Methane yield of perennial grasses as affected by the chemical composition of their biomass

Kristina AMALEVICIUTE-VOLUNGE, Alvyra SLEPETIENE, Bronislava BUTKUTE

Lithuanian Research Centre for Agriculture and Forestry, Institute of Agriculture
Instituto 1, Akademija, Kėdainiai distr., Lithuania
E-mail: kristina.amaleviciute-volunge@lammc.lt

Abstract
The aim of the research work was to study the chemical properties of most cultivated perennial Fabaceae and Poaceae plants: common cocksfoot (Dactylis glomerata L.), reed canary grass (Phalaris arundinacea L.), tall fescue (Festuca arundinacea Schreb.), perennial ryegrass (Lolium perenne L.), common lucerne (Medicago sativa L.), common samfion (Onobrychis vicifolia Scop.) and switchgrass (Panicum virgatum L.), and to link them to methane (CH₄) and biogas yields under laboratory conditions. The results of chemical analyses showed that the biomass of legumes was most suitable for biogas production: high digestibility: 78.4–73.5% dry matter (DM) and low cellulose concentration 20.8–24.4% DM, were determined to be most suitable for anaerobic digestion. The biomass of L. perenne and F. arundinacea was best-suited for anaerobic digestion due to the highest content of water-soluble carbohydrates (WSC) 16.4–20.3% DM, and the lowest content of cellulose 27.3–28.9% DM and acid detergent fibre (ADF) 31.3–32.8% DM. The best yields of methane were obtained from O. vicifolia – 277.7 L CH₄ kg⁻¹, D. glomerata – 213.9 L CH₄ kg⁻¹ and L. perenne – 205.7 L CH₄ kg⁻¹. The correlations corroborated that the methane yield depended on the chemical composition of the biomass. Methane yield positively and significantly correlated with WSC – 0.761** and digestibility – 0.744** (P ≤ 0.01), and negatively correlated with cellulose – 0.793*** and ADF – 0.762***. The best results of specific methane yields were demonstrated by O. vicifolia (1453 m³ CH₄ ha⁻¹ DM), M. sativa (1326 m³ CH₄ ha⁻¹ DM) and L. perenne (1060 m³ CH₄ ha⁻¹ DM).

This study with seven species of perennial grasses could serve as a basis for more advanced experiments on how to choose grass species for the best methane yield.

Key words: anaerobic digestion, biomass, biogas, methane, perennial plants.

Introduction

Due to the growing demand for energy and concerns about the increasing greenhouse gas emissions, perennial grasses have attracted worldwide attention as renewable energy sources offering several advantages over annual crops, such as lower establishment costs, higher biomass productivity, improved soil health, increased water quality and reduced soil erosion (Nazli, Tansi, 2019). Europe is the world’s leading producer of biogas. Most biomethane production plants are in Germany (185), United Kingdom (80) and Sweden (61). In the other countries the biomethane production volumes are still marginal (EBA, 2018; Scarlat et al., 2018).

Perennial grasses are attractive sources of biomass for Northern Europe and North America, as they meet agronomic, environmental and societal requirements for successful deployment as energy grass crops (Price et al., 2004; Monti et al., 2009; Rösch et al., 2009; Allison et al., 2012). Perennial grasses will likely be a dominant feedstock for on-farm anaerobic digestion in Northeast Europe. Sustainable development is a current notion strictly related to the concept of a circular economy. In this respect, the production of renewable energy by the valorisation of wastes or by-products is considered as one of the most dominant future renewable energy sources (Appels et al., 2011; Coppolecchia et al., 2015; Chiumenti et al., 2018). The use of energy crops has increased in several countries, particularly in Germany and Austria, due to their exceptionally high methane yields which increase the profitability of biogas production. Co-digestion of various substrates also contributes significantly to the improvement of the digestion process, the improvement of biogas yield and biogas plant performance. However, the sustainability debated on the use of energy crops and their impact on land use changes and on food security has led to limitations for the use of energy crops used for biogas production in Germany, Austria and Denmark. Thus, it is expected that the use of energy crops and their potential in the future biogas production in the EU will be increasingly limited due to sustainability considerations and support directed only to the use of waste and residues. Alternatively, landscape grass, consisting of herbaceous plant composite, could represent one of the most promising feedstocks to improve the sustainability of the biogas sector (Scarlat et al., 2018).

