tested local ecotype of *Agropyron intermedium* reached a height of 128 cm, whereas the control grass species, ranged from 89 to 114 cm. The fresh biomass

yield of the local ecotype of *Agropyron intermedium* was 4.99 kg/m², corresponding to 1.15 kg/m² of dry matter. In comparison, the control species produced the following yields: *Arrhenatherum elatius* - 4.18 kg/m² fresh mass (1.01 kg/m² dry matter), *Festuca arundinacea* - 4.25 kg/m² fresh mass (1.02 kg/m² dry matter),

and *Festuca pratensis* - 3.89 kg/m² fresh mass (1.04 kg/m² dry matter). The proportion of leaves and inflorescences in the fresh biomass of *Agropyron intermedium* was higher than that in *Arrhenatherum elatius*, but lower than in the *Festuca* species.

Table 1. Some agrobiological peculiarities of the studied grass species

Species	Plant height, cm	Yield, kg/m ²		Content of leaves and inflorescences in fodder, %
		fresh mass	dry matter	
<i>Agropyron intermedium</i>	128	4.99	1.15	60.00
<i>Arrhenatherum elatius</i>	114	4.18	1.01	58.76
<i>Festuca arundinacea</i>	109	4.25	1.02	62.20
<i>Festuca pratensis</i>	89	3.89	1.04	67.00

Previous studies support these observations. Culman et al. (2013) reported vegetative biomass yields of intermediate wheatgrass ranging from 12,202 kg/ha under organic management to 13,083-17,131 kg/ha under conventional management. Sakiroglu et al. (2020) mentioned that, in Minnesota, summer aboveground biomass from dual-use Kernza intermediate wheatgrass in the third growing year averaged 1.06 kg/m². Pototskaya et al. (2022) observed that the wheatgrass cultivar ‘Sova’ produced 21.0 t/ha of green mass or 7.1 t/ha of hay. Bugayov et al. (2024) reported that fodder productivity of the cultivar ‘Kernza’ of intermediate wheatgrass across two cuts ranged from 1.49 to 2.65 kg/m² dry matter, while the cultivar ‘Horse’ ranged from 1.18 to 1.79 kg/m² dry matter. The quality indices of the harvested biomass from the studied grass species are presented in Table 2. The comparative analysis of the

biochemical composition of the dry matter indicated that the green forage of *Agropyron intermedium* contained higher crude protein (131 g/kg) and mineral content (91 g/kg) than *Arrhenatherum elatius* (80 g/kg and 83 g/kg, respectively) and the *Festuca* species (107-124 g/kg protein and 83-89 g/kg minerals). Neutral detergent fibre (NDF) and acid detergent lignin (ADL) concentrations in *Agropyron intermedium* were lower than in *Arrhenatherum elatius* but higher than in the *Festuca* species. The total soluble sugar content in *Agropyron intermedium* forage was lower than in the control grass species. The cellulose content in *Agropyron intermedium* was comparable to that of *Arrhenatherum elatius* but exceeded that of the *Festuca* species. Hemicellulose levels in *Agropyron intermedium* were higher than in *Festuca pratensis* but lower than in *Arrhenatherum elatius* and *Festuca arundinacea*.

Table 2. The biochemical composition and the nutritive value of the green mass from studied grass species

Indices	<i>Agropyron intermedium</i>	<i>Arrhenatherum elatius</i>	<i>Festuca arundinacea</i>	<i>Festuca pratensis</i>
Crude protein, g/kg DM	131	80	107	124
Crude fibre, g/kg DM	390	382	354	315
Minerals, g/kg DM	91	83	89	83
Acid detergent fibre, g/kg DM	407	411	388	339
Neutral detergent fibre, g/kg DM	672	710	666	574
Acid detergent lignin, g/kg DM	36	38	34	30
Cellulose, g/kg DM	371	373	354	309
Hemicellulose, g/kg DM	265	299	278	235
Total soluble sugars	75	118	122	108
Digestible dry matter, g/kg DM	572	561	587	625
Relative feed value	79	74	87	110
Digestible energy, MJ/ kg DM	11.37	11.18	11.64	12.20
Metabolizable energy, MJ/ kg DM	9.34	9.18	9.56	10.01
Net energy for lactation, MJ/ kg DM	5.55	5.31	5.57	6.13

Regarding feed value, *Agropyron intermedium* forage exhibited favourable dry matter digestibility, relative feed value (RFV), and energy concentration compared to *Arrhenatherum elatius*, although these values remained lower than those observed in the *Festuca* species.

