# BIOGAS PRODUCTION FROM LATVIAN FOREST MUSHROOMS

**Vilis Dubrovskis, Imants Plume, Dagnis Dubrovskis**
Latvia University of Life Sciences and Technologies, Latvia
vilisd@inbox.lv, imants.plume@llu.lv, dagnis.dubrovskis@llu.lv

**Abstract.** In Latvia, edible mushrooms are widely used as a means of nutrition. For this purpose, both picked in the forests and specially grown are used. Several countries have special mushroom farms, where they are grown on a large scale. Researchers in several countries have found that a mixture of mushroom residues, straw and manure used for mushroom production can be used well for biogas production. Digestate from biogas plants can also be used as a substrate for a new mushroom crop. The aim of this study was to find out whether and how much methane can be obtained from several mushrooms commonly found in Latvian forests. Lactarias rufus, Lactarias forminosus, Amanita mappa, Amanita muscaria, Boletus elegans, Russula paludosa, Russula foetens and Russula xerampelina were fermented in the laboratory bioreactors under anaerobic conditions. The mushrooms were crushed and stuffed in fourteen bioreactors. Anaerobic fermentation took place at 38 °C. In order to find out how much gas could still be obtained from the inoculum, it was fermented in two bioreactors. After 21 days of fermentation, the highest methane yield of 0.433 $L \cdot g^{-1}_{DOM}$ was obtained from russula paludosa. The study shows that mushrooms are a good raw material for biogas production and there is no need to fear that if they end up in household waste used for biogas production, even Amanita mappa, yield could not be reduced. Latvian mushroom growers can also safely add their mushroom residues to other raw materials and use them in biogas plants.

**Keywords:** anaerobic digestion, methane, mushrooms.

## Introduction

Currently, there are 39 biogas plants in Latvia that use various raw materials, including agricultural waste [1]. New raw materials that could catalyze the anaerobic fermentation process are also being sought [2]. There are research results that confirm the beneficial effects of mushrooms on this process. Several countries, Latvia too, have special mushroom farms, where they are grown on a large scale. Argentine researchers studied the possibilities of using mushrooms and concluded the following. Mushrooms can grow on several lignocellulosic residues from agroindustry. Spent mushroom substrate (SMS) can be used to produce high yields of biogas. Fungi act as a pre-treatment for raw material prior to anaerobic digestion. Spent mushroom substrate is a useful resource for the generation of biogas and increasing the yield of methane production [3; 4]. The potential production of methane using SMS pre-treated with cultivated mushrooms should be added to the list of other common residues used, such as food/vegetable waste and pig manure [5]. The researchers [6] found that straw on which mushrooms were grown yielded twice as much biogas as straw on which no mushrooms were grown. The oyster-mushroom " Pleurotus sp. florida" showed fastest delignification of all tested mushrooms. Straw pre-treated by these mushrooms was fermented anaerobically to biogas [7]. Another study [8] found that mushroom cultivation on spent biomass feedstocks from biogas plants (digestate) is an attractive option to utilize the residual lignin and cellulosics to create cash flow from the use of biogas plants and make their use and operation lucrative in villages. The cultivation of oyster mushroom, Pleurotus was successful. Indian researchers [9] compared different substrates for growing mushrooms. The best turned out to be a digestate from a biogas plant. It not only resulted in the best productivity and reduced the cropping schedule but induced better quality solid mushrooms with hard texture that could be kept for about 6 days more without opening or softening. The aim of this study was to find out whether and how much methane can be obtained from several mushrooms commonly found in Latvian forests. Lactarias rufus, Lactarias forminosus, Amanita mappa, Amanita muscaria, Boletus elegans, Russula paludosa, Russula foetens and Russula xerampelina were fermented in the laboratory bioreactors under anaerobic conditions. We wanted to find out whether any of them have catalytic properties of the anaerobic fermentation process. We also wanted to find out whether mushrooms that are very poisonous to humans will affect the process.

## Materials and methods

The traditional methodology was used for the research. It is similar to that used to determine the methane potential of biomass [10-12]. 16 bioreactors, volume 0.75 l were charged with 500 g of each inoculum taken from a 110 l bioreactor operating continuously in the laboratory. With this method, the bacteria multiply rapidly and the anaerobic fermentation process proceeds rapidly. The mushrooms were collected at 6.09 in the forest in the morning of the same day when the study began. The raw materials were chopped into 2-10 mm pieces. Bioreactors R2, R3 were charged with 20 g of Lactarius rufus, R4, R5 with 20 g of Lactarius terminosus, R6, R7 with 20 g of Amanita mappa, R8, R9 with 20 g of Amanita muscaria. Bioreactors R10, R11, R12 were charged with 20 g of Boletus elegans, bioreactors R13 with 20 g of Russula paludosa, R14 with 20 g of Russula foetens and R15 with 20 g of Russula xerampelina. All bioreactors were sealed and gas storage bags were added. All bioreactors together with the gas storage bags were then weighed. All bioreactors were placed in a SNOL oven, and the anaerobic fermentation operating temperature was set at 38 ± 0.5 ºC. The composition of the emitted gas was measured with a GA 2000 gas analyzer. Prior to loading into bioreactors, all feedstocks were analyzed using Shimazu and Nabertherm equipment. The PP-50 was used to determine the pH. After 21 days, the anaerobic fermentation process was stopped, the bioreactors were removed from the oven and weighed together with the gas bags. The bioreactors were then opened. Each digestate of the bioreactor was sampled and analyzed.