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In Lithuania, particular attention has been paid to the productivivity of local and introduced plant species with high energy value and the ability to use them as a biofuel in the environment (Kazlauskas et al., 2014; Slepetiene et al., 2016). Native, naturally growing perennial grasses, such as Festuca arundinacea, Dactylis glomerata and Phalaris arundinacea, have already been studied in Lithuania (Tilvikiene et al., 2012; Nekrošius et al., 2014; Butkutė et al., 2014; Pociene, Kadziauskiené, 2016; Slepetiene et al., 2016; Tilvikiene et al., 2016) as well as Panicum virgatum, which is an introduced grass (Norkevičienė et al., 2016). Agricultural researchers also show interest in other perennial plants: Medicago sativa, Onobrychis vicifolia and Lolium perenne (Slepetys et al., 2012; Slepetiene et al., 2016; Kemesyte et al., 2017).

The stability and productivity of anaerobic digestion of biomass are mostly influenced by its chemical composition (Slepetiene et al., 2016; Tilvikiene et al., 2016). Higher concentrations of fibre components (cellulose, hemicellulose and lignin) are a disruptive factor in the process of methane production (Sepplé et al., 2009; Triolo et al., 2011; McEniry, O’Kiely, 2013; Yang et al., 2015; Tilvikiene et al., 2016). Each fibre component in biomass has different chemical bonds and is therefore interdependent (Klimiuk et al., 2010; Butkutė et al., 2014). Hemicellulose is most susceptible to anaerobic conversion or to pre-treatment effects, whereas cellulose is not readily degraded and needs more enzymes for its digestion (Yang et al., 2015). Therefore, cellulose, hemicellulose and lignin levels and their ratio are important in grass biomass composition (Kuprys-Caruk, et al., 2019).

Anaerobic digestion is a biological process, wherein diverse group of microorganisms decompose the complex organic matter in the absence of oxygen. Thermochemical conversion destroys every component in biomass converting it to carbon dioxide, carbon monoxide, hydrogen, methane, nitrogen oxides and water in various amounts. Methane fermentation technologies exhibit higher specificity: lignin is not converted, 34–92% of proteins are hydrolysed and fermented depending on various conditions, 70–95% of lipids, 65–70% of polymerised sugars and 95% of sugar oligomers are destroyed (Bertsen, Felby, 2012). High biomass yield and high specific methane yield are parameters important when choosing the most appropriate crops for biogas production (Kuprys-Caruk et al., 2019). However, there is a lack of knowledge on the use of fresh perennial herbaceous plants for methane production and their potential and the most efficient and rational to use as a renewable source.

The aim of the research was to study the chemical properties of perennial herbaceous plants and to link these properties with high productivity and biogas yields under laboratory conditions.

### Materials and methods

#### Experimental plots.

Seven common grass species: common cocksfoot (Dactylis glomerata L.), reed canary grass (Phalaris arundinacea L.), tall fescue (Festuca arundinacea Schreb.), perennial ryegrass (Lolium perenne L.), common lucerne (Medicago sativa L.), common sainfoin (Onobrychis vicifolia Seep.) and switchgrass (Panicum virgatum L.) were grown in the field plots (0.5 m$^2$) within three replicate blocks in 2018 at the Institute of Agriculture (55°23′49″ N, 23°51′40″ E), Lithuanian Research Centre for Agriculture and Forestry. Herbage samples were taken in May–June (1st cut – heading / inflorescence emergence, the first harvest year) of 2018 and were cut to 1 cm pieces and stored at −18°C temperature for biogas analyses. Biomass yield was measured gravimetrically.