The nutrient composition of biomass harvested from *Agropyron* species has been extensively documented in the literature. Wills et al. (1998) reported that intermediate wheatgrass forage contained 2.7% nitrogen, 13.6% soluble carbohydrates, 3.6% lipids, 44.0% NDF, 24.8% ADF, and 683 g/kg organic matter digestibility (OMD), whereas tall oatgrass exhibited 1.8% nitrogen, 13.6% soluble carbohydrates, 2.6% lipids, 44.0% NDF, 28.6% ADF, and 709 g/kg OMD. Glover et al. (2004) found that intermediate wheatgrass grown on saline soils yielded 3,320 kg/ha dry matter, containing 10.4% crude protein (CP), 0.24% phosphorus (P), 55.4% NDF, and 29.8% ADF, while reed canary grass produced 11.0% CP, 0.25% P, 57.7% NDF, and 32.9% ADF. Zhao et al. (2008) reported crude protein concentrations of 135 g/kg dry matter (DM) in intermediate wheatgrass forage, compared with 111 g/kg DM in tall fescue and festulolium. Phillips et al. (2009) observed that forage from intermediate wheatgrass contained 349 g/kg DM, with 2.09% nitrogen, 66.94% NDF, and 31.72% ADF. Albayrak et al. (2011) documented that *Agropyron intermedium* forage contained 9.90-10.42% CP, 57.72-58.86% NDF, 41.88-44.29% ADF, 44.18-47.28% total digestible nutrients (TDN), and a relative feed value (RFV) of 86.00-92.51, whereas *Bromus inermis* contained 10.45-11.30% CP, 57.85-59.70% NDF, 41.75-43.50% ADF, 45.16-47.45% TDN, and RFV of 85.57-88.11. Jensen et al. (2017) reported that the nutritive value of intermediate wheatgrass at the late reproductive stage included 10.0% CP, 39.4% ADF, 63.6% NDF, and 630 g/kg neutral detergent fiber digestibility (NDFD). Newell & Hayes (2017) indicated that first-year intermediate wheatgrass forage contained 18.97% CP, 80.3% digestible dry matter (DMD), 12.3 MJ/kg metabolizable energy (ME), 32.0% NDF, 12.3% ADF, 21.2% water-soluble carbohydrates (WSC), and 5.0% ash. Favre et al. (2019) reported seasonal variation in forage quality, with spring forage containing

22.5% CP, 45.6% NDF, 24.9% ADF, and a relative forage quality (RFQ) of 175; summer forage showed 5.1% CP, 70.2% NDF, 42.7% ADF, RFQ = 65; and fall forage had 11.9% CP, 59.0% NDF, 33.7% ADF, RFQ = 116. Gürsoy et al. (2021) reported that green biomass of *Agropyron intermedium* contained 9.14% ash, 19.56% CP, 3.06% ether extract (EE), 20.88% crude fiber (CF), 57.82% NDF, 26.49% ADF, 5.45% acid detergent lignin (ADL), 71.40% TDN, 65.92% true dry matter digestibility (TDMD), 56.02% total NDF digestibility (TNDFD), 97.75% total organic matter digestibility (TOMD), RFQ = 138.29, 8.82 MJ/kg ME, and 5.42 MJ/kg net energy for lactation (NEI). By comparison, green mass from *Dactylis glomerata* contained 7.84% ash, 13.47% CP, 1.46% EE, 27.49% CF, 66.23% NDF, 34.14% ADF, 8.52% ADL, 68.79% TDN, 67.85% TDMD, 58.40% TNDFD, 97.70% TOMD, RFQ = 122.24, 6.85 MJ/kg ME, and 3.73 MJ/kg NEI. Pinto et al. (2022) observed that intermediate wheatgrass harvested during the vegetative period contained 17.3% CP, 55.1% NDF, 30.0% ADF, RFV = 111, whereas the forage collected during the reproductive stage of plants had 5.6% CP, 69.6% NDF, 42.6% ADF, RFV = 75. Puka-Beals et al. (2022) reported that first-cut forage at the boot stage contained 10.05% CP, 37.45% ADF, and 64.53% NDF, while the harvest at anthesis stage contained 63.2% CP, 43.27% ADF, 73.28% NDF, and at the dough stage contained 4.62% CP, 40.11% ADF, and 69.68% NDF. Bowden (2023) reported that non-fertilized intermediate wheatgrass forage contained 7.08-9.50% CP, RFV = 95-117, whereas manure-treated variants contained 9.49-11.20% CP, RFV = 93-120. Locatelli et al. (2023) documented that in Uruguay, winter-harvested intermediate wheatgrass contained 19.73% CP, 50.07% NDF, 30.47% ADF, RFV = 108.5, while summer-harvested plants contained 8.20% CP, 70.58% NDF, 42.99% ADF, RFV = 73.2. Yavuz and Kır (2023) observed that intermediate wheatgrass forage contained 11.5-12.0% CP, 54.2-57.4% NDF, 41.2-43.5% ADF, 6.1-7.6% ADL, whereas smooth brome forage contained 12.3-12.5% CP, 53-56% NDF, 39.0-42.6% ADF, 5.3-6.5% ADL. Fagnant et al. (2024) highlighted that forage quality of intermediate wheatgrass varied with phenological stage, with ash content