## Results and discussion

Crushed mushrooms were analyzed before filling in bioreactors. The results are shown in Table 1.

Table 1

**Analyses of raw material samples before anaerobic digestion**

| Bioreactor | pH | TS, % | TS, g | Ash, % | DOM, % | DOM, g | Weight, g |
|---|---|---|---|---|---|---|---|
| R1, R16 | 7.71 | 1.12 | 5.600 | 33.53 | 66.47 | 3.722 | 500 |
| R2-R3 | | 7.41 | 1.482 | 15.59 | 84.41 | 1.251 | 20 |
| R2-R3 + In | | 1.36 | 7.082 | 29.78 | 70.22 | 4.973 | 520 |
| R4-R5 | | 8.98 | 1.746 | 6.64 | 93.36 | 1.677 | 20 |
| R4-R5 + In | | 1.41 | 7.346 | 26.50 | 73.50 | 5.399 | 520 |
| R6,R7 | | 5.22 | 1.044 | 15.80 | 84.20 | 0.879 | 20 |
| R6-R7 + In | | 1.28 | 6.644 | 30.75 | 69.25 | 4.601 | 520 |
| R,R9 | | 6.15 | 1.230 | 15.63 | 84.37 | 1.038 | 20 |
| R8-R9 + In | | 1.31 | 6.830 | 30.31 | 69.69 | 4.760 | 520 |
| R10-R12 | | 8.56 | 1.712 | 9.16 | 90.84 | 1.555 | 20 |
| R1-R12 + In | | 1.41 | 7.312 | 27.83 | 72.17 | 5.277 | 520 |
| R13-R15 | | 7.83 | 1.566 | 8.37 | 91.63 | 1.435 | 20 |
| R13-R15 + In | | 1.38 | 7.166 | 28.04 | 71.96 | 5.157 | 520 |

Notes: R – bioreactor, In – inoculum, TS – total solids, DOM – dry organic matter

The results of the analyzes show that the mushrooms have a low content of TS (5.22-8.98%), but a high content of organic dry matter in the dry matter (84.2- 93.36%). The lowest DOM content was in the Amanita mappa and the highest in Lactarius terminosus. Although the inoculum in the bioreactors was of low quality because it contained less bacteria than usual, the anaerobic fermentation process was rapid. Analyzes showed the highest methane content in biogas from all bioreactors after 7-9 days. The highest it was from R14 with Russula foetens (60.1%). When the gas was no longer produced by the bioreactors (after 21 days), the anaerobic fermentation process was stopped. The biogas and methane yields from each bioreactor are shown in Table 2. The average yields from bioreactors R1 and R16 have already been deducted.

The table shows that humans and bacteria have different eating tastes. The good edible mushroom Boletus elegans produced about twice less methane as the highly poisonous Amanita mappa and Amanita muscaria. The highest methane yield was obtained from Russula paludosa. The results show that high biogas yields were obtained, but with a relatively low methane content. This is partly due to the low dry matter and organic solids content in the bioreactors. None of the mushrooms tested (including poisonous ones) showed inhibitory or catalytic properties of the anaerobic fermentation process. Also, from Boletus elegans R11 produced 0.314 $L \cdot g^{-1}_{DOM}$ and just because the other two produced little, the overall result was so poor.

Table 2

**Biogas and methane yields**

| Bioreactor/Raw material | Biogas, L | Biogas, $L \cdot g^{-1}_{DOM}$ | Methane, aver.% | Methane, L | Methane, $L \cdot g^{-1}_{DOM}$ |
|---|---|---|---|---|---|
| R1 In | 0.1 | 0.027 | 5.30 | 0.005 | 0.001 |
| R16 In | 0.2 | 0.054 | 1.20 | 0.002 | 0.0005 |
| R1, R16 average | 0.15 | 0.41 | 3.25 | 0.0035 | 0.0008 |
| R2 LR | 1,2 | 0,953 | 4042 | 0.485 | 0.388 |
| R3 LR | 1.3 | 1.039 | 37.85 | 0,492 | 0,393 |
| R2, R3 average | 1.25 ± 0.05 | 0.999 ± 0.043 | 39.08 ± 1.285 | 0.489 ± 0.004 | 0.391 ± 0.003 |
| R4 LT | 0.7 | 0.417 | 25.43 | 0.178 | 0.106 |
| R5 LT | 2.6 | 1.550 | 32.08 | 0.834 | 0.497 |
| R4, R5 average | 1.65 ± 0.95 | 0.984 ± 0.566 | 30.66 ± 3.325 | 0.506 ± 0.328 | 0.301 ± 0.196 |
| R6 AMA | 1.3 | 1.479 | 33.23 | 0.432 | 0.491 |
| R7 AMA | 1.1 | 1.251 | 21.00 | 0.231 | 0.263 |
| R6, R7 average | 1.2 ± 0.1 | 1.365 ± 0.114 | 27.12 ± 6.115 | 0.332 ± 0.101 | 0.377 ± 0.114 |
| R8 AMU | 1.1 | 1.059 | 19.27 | 0.212 | 0.204 |
| R9 AMU | 2.0 | 0.927 | 29.30 | 0.586 | 0.565 |
| R8, R9 average | 1.55 ± 0.45 | 1.493 ± 0.066 | 24.29 ± 