#### Chemical analyses.

Chemical analyses were done at Chemical Research Laboratory of Institute of Agriculture, Lithuanian Research Centre for Agriculture and Forestry. For chemical analyses and biogas experiment, air-dried samples were fixed at 105 ± 5°C temperature for 20 min and dried at 65 ± 5°C temperature for 24 hours, and after that ground in a laboratory mill. Chemical composition of the plant samples was determined according to the standard methods: for ash and organic matter (OM) content the dried samples were incinerated at 550°C temperature. Before testing, the samples for biogas production and chemical composition were ground by an ultracentrifugal mill GM 2200 (Retsch, Germany) using 1 mm mesh size. Total organic carbon (C) content was determined by a spectrophotometric procedure at a wavelength of 590 nm using glucose as a standard after wet combustion according to Tyurin method modified by Nikitin (1999). Nitrogen (N) content was determined by the Kjeldahl method using a spectrophotometric procedure at the wavelength of 655 nm.

Neutral detergent fibre (NDF), acid detergent fibre (ADF) and acid detergent lignin (ADL) in plant biomass were determined using the NDF method according to Van Soest et al. (1991). NDF and ADF extraction was done on a fibre analyser ANKOM 220 (ANKOM Technology, USA) using F57 filter bags (25-µm porosity). Lignin was determined in beakers on the remaining material from the ADF procedure as a residue insoluble in sulfuric acid (72% w/w). Cellulose (Cel) was determined as the difference between ADF and ADL and hemicellulose (Hcel): Hcel = NDF − ADF. The concentrations of water-soluble carbohydrates (WSC) were determined by arc titration method. Digestion of plant biomass and crude solids (CP) was measured by a NIRS 6500 system (Foss, Denmark).

The feedstocks (biomass + inoculum) before anaerobic digestion were homogenised (TS, volatile solids (VS) and mineral solids (MS) by a gravimetric method. This method includes evaporation of the sample, drying of the residue at 105°C temperature to a constant weight, and then repeating the steps taking into account the dry residue. (N) content was determined by a spectrophotometer Lange DR3900 (Hach GmbH, Germany) (pH) was determined in 1 M KCl (soil to solution ratio 1:2.5, w/v) using a potentiometric method. Organic acid to alkalinity ratio (FOS:TAC) was measured by titration with sulfuric acid 0.1% solution.

#### Anaerobic digestion experiments.

The investigation of biogas production in this paper is based on the results of laboratory experiments conducted in the Chemical Research Laboratory of Institute of Agriculture, Lithuanian Research Centre for Agriculture and Forestry. The tests were conducted under the gas and pressure control system OxiTop (WTW, Germany).

Biogas production was continuously measured using a glass vessel of 250 mL volume. The manometric device consists of a glass vessel provided with a pressure transducer located in a measuring head. During anaerobic tests, the vessels were mixed by a magnetic stirrer. The pressure due to biogas accumulation in the headspace was automatically registered by the measuring heads. Each bottle was filled with 0.05 L of inoculum (pH 8.4, FOS:TAC 0.10, TS 2.29%, VS 46.87% TS, N 2.33 g L$^{-1}$ and COD 4.68 g L$^{-1}$) and 0.25 g OM of biomass, with a starting organic load 5 g L$^{-1}$ (Dudek et al., 2017; Zielinski et al., 2017). A sufficient headspace volume was provided not to exceed the maximum pressure value of 400 hPa. Before starting, the vessel headspace was flushed with N$_2$ for 120 s. The biogas production was accompanied by daily automatic measurements with an interval of 4 min. The maximum of each replicate and average gas production rates in hPa were extracted to evaluate the repeatability of the method, the tests were conducted in triplicate. OxiTop vessels were incubated at 37 ± 1°C temperature under mesophilic conditions for 35 days in a thermostat. The total biogas production was measured by a gas data analyser GFM406 (Gas Data, UK).