ranging 3.1-13.3%, CP 28.0-31.3%, NDF 27.0-79.9%, ADF 16.4-51.4%, ADL 1.1-11.2%, cellulose 14.4-52.8%, and OMD 280-930 g/kg. Culman et al. (2023) reported that spring-harvested intermediate wheatgrass contained 15.1-21.1% CP, 51.2-55.7% NDF, 26.1-28.7% ADF, 9.6-10.6% ash, and RFV = 112-126.8, whereas summer-harvested forage contained 4.1-4.8% CP, 67.2-70.2% NDF, 38.2-42.6% ADF, 6.0-6.5% ash, and RFV = 75.0-80.9. Fagnant (2024) reported that the forage quality of intermediate wheatgrass at the first harvest in early May was characterized by 18.2% CP, 25.5% Cel, 51.9% NDF, 26.2% ADF, and 2.2% ADL, with an OMD of 785 g/kg and a NEL of 1524 kcal/kg. When harvested later, at the end of May, forage quality declined markedly, with CP decreasing to 9.8%, while fiber fractions increased to 34.7% Cel, 61.7% NDF, 32.5% ADF, and 3.2% ADL. At the same time, OMD decreased to 654 g/kg and NEL to 1281 kcal/kg. Similarly, Pan et al. (2025) reported that the

chemical composition of fresh intermediate wheatgrass biomass included 264.3 g/kg DM, 14.31% CP, 61.81% NDF, 31.79% ADF, 14.17% WSC, 3.58% EE, and 8.91% ash. Pinto et al. (2026) observed seasonal variation in forage quality, reporting CP concentrations of 10.6-21.6%, NDF of 45.3-67.7%, and ADF of 24.9-37.2% during the spring period. Hay represents the principal dried forage source for ruminants (cattle, sheep, and goats), horses, and small herbivores such as rabbits, providing essential structural fiber necessary for maintaining digestive health. It is a fundamental component of livestock diets, particularly during winter or periods, when grazing is limited. The nutritional value of hay depends primarily on plant species, stage of maturity at harvest, and the effectiveness of the drying process. The biochemical composition and nutritive value of hay produced from the studied grass species are presented in Table 3.

Table 3. The biochemical composition and the nutritive value of the hay from studied grass species

Indices	<i>Agropyron intermedium</i>	<i>Arrhenatherum elatus</i>	<i>Festuca arundinacea</i>	<i>Festuca pratensis</i>
Crude protein, g/kg DM	122	77	100	122
Crude fibre, g/kg DM	398	414	393	350
Minerals, g/kg DM	94	80	89	80
Acid detergent fibre, g/kg DM	412	436	427	385
Neutral detergent fibre, g/kg DM	682	740	720	621
Acid detergent lignin, g/kg DM	37	40	35	34
Cellulose, g/kg DM	375	396	372	351
Hemicellulose, g/kg DM	270	304	292	236
Total soluble sugars	49	98	55	102
Digestible dry matter, g/kg DM	558	541	556	589
Relative feed value	76	68	74	93
Digestible energy, MJ/ kg DM	11.30	10.82	11.09	11.68
Metabolizable energy, MJ/ kg DM	9.28	8.88	9.10	9.59
Net energy for lactation, MJ/ kg DM	5.30	5.02	5.13	5.61

The hay prepared from *Agropyron intermedium* contained 122 g/kg CP, 94 g/kg ash, 398 g/kg CF, 412 g/kg ADF, 682 g/kg NDF, 37 g/kg ADL, 375 g/kg cellulose, 270 g/kg hemicellulose, and 49 g/kg TSS. The nutritive and energy value of this hay was characterized by a DMD of 55.8%, DE of 11.30 MJ/kg, ME of 9.28 MJ/kg, and NEL of 5.30 MJ/kg. During the haymaking process, the concentration of structural carbohydrates increased, whereas the content of total soluble sugars decreased in hay obtained from all investigated species. Compared with the control grass species, *Agropyron intermedium* hay exhibited a crude

protein concentration similar to that of *Festuca pratensis* hay, but considerably higher than that of *Arrhenatherum elatus* and *Festuca arundinacea* hay. The hay from *Agropyron intermedium* contained lower levels of total soluble sugars and minerals. The concentration of cell wall fractions in *Agropyron intermedium* hay was lower than that observed in *Arrhenatherum elatus* hay, but higher than in *Festuca pratensis* hay. Furthermore, the dry matter digestibility, relative feed value, and energy concentration of *Agropyron intermedium* hay were more favorable compared with those

of *Arrhenatherum elatius* and *Festuca arundinacea* hay.

Several studies have reported the nutritive characteristics and quality indices of hay derived from *Agropyron* species. Moore et al. (1995) indicated that intermediate wheatgrass hay harvested during the spring vegetative stage contained approximately 20% CP, 50.0% NDF, and exhibited a DMD ranging from 700 to 800 g/kg. Maheri-Sis et al. (2008) evaluated the chemical composition of hay obtained from *Agropyron repens* (quackgrass) harvested at a late stage of maturity and reported values of 950 g/kg DM and 887 g/kg OM. The hay contained 8.9% CP, 1.44% EE, 34.30% CF, 66.0% NFE, 8.96% NFC, 69.60% NDF, 38.30% ADF, 5.70% ADL, and 11.2% ash. In addition, the authors reported an OMD of 43.54% and a ME concentration of 6.58 MJ/kg. Similarly, Phillips et al. (2009) reported that baled intermediate wheatgrass hay contained 2.08% N, 67.21% NDF and 32.42% ADF. More recent studies by Hunter et al. (2020) highlighted the strong seasonal variability in the nutritive value of intermediate wheatgrass hay. Spring-harvested hay exhibited high protein concentrations (22.0-28.0% CP) and a relative feed value (RFV) of 148-161, whereas hay harvested in autumn showed a more reduced protein content (12.8-13.2% CP) and lower RFV values (89-107). Miron et al. (2023) investigated the nutritive composition of hay obtained from *Elytrigia repens* (syn. *Agropyron repens*) grasslands and reported CP concentrations ranging from 6.62 to 11.79%, EE from 1.31 to 2.47%, CF from 30.78 to 37.17%, and NFE from 40.64 to 53.01%. The hay also contained 4.31-6.82% soluble sugars, 1.40-1.79% starch, and 7.12-7.69% ash. Mineral concentrations ranged from 0.27 to 0.49% Ca and 0.16 to 0.21% P. Energy values were reported as 18.22-19.94 MJ/kg GE, 8.05-9.73 MJ/kg ME, and 4.39-5.50 MJ/kg NEI. More recently, Nazare et al. (2026) reported that the hay produced from *Elytrigia repens* grasslands contained 8.5% CP, 42.4% CF, 75.3% NDF, 45.0% ADF, and 4.4% ADL, with structural carbohydrates composed of 41.6% cellulose and 30.3% hemicellulose. The hay also contained 8.6% ash and demonstrated a dry matter digestibility of 54.3%, with energy concentrations of 8.84 MJ/kg ME and 4.86 MJ/kg NEI.

Silage production represents an important preservation strategy that stabilizes the nutritional value of fresh forage while ensuring a continuous supply of high-quality feed for ruminants throughout the year. By maintaining the biochemical integrity of harvested biomass through controlled anaerobic fermentation, silage helps bridge seasonal forage shortages and prevents fluctuations in feed quality.

The fermentation characteristics, biochemical composition, and nutritional value of silage produced from the investigated grass species are presented in Table 4. The silage prepared from *Agropyron intermedium* was characterized by a pleasant smell and dark-green color, with a pH of 3.97 and a total organic acid concentration of 37.1 g/kg, including 33.8 g/kg lactic acid. Butyric acid was not detected, indicating a well-preserved fermentation process. In addition, the concentration of acetic acid in *Agropyron intermedium* silage was lower than that observed in the control silages. The nutrient composition and energy value of *Agropyron intermedium* silage dry matter were as follows: 144 g/kg CP, 120 g/kg ash, 395 g/kg CF, 399 g/kg ADF, 654 g/kg NDF, and 21 g/kg ADL. The digestibility and energy indices were 57.8% DMD, 11.48 MJ/kg DE, 9.43 MJ/kg ME, and 5.45 MJ/kg NEI. During the ensiling process, the concentration of ADL decreased. Compared with the initial green biomass, the silage exhibited increased concentrations of crude protein and minerals, while the contents of crude fiber, ADF, and ADL decreased, contributing to improved digestibility and higher energy value of the resulting forage.

In comparison with silages obtained from the control grass species, *Agropyron intermedium* silage was distinguished by a higher crude protein concentration. Moreover, the dry matter digestibility and energy content of this silage were superior to those recorded for silages produced from *Arrhenatherum elatius* and *Festuca arundinacea*.

Similar findings were reported by Pan et al. (2025), who described intermediate wheatgrass silage with a pH of 4.90, containing 4.46% lactic acid and 0.68% acetic acid. The silage had a dry matter content of 289.9 g/kg and included 13.43% CP, 45.20% NDF, 30.20% ADF, 8.61% WSC, 1.68% EE, and 10.56% ash.

Table 4. The quality indices of the silage from studied grass species

Indices	<i>Agropyron intermedium</i>	<i>Arrhenatherum elatius</i>	<i>Festuca arundinacea</i>	<i>Festuca pratensis</i>
pH index	3.97	4.26	4.05	4.11
Organic acids, g/kg DM	37.1	24.2	36.7	39.6
Total acetic acid, g/kg DM	3.3	5.9	6.7	10.0
Total butyric acid, g/kg DM	0	0	1.3	0
Total lactic acid, g/kg DM	33.8	18.3	28.7	29.6
Acetic acid, % of organic acids	8.9	24.4	18.3	25.3
Butyric acid, % of organic acids	0	0	3.5	0
Lactic acid, % of organic acids	91.1	75.6	78.2	74.7
Crude protein, g/kg DM	144	88	106	130
Crude fibre, g/kg DM	395	405	380	320
Minerals, g/kg DM	120	100	105	91
Acid detergent fibre, g/kg DM	399	420	400	339
Neutral detergent fibre, g/kg DM	654	724	690	562
Acid detergent lignin, g/kg DM	21	26	19	24
Cellulose, g/kg DM	378	394	381	315
Hemicellulose, g/kg DM	255	304	290	223
Digestible dry matter, g/kg DM	578	565	577	625
Relative feed value	82	73	82	112
Digestible energy, MJ/ kg DM	11.48	11.25	11.46	12.32
Metabolizable energy, MJ/ kg DM	9.43	9.19	9.41	10.12
Net energy for lactation, MJ/ kg DM	5.45	5.21	5.43	6.13