#### Calculations.

Biogas production was calculated by the formula:

$$n (\text{mol}) = \frac{p \times V}{R \times T},$$

where $p$ is headspace pressure (hPa), $V$ – bottle volume (ml), $R$ – ideal gas constant (8.314 J / (mol × K)), $T$ – incubation temperature (K).

Biogas volume was calculated by the formula:

$$L\text{ biogas} = n (\text{mol}) \times 22.4;$$

under normal conditions $(t = 0°C; P = 1.013 \times 10^5 \text{ Pa})$ one mol of gas takes 22.4 L.
The specific methane (CH₄) yield was calculated as the cumulative sum of the methane volume produced over a 35-day incubation period relative to the substrate by dry matter (DM) concentration added to the test. In addition, the area-specific methane yield (m³ CH₄ ha⁻¹) was calculated using values for specific methane yield and the total solids yield per hectare (McEniry, O’Kiely, 2013; Chiumenti et al., 2018).

Statistical analysis: The data structuring analysis and processing were conducted using analysis of variance (ANOVA) software SAS, version 9.4 (SAS Institute Inc., USA); P value < 0.05 was considered statistically significant. The different letters a–c in the column indicate significant differences (P < 0.05) between the components. The correlations between chemical composition and methane yield were estimated by Pearson method. ** – significant at P = 0.01, * – significant at P ≤ 0.05 level.

### Results and discussion

Different plant species produced different biomass yields. Biomass production per surface of land area amounted to 0.54–7.9 t ha⁻¹ DM (Table 1). *Medicago sativa* produced the highest biomass yield of 7.9 t ha⁻¹ DM. Biomass yields of *Onobrychis viciifolia* and *Lolium perenne* were 5.32 and 5.15 t ha⁻¹ DM, respectively. The lowest biomass yields were produced by *Festuca arundinacea* and *Phalaris arundinacea – 0.54 and 2.37 t ha⁻¹ DM, respectively. Biomass yields of *Dactylis glomerata* and *Panicum virgatum* were 3.39 and 3.63 t ha⁻¹ DM.

The chemical characteristics of the investigated perennial grasses are summarised in Table 1. Each perennial herbaceous plant species had different chemical composition. Results of our research are similar to other researchers’ data on the chemical composition of the plants: *NDF* 49.6–74.15 (Allison et al., 2012; McEniry, 2013).

![Table 1](image-url)

**Note.** DM – dry matter; NDF – neutral detergent fibre, ADF – acid detergent fibre, ADL – acid detergent lignin, WSC – water-soluble carbohydrates, Cel – cellulose, HCel – hemicellulose, C – carbon, N – nitrogen, CP – crude proteins, C:N – carbon to nitrogen ratio; different letters a–c in the column indicate significant differences (P < 0.05) in concentration of respective biomass component.

O’Kiely, 2013; Butkutė et al., 2014, ADF 35.5–37.5 (Butkutė et al., 2014), ADF 1.2–31 (McEniry, O’Kiely, 2013; Butkutė et al., 2014), WSC 4.9–18.8 (McEniry, 2013; Butkutė et al., 2014), ADL 1.2–31 (McEniry, O’Kiely, 2013; Butkutė et al., 2014), ADL 35.5–37.5 (McEniry, O’Kiely, 2013; Butkutė et al., 2014), ADF 35.5–37.5 (McEniry, O’Kiely, 2013; Butkutė et al., 2014), ADL 1.2–31 (McEniry, O’Kiely, 2013; Butkutė et al., 2014).