Perennial species of the Poaceae family are increasingly recognized as valuable feedstock for renewable energy production due to their high biomass productivity, relatively low input requirements, and adaptability to marginal lands. These grass species can be utilized for the production of biogas, solid biofuels, and bioethanol, thereby contributing to the development of sustainable bioenergy systems and reducing greenhouse gas emissions (Dandikas et al., 2015; Muntean et al., 2018; Țiței & Roșca, 2021; Cozari et al., 2023; Țiței, 2024).

Efficient anaerobic digestion requires an optimal carbon-to-nitrogen (C/N) ratio, generally ranging from 20:1 to 30:1. Ratios exceeding 30:1 may slow microbial degradation due to nitrogen limitation, whereas ratios below 10:1 can lead to ammonia accumulation, reduced pH, and inhibition of methanogenic activity. A balanced ratio of approximately 25:1 is therefore considered optimal for maximizing methane production efficiency. The quality indices of the prepared substrates from the studied grass species and their biochemical methane production potential are presented in Table 5. The C/N ratio of both green biomass and silage substrates from *Agropyron intermedium* was within a favorable range for anaerobic digestion. Hemicellulose content in these substrates varied between 255 and

265 g/kg, while acid detergent lignin ranged from 21 to 36 g/kg. The biochemical methane potential ranged from 352 to 378 L/kg volatile solids (VS). The highest methane yield was obtained from the silage substrate, which also exceeded the methane production potential of silage derived from *Festuca pratensis*.

Previous studies have reported comparable methane production potentials for biomass from *Agropyron* species. Dickeduisberg (2017) reported methane yields ranging from 204 to 282 L/kg for substrates derived from tall wheatgrass herbage. In earlier investigations, Țiței (2021) reported biochemical methane potentials of 328 L/kg VS for green biomass substrates from both *Agropyron cristatum* and *Agropyron desertorum*. Lalak et al. (2022) noted that substrates derived from *Agropyron elongatum* contained 46.24% cellulose, 28.51% hemicellulose, and 28.27% lignin, achieving a biogas production potential of 398 L/kg in treated substrates. Miron et al. (2023) reported that the specific methane yield of biomass substrates from *Elytrigia repens* (syn. *Agropyron repens*) grasslands ranged between 242 and 282 L/kg VS. Similarly, Nazare et al. (2026) indicated that substrates derived from *Elytrigia repens* grasslands had a C/N ratio of 37.34, 4.4% ADL, and 30.3% hemicellulose, with a biomethane potential reaching 333 L/kg VS.

Table 5. The biochemical biomethane production potential of the investigated substrates from studied grass species

	<i>Agropyron intermedium</i>		<i>Arrhenatherum elatius</i>		<i>Festuca arundinacea</i>		<i>Festuca pratensis</i>	
	green mass	silage	green mass	silage	green mass	silage	green mass	silage
Crude protein, g/kg DM	131.00	144.00	80.00	88.00	107.00	106.00	124.00	130.00
Nitrogen, g/kg DM	20.96	23.04	12.80	14.10	17.12	16.96	19.84	20.80
Ash, g/kg DM	91.00	120.00	83.00	100.00	89.00	105.00	83.00	91.00
Carbon, g/kg DM	505.00	488.89	509.00	500.00	506.11	497.22	509.44	505.00
Ratio carbon/nitrogen	24.09	21.22	39.80	35.50	29.42	29.32	25.68	24.28
Acid detergent lignin, g/kg DM	36.00	21.00	38.00	26.00	34.00	19.00	30.00	24.00
Hemicellulose, g/kg DM	265.00	255.00	299.00	304.00	278.00	290.00	235.00	223.00
Biomethane potential, L/kg VS	352.00	378.00	341.00	361.00	351.00	376.00	360.00	369.00
Biomethane potential, L/kg DM	320.00	333.00	313.00	325.00	320.00	337.00	330.00	336.00

CONCLUSIONS

The local ecotype of intermediate wheatgrass (*Agropyron intermedium*) is well-suited for establishing temporary pastures, either in pure stands or in mixtures with other grasses and legumes, as well as for the restoration of eroded lands and the reintegration of degraded soils into agricultural use.

The harvested biomass of intermediate wheatgrass can be utilized as fresh forage, hay, or silage for livestock, and it also represents a viable feedstock for biogas production, contributing to biomethane generation as a renewable energy source.

Moreover, the evaluated local ecotype of *Agropyron intermedium* represents a valuable genetic resource for breeding programs aimed at developing new cultivars for forage production and other applications 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".

REFERENCES

- Albayrak, S., Mevlüt, T., Yüksel, O., Yilmaz, M. (2011). Forage yield and the quality of perennial legume-grass mixtures under rainfed conditions. *Notulae Botanicae Horti Agrobotanici Cluj-Napoca*, 39(1):114-118.
- Bowden, J.H. (2023). *Organically managed intermediate wheatgrass (Thinopyrum intermedium) as a dual-use grain and forage crop*. Master thesis. University of Minnesota, 71p.
- Bugayov, V.D., Bugayov, V.V., Veklenko, Y.A., Voronetska, I.S. (2023). Potential of fodder and seed productivity of intermediate wheatgrass (*Thinopyrum intermedium*) Kernza by years of grassland use. *Feeds and Feed Production*, 96:30-39 [In Ukrainian].
- Badger, C.M., Bogue, M.J., Stewart, J. (1979). Biogas production from crops and organic wastes. *New Zeland Journal of Science*, 22:11-20.
- Glover, D.E., Kielly, G.A., Jefferson, P.G., Cohen, R.D.H. (2004). Agronomic characteristics and nutritive value of 11 grasses grown with irrigation on a saline soil in southwestern Saskatchewan. *Canadian Journal of Plant Science*, 84:1037-1050.
- Cozari, S., Țiței, V., Ababii, A., Doroftei, V., Guțu, A., Cîrlig, N., Andreoiu, A., Blaj, V., Mazăre, V., Lisii, R., Covalciuc, D., Gadibadi, M., Cerempei, V., (2023). The evaluation of the biomass quality of *Spartina pectinata*, and prospects of its use in Moldova. *Scientific Papers. Series A. Agronomy*, 66(1): 677-684.
- Culman, S., Pinto, P., Pugliese, J., Crews, T., DeHaan, L., Jungers, J., Larsen, J., Ryan, M., Schipanski, M., Sulc, M., Wayman, S., Wiedenhoef, M., Stoltenber, D., Picasso, V. (2023). Forage harvest management impacts "Kernza" intermediate wheatgrass productivity across North America. *Agronomy Journal*, 115 :2424-2438.
- Culman, S.W., Snapp, S.S., Ollenburge, M., Basso B., DeHaan, L.R. (2013). Soil and water quality rapidly responds to the perennial grain Kernza wheatgrass. *Agronomy Journal*, 105(3): 735-744.
- Dandikas, V., Heuwinkel, H., Lichti, F., Drewes, J.E., Koch, K. (2015) Correlation between biogas yield and chemical composition of grassland plant species. *Energy Fuels*, 29 (11): 7221-7229.
- Dickeduisberg, M. (2017). *Management and drought effects on growth and herbage yield of tall wheatgrass (Agropyron elongatum) for biogas production in Central Europe*. Dissertation zur Erlangung des Doktorgrades. Göttingen, 114p.
- DeHaan, L.R., Wang, S., Larson, S.R., Cattani, D.J., Zhang, X., Kantarski, T. (2014). Current efforts to develop perennial wheat and domesticate *Thinopyrum intermedium* as a perennial grain. *Perennial Crops for Food Security Proceedings of the FAO Expert Workshop*, 72-89.
- De Oliveira, G., Brunzell, N.A., Crews, T. E., DeHaan, L.R., Vico, G. (2020). Carbon and water relations in perennial Kernza (*Thinopyrum intermedium*): an overview. *Plant Science*, 295 :110279.