The volatile FOS:TAC value ranged from 0.05 to 0.13 (Table 2), biomasses of *Medicago sativa* and *Onobrychis viciifolia* had too low C:N – 13.3 and *Dactylis glomerata* and *D. glomerata* had too high C:N – 42.1,

### Table 1: Biomass yield and chemical composition of the seven perennial herbaceous plant species

<table>
<thead>
<tr>
<th>Grass species</th>
<th>Yield t ha⁻¹ DM</th>
<th>Ash</th>
<th>NDF</th>
<th>ADF</th>
<th>WSC</th>
<th>Cel</th>
<th>HCel</th>
<th>C : N</th>
<th>Digestibility</th>
<th>CP</th>
<th>C : N</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Dactylis glomerata</em></td>
<td>3.96</td>
<td>7.63</td>
<td>55.26</td>
<td>33.2</td>
<td>4.18</td>
<td>13.5</td>
<td>29.0</td>
<td>4.8</td>
<td>22.04</td>
<td>58.4</td>
<td>13.9</td>
</tr>
<tr>
<td><em>Phalaris arundinacea</em></td>
<td>2.37</td>
<td>7.43</td>
<td>62.71</td>
<td>37.8</td>
<td>4.65</td>
<td>16.4</td>
<td>33.1</td>
<td>4.5</td>
<td>24.96</td>
<td>46.6</td>
<td>12.5</td>
</tr>
<tr>
<td><em>Festuca graminea</em></td>
<td>0.54</td>
<td>7.52</td>
<td>52.19</td>
<td>31.3</td>
<td>4.02</td>
<td>16.5</td>
<td>37.3</td>
<td>4.8</td>
<td>24.96</td>
<td>46.6</td>
<td>12.5</td>
</tr>
<tr>
<td><em>Lolium perenne</em></td>
<td>5.15</td>
<td>6.67</td>
<td>54.75</td>
<td>32.8</td>
<td>3.97</td>
<td>20.3</td>
<td>28.9</td>
<td>4.9</td>
<td>15.83</td>
<td>75.2</td>
<td>18.2</td>
</tr>
<tr>
<td><em>Medicago sativa</em></td>
<td>7.9</td>
<td>9.17</td>
<td>37.38</td>
<td>35.3</td>
<td>10.9</td>
<td>11.1</td>
<td>24.4</td>
<td>4.8</td>
<td>24.96</td>
<td>46.6</td>
<td>12.5</td>
</tr>
<tr>
<td><em>Onobrychis viciifolia</em></td>
<td>5.32</td>
<td>6.92</td>
<td>32.66</td>
<td>32.1</td>
<td>11.3</td>
<td>16.9</td>
<td>20.8</td>
<td>4.8</td>
<td>24.96</td>
<td>46.6</td>
<td>12.5</td>
</tr>
<tr>
<td><em>Panicum virgatum</em></td>
<td>3.63</td>
<td>5.80</td>
<td>70.49</td>
<td>39.9</td>
<td>5.70</td>
<td>5.91</td>
<td>34.2</td>
<td>4.8</td>
<td>24.96</td>
<td>46.6</td>
<td>12.5</td>
</tr>
</tbody>
</table>

### Note.
- DM – dry matter; NDF – neutral detergent fibre, ADF – acid detergent fibre, ADL – acid detergent lignin, WSC – water-soluble carbohydrates, Cel – cellulose, HCel – hemicellulose, C – carbon, N – nitrogen, CP – crude proteins, C:N – carbon to nitrogen ratio; different letters a–c in the column indicate significant differences (P < 0.05) in concentration of respective biomass component.
- P value ≤ 0.01, * – significant at P < 0.05 in concentration of respective biomass component.
O. viciifolia had the most favourable FOS/TAC value for the anaerobic process. The content of total solids (TS) of the feedstock for anaerobic digestion was 2.94–4.14%, mineral solids (MS) – 36.3–54.6 (% TS) and volatile solids (VS) – 45.4–65.7 (% TS). Feedstock of Festuca arundinacea, P. virgatum and D. glomerata had the highest concentration of total nitrogen (N) 2.33–2.34 g L⁻¹. In contrast, the feedstock of P. arundinacea had less N – 1.68 g L⁻¹. The highest chemical oxygen demand (COD) was found when using biomass of O. viciifolia – 7.1 g L⁻¹ and L. perenne – 6.9 g L⁻¹. The COD value of other biomasses varied between 6.0–6.7 g L⁻¹. The lowest COD 5.5 value was determined for the feedstock of P. virgatum. Zielinski et al. (2017) indicated COD range of feedstock 2.8–12.7 g L⁻¹.