- Fagnant, L. (2024). *Evaluation et optimisation agronomique des services de production d'une céréale pérenne, Thinopyrum intermedium*. Dissertation originale présentée en vue de l'obtention du grade de docteur en sciences agronomiques et ingénierie biologique, 269. https://orbi.uliege.be/bitstream/2268/321338/1/Th%C3%A8se_L_Fagnant_compressed.pdf
- Fagnant, L., Olivier, D., Bindelle, J., Beckers, Y., Decruyenaere, V., Dumont, B. (2024). Forage potential of *Thinopyrum intermedium* through near-infrared spectrometry and grown in mixture with various legumes. *Grass and Forage Science*, 1-14 DOI: 10.1111/gfs.12684
- Favrea, J. R., Castiblanco, T.M., Combs, D.K., Wattiaux, M.A., Picasso, V.D. (2019). Forage nutritive value and predicted fiber digestibility of Kernza intermediate wheatgrass in monoculture and in mixture with red clover during the first production year. *Animal Feed Science and Technology*, 258:114298. [10.1016/j.anifeeds.2019.114298](https://doi.org/10.1016/j.anifeeds.2019.114298).
- Gürsoy, E., Kaya, A., Gül, M. (2021). Determining the nutrient content, energy, and *in vitro* true digestibility of some grass forage plants. *Emirates Journal of Food and Agriculture*, 33(5): 417-422.
- Gutknecht, J.L., Anderson, J.A., Annor, G., Bajgain, P., Crews, T.E., Cureton, C., DeHaan, L., Meier, E., Peters, T., Picasso, V., Reilly, E.C., Reser, A., Ritter, T., Krug, A.S., Tautges, N., Jungers, J.M. (2026). Development and adoption of Kernza a perennial grain crop for sustainable agriculture. *Plants, People, Planet*, 1-16. <https://doi.org/10.1002/ppp.3.70164>
- Hybner, R.M., Jacobs, J. (2012). Intermediate wheatgrass (*Thinopyrum intermedium* L.): An Introduced Conservation Grass for Use in Montana and Wyoming. *NRCS-Montana-Technical Note-Plant Materials-MT-80*. <https://www.nrcs.usda.gov/plantmaterials/mtpmctn11288.pdf>
- Hunter, M.C., Sheaffer, C.C., Culman, S.W., Lazarus, W.F., Jungers, J.M. (2020). Effects of defoliation and row spacing on intermediate wheatgrass II: Forage yield and economics. *Agronomy Journal*, 112(3): 1862-1880.
- Jungers, J.M., DeHaan, L.R., Betts, K.J., Sheaffer, C.C., Wyse, D.L. (2017). Intermediate wheatgrass grain and forage yield responses to nitrogen fertilization. *Agronomy Journal*, 109(2): 462-472.
- Jungers, J., Frahm, C., Tautges, N., Ehlke, N., Wells, M., Wyse, D., Sheaffer, C. (2018). Growth, development, and biomass partitioning of the perennial grain crop *Thinopyrum intermedium*. *Annals of Applied Biology*, 172:346-354.
- Jungers, J.M., Schiffner, S., Sheaffer, C., Ehlke, N.J., DeHaan, L., Torrión, J., Noland, R.L., Franco, J.G. (2022). Effects of seeding date on grain and biomass yield of intermediate wheatgrass. *Agronomy Journal*, 114 :2342-2351.
- Lalak, J., Kasprzycka, A., Martyniak, D., Tys, J. (2016). Effect of biological pretreatment of *Agropyron elongatum* 'BAMAR' on biogas production by anaerobic digestion. *Bioresource Technology*, 200:194-200. doi: 10.1016/j.biortech.2015.10.022.
- Lanker, M., Bell, M., Picasso, V.D. (2020). Farmer perspectives and experiences introducing the novel perennial grain Kernza intermediate wheatgrass in the US Midwest. *Renewable Agriculture and Food Systems*, 35(6):653-662.
- Law, E.P., Wayman, S., Pelzer, C.J., Culman, S.W., Gómez, M.I., Ditommaso, A., Ryan, M.R. (2022). Multi-Criteria assessment of the economic and environmental sustainability characteristics of intermediate wheatgrass grown as a dual-purpose grain and forage crop. *Sustainability* (Switzerland), 14(6), 3548. <https://doi.org/10.3390/SU14063548/S1>
- Li, W., Li, H., Zheng, Q. (2025). *Thinopyrum* species as a genetic resource: enhancing salt tolerance in wheat and forage crops for sustainable agriculture. *Frontiers in Plant Science*, 16:1728305. doi: 10.3389/fpls.2025.1728305
- Locatelli, A., Gutierrez, L., Duchon, O., Speranza, P.R., Risso, V.D.P., Wang, Z. (2023). Agronomic assessment of two populations of intermediate wheatgrass Kernza (*Thinopyrum intermedium*) in temperate South America. *Grassland Research*, 1-17.
- Maheri-Sis, N., Mirzaei-Aghsaghali, A., Safaei, A.R., Mirza-Aghazadeh, A., Bibalani, G.H. (2008). Quack grass (*Agropyron repens* L.): as ruminant feed. *Research Journal of Environmental Sciences*, 2: 228-233.
- Miron, A., Țiței, V., Ababii, A., Teleuță, A., Guțu, A., Talmaci, L., Coșman, V., Galupa, A., Cozari, S., Nazare, A.-I., Mardari, L., Lazu, S., Cirlig, N. (2023). Grasslands as fodder for animals and renewable source of energy biomass. *Scientific Papers, Series A, Agronomy*, 66 (1): 772-779.
- Moore, K.J., Vogel, K.P., Klopfenstein, T.J., Masters, R.A., Anderson, B.E. (1995). Evaluation of four intermediate wheatgrass populations under grazing. *Crops*, 87(10381): 744-747.
- Muntean, I., Țiței, V., Gudima, A., Armaș, A., Gadibadi, M. (2018). Biomass quality of some *Poaceae* species and possible use for renewable energy production in Moldova. *Scientific Papers. Series A. Agronomy*, 61(1):497-502
- Nazare, A.I., Țiței, V., Guțu, A., Samuil, C., Miron, A., Gadibadi, M., Andreoiu, A., Vintu, V. (2026). The biomass productivity and quality indices of some permanent grasslands. *Romanian Agricultural Research*, 43: 373-385.
- Newell, M.T., Hayes, R.C. (2017). An initial investigation of forage production and feed quality of perennial wheat derivatives. *Crop & Pasture Science*, 68(12): 1141-1148.
- Ogle, D.G., John, S.L., Tober, D., Jensen, K. (2011). Plant guide for Intermediate wheatgrass (*Thinopyrum intermedium*). USDA-Natural Resources Conservation Service, Idaho and North Dakota Plant Materials Centers. <https://www.nrcs.usda.gov/plantmaterials/idpmspg10369.pdf>
- Park, H.S., Choi, K.C., Yang, S.H., Jung, J.S., Lee, B.H. (2022). Evaluation of growth characteristics and yield potential of summer emergency forage crops. *Journal of The Korean Society of Grassland and Forage Science*, 42:26-31.