The data of Table 3 show the results of biogas production for 35 days in anaerobic conditions by system OxTop. Biogas release from the biomass of seven plant species having the same organic matter content slightly differed until day 30 of the anaerobic experiment, but later the treatments differentiated and produced different amounts of biogas. During this investigation, 0.076–0.096 L of biogas was produced. Biomass of L. perenne produced the highest amount of biogas – 0.096 L. Lower biogas yields were generated from the biomass of F. arundinacea – 0.087 L, O. viciifolia – 0.086 L and D. glomerata – 0.084 L. The lowest biogas production was determined for P. virgatum – 0.076 L and P. arundinacea – 0.077 L.

According to the results of the biogas analysis, biomas and methane yield in dry (DM) and in fresh (FM) mass was calculated (Table 3). Biogas yield ranged from 63.2 to 114.3 NL kg⁻¹ FM and from 210.0 to 435.3 NL kg⁻¹ DM.

Legumes and grasses were compared according to biogas production. Assessment of the production of biomas from the biomass of different perennial grasses (in DM) showed that the highest biogas yield was produced by O. viciifolia – 435.3 NL kg⁻¹ (63.2 NL kg⁻¹ FM), D. glomerata – 329.0 NL kg⁻¹ (80.0 NL kg⁻¹ FM) and P. arundinacea – 307.5 NL kg⁻¹ (90.6 NL kg⁻¹ FM) compared with the other treatments. Lower amount of biogas was produced by L. perenne – 304.3 NL kg⁻¹ (114.3 NL kg⁻¹ FM) and M. sativa – 266.9 NL kg⁻¹ (90.0 NL kg⁻¹ FM). The least-suited biomass for biogas production was that of P. virgatum and P. arundinacea.

Chiumenti et al. (2018), who studied the perennial grasses, reported biogas yield of 164.6 NL kg⁻¹ FM and 307.5 NL kg⁻¹ DM, and methane yield – 87.4 L CH₄ kg⁻¹ FM and 269.5 L CH₄ kg⁻¹ DM. Scarlet

### Table 2. Properties of feedstock (plant biomass and inoculum) for anaerobic digestion

<table>
<thead>
<tr>
<th>Feedstock (biomass + inoculum)</th>
<th>pH</th>
<th>FOS/TAC</th>
<th>TS</th>
<th>MS</th>
<th>VS</th>
<th>N</th>
<th>COD</th>
</tr>
</thead>
<tbody>
<tr>
<td>Dactylis glomerata</td>
<td>8.4</td>
<td>0.01</td>
<td>0.10</td>
<td>0.001</td>
<td>2.94</td>
<td>3.70</td>
<td>6.30</td>
</tr>
<tr>
<td>Phalaris arundinacea</td>
<td>8.4</td>
<td>0.02</td>
<td>0.13</td>
<td>0.001</td>
<td>3.50</td>
<td>3.64</td>
<td>6.36</td>
</tr>
<tr>
<td>Festuca arundinacea</td>
<td>8.4</td>
<td>0.02</td>
<td>0.10</td>
<td>0.001</td>
<td>4.04</td>
<td>4.45</td>
<td>5.55</td>
</tr>
<tr>
<td>Lolium perenne</td>
<td>8.4</td>
<td>0.02</td>
<td>0.10</td>
<td>0.001</td>
<td>4.14</td>
<td>3.81</td>
<td>6.19</td>
</tr>
<tr>
<td>Medicago sativa</td>
<td>8.4</td>
<td>0.02</td>
<td>0.11</td>
<td>0.001</td>
<td>3.01</td>
<td>4.46</td>
<td>5.43</td>
</tr>
<tr>
<td>Onobrychis vicifolia</td>
<td>8.4</td>
<td>0.02</td>
<td>0.12</td>
<td>0.001</td>
<td>3.75</td>
<td>3.63</td>
<td>6.37</td>
</tr>
<tr>
<td>Panicum virgatum</td>
<td>8.4</td>
<td>0.02</td>
<td>0.10</td>
<td>0.001</td>
<td>4.71</td>
<td>3.23</td>
<td>7.74</td>
</tr>
</tbody>
</table>


### Table 3. Yield of biogas (NL kg⁻¹) and methane (L CH₄ kg⁻¹) in fresh (FM) and in dry matter (DM)

<table>
<thead>
<tr>
<th>Grass species</th>
<th>Biogas volume</th>
<th>NL kg⁻¹</th>
<th>L CH₄ kg⁻¹</th>
<th>NL kg⁻¹</th>
<th>L CH₄ kg⁻¹</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>FM</td>
<td></td>
<td></td>
<td>DM</td>
<td></td>
</tr>
<tr>
<td>Dactylis glomerata</td>
<td>0.084 b</td>
<td>80.0 d</td>
<td>0.01</td>
<td>114.3 a</td>
<td>77.3 a</td>
</tr>
<tr>
<td>Phalaris arundinacea</td>
<td>0.077 c</td>
<td>67.4 a</td>
<td>0.01</td>
<td>110 ab</td>
<td>63.0 c</td>
</tr>
<tr>
<td>Festuca arundinacea</td>
<td>0.087 b</td>
<td>90.0 c</td>
<td>0.01</td>
<td>114.3 a</td>
<td>77.3 a</td>
</tr>
<tr>
<td>Lolium perenne</td>
<td>0.096 a</td>
<td>114.3 a</td>
<td>0.01</td>
<td>123.1 c</td>
<td>107.9 a</td>
</tr>
<tr>
<td>Medicago sativa</td>
<td>0.081 c</td>
<td>90.0 c</td>
<td>0.01</td>
<td>123.1 c</td>
<td>107.9 a</td>
</tr>
<tr>
<td>Onobrychis vicifolia</td>
<td>0.086 c</td>
<td>63.2 c</td>
<td>0.01</td>
<td>110 ab</td>
<td>63.0 c</td>
</tr>
<tr>
<td>Panicum virgatum</td>
<td>0.076 c</td>
<td>107.2 b</td>
<td>0.01</td>
<td>123.1 c</td>
<td>107.9 a</td>
</tr>
</tbody>
</table>

Note. Different letters a–e in the column indicate significant differences (P < 0.05) in concentration of respective biomass component.

The correlations between other chemical composition indicators were also found (Table 4). ADF positively correlated with cellulose 0.711** and carbon 0.681** (P ≤ 0.01), but negatively – with WSC −0.873** digestibility −0.693** (P ≤ 0.01) and CP −0.428** (P ≤ 0.05). ADL negatively correlated with hemicellulose −0.875** and cellulose −0.724** (P ≤ 0.01), but positively – with CP 0.792** and digestibility 0.721** (P ≤ 0.01).

WSC positively correlated with digestibility 0.570** too, but negatively – with cellulose −0.592** (P ≤ 0.01). Strong correlation was determined between cellulose and hemicellulose 0.921** (P < 0.01). The strongest negative correlation was determined of cellulose with digestibility −0.986** (P ≤ 0.01) as well as with crude proteins −0.853** (P ≤ 0.01). Hemicellulose negatively correlated with digestibility −0.935** (P < 0.01), crude proteins −0.933** (P ≤ 0.01) and nitrogen −0.748** (P ≤ 0.01). Carbon positively correlated with C:N
The results revealed a methane yield of 103 to 1453 m³ CH₄ ha⁻¹ DM. Chiumenti et al. (2018) have documented methane yields of 181 m³ CH₄ ha⁻¹ DM, McEniry and O’Kiely (2013) received higher values – 1157–2252 m³ CH₄ ha⁻¹ DM.

According to the biomass and methane yields, the results of specific methane yield were obtained: O. vicifolia biomass produced the highest amount of 1453 m³ CH₄ ha⁻¹ DM, M. sativa – 1326 m³ CH₄ ha⁻¹ DM and L. perenne – 1060 m³ CH₄ ha⁻¹ DM. Less suitable for methane production were F. arundinacea (103 m³ CH₄ ha⁻¹ DM), P. avicennae (792 m³ CH₄ ha⁻¹ DM) and P. virginatum (521 m³ CH₄ ha⁻¹ DM).

The highest methane yield was generated from the biomass of O. vicifolia – 277.7 L CH₄ kg⁻¹ DM; its biomass contained the highest amount of WSC and digestibility, while less ADF and cellulose.

High content of ADF and cellulose and the lowest WSC amount and digestibility of the biomass of P. arundinacea and P. virginatum led to the lowest methane yields – 123.1 L CH₄ kg⁻¹ DM and 143.4 L CH₄ kg⁻¹ DM, respectively. Based on the correlations, it can be inferred that methane yield depended on the chemical composition of plant biomass. High content of WSC and low content of ADF and cellulose in plant biomass favoured methane production.

Assessment of grass biomass and methane yields suggested that the biomass of Fabaceae (O. vicifolia and M. sativa) and L. perenne from Poacea families was most suitable for methane production. Current study provided valuable information on the suitability of different grass species, grown under the same or similar management, suitable for methane production.

Conclusions

1. Chemical composition of the biomass of the Fabaceae species tested, specifically due to the high digestibility (75.5–78.4% DM) and low cellulose (20.8–24.4% DM) content, was determined most suitable for the anaerobic digestion process. The biomass of Lolium perenne and Festuca arundinacea was found to be appropriate for anaerobic digestion compared with that of the other investigated Poacea plants, because it had the highest content of water-soluble carbohydrates (WSC) (20.3–16.4% DM) and the lowest content of cellulose (28.9–27.3% DM) and acid detergent fibre ADF (32.8–31.3% DM).

2. The results of anaerobic digestion analysis showed that the highest methane (CH₄) yields were obtained from O. vicifolia – 277.7 L CH₄ kg⁻¹ DM (40.3 L CH₄ kg⁻¹ FM), Dactylis glomerata – 213.9 L CH₄ kg⁻¹ DM (52.0 L CH₄ kg⁻¹ FM) and L. perenne – 205.7 L CH₄ kg⁻¹ DM (77.3 L CH₄ kg⁻¹ FM). The correlations corroborated that the methane yield depended on the chemical composition of the biomass. Methane positively and significantly correlated with WSC 0.761** (P ≤ 0.01) and digestibility 0.744** (P ≤ 0.01). Cellulose and ADF had negative impact on the methane production, significant correlations of cellulose – 0.793** and ADF – 0.762** (P ≤ 0.01) were found.

3. After assessing the biomass yield of the first cut and methane yield of seven perennial grass species, specific methane yields were calculated: O. vicifolia – 1453 m³ CH₄ ha⁻¹ DM, Medicago sativa – 1326 m³ CH₄ ha⁻¹ DM and L. perenne – 1060 m³ CH₄ ha⁻¹ DM, gave the best results. F. arundinacea and Phalaris arundinacea were found to be less suitable for methane production in terms of specific methane yields.

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K. Almelevičiūtė-Volungė, A. Šlepetyienė, B. Butkutė

Lietuvos agrarinių ir miškų mokslo centro Žemdirbystės institutas

Santrauka