- Pan, Y., Zhang, G., Lu, Q., Valentin, P.R., Dong, C. (2023). The combination of *L. plantarum* and *B. coagulans* enhanced the microbial community assembly of *T. intermedium* silage. <http://dx.doi.org/10.2139/ssrn.4613152>
- Phillips, W.A., Northup, B. N., Venuto, B.C. (2009). Dry matter intake and digestion of perennial and annual cool-season grasses by sheep. *The Professional Animal Scientist*, 25(5):610-618.
- Pinto, P., Cartoni-Casamitjana, S., Stolenberg, D.E., Zimbric, J., Picasso, V.D. (2022). Intercropping legumes and intermediate wheatgrass increases forage yield, nutritive value, and profitability without reducing grain yields. *Frontiers in Sustainable Food Systems*, 6:977841. doi.org/10.3389/fsufs.2022.977841
- Pinto, P., Tautges, N.E., Jungers, J.M., Sheaffer, C.C., Hunter, M., Picasso, V.D. (2026). Fall grazing improves the performance of Kernza intermediate wheatgrass as a dual-purpose crop. *Agronomy Journal*, 118(1):e70299. doi: 10.1002/agj2.70299
- Pototskaya, I.V., Shamanin, V.P., Aydarov, A.N., Morgounov, A.I. (2022). The use of wheatgrass (*Thinopyrum intermedium*) in breeding. *Vavilov Journal of Genetics and Breeding*, 6(5):413-421
- Puka-Beals, J., Sheaffer, C. C., Jungers, J.M. (2022). Forage yield and profitability of grain-type intermediate wheatgrass under different harvest schedules. *Agrosystems, Geosciences and Environment*, 5:e20274. <https://doi.org/10.1002/agg2.20274>
- Rusch, H.L., Hunter, M.C., Kraus, A., Tautges, N.E., Jungers, J.M. (2025) Intermediate wheatgrass as a dual use crop for grain and grazing. *Frontiers in Agronomy*, 7:1534962. doi: 10.3389/fagro.2025.1534962.
- Stybayev, G., Zargar, M., Serekpayev, N., Zharlygassov, Z., Baitelenova, A., Nogaev, A., Mukhanov, N., Elsergani, M.I.M., Abdiec, A.A.A. (2023). Spring-planted cover crop impact on weed suppression, productivity, and feed quality of forage crops in Northern Kazakhstan. *Agronomy*, 13(5):1278.
- Sakiroglu, M., Dong, C., Hall, M.B., Jungers, J., Picasso, V. (2020). How does nitrogen and forage harvest affect belowground biomass and nonstructural carbohydrates in dual-use Kernza intermediate wheatgrass? *Crop Science*, 60:2562–2573.
- Țiței, V. (2024). The agro-economic value of common millet, *Panicum miliaceum*, under the conditions of the Republic of Moldova. *Scientific Papers. Series Management, Economic Engineering in Agriculture and Rural Development*, 24(4):845-852.
- Țiței, V., Andreoiu, A.C., Coșman, V., Coșman, S., Nazare, A., Stavarache, M. (2021). Biomass quality of *Agropyron cristatum* and *Agropyron desertorum* and possibilities for utilization [Calitatea biomasei de *Agropyron cristatum* și *Agropyron desertorum* și posibilități de valorificare] In. *Conservarea diversității biologice - o șansă pentru remedierea ecosistemelor*, Chișinău, 372-379 [In Romanian].
- Țiței, V., Roșca, I. (2021). Good practices for the use of degraded lands in the cultivation of crops with energy biomass potential. Chișinău, 80 p. <https://www.ucipifad.md/wpcontent/uploads/2018/12/Bunele-practici-de-utilizare-aterenurilor-degradate-%C3%AEn-cultivarea-culturilorcu-poten%C5%A3ial-de-biomas%C4%83-energetic%C4%83.pdf> [In Romanian].
- Wills, B.J., Douglas, G.B., McKenzie, J., Trainor, K.D., Foote, A.G. (1998). *Thinopyrum intermedium* (Host) Barkw. & Dewey – a review, and evaluation of intermediate and pubescent wheatgrass for dryland agriculture in New Zealand. *Proceedings of the New Zealand Grassland Association*, 60:233-241.
- Yavuz, T., Kir, H. (2023). Performances of some perennial legume and grass mixtures under rainfed conditions of a continental climate region. *Turkish Journal of Range and Forage Science*, 4(2): 73-84.
- Zhao, D., MacKown, C.T., Starks, P.J., Kindiger, B. K. (2008). Interspecies variation of forage nutritive value and nonstructural carbohydrates in perennial cool-season grasses. *Agronomy Journal*, 100(3): 837-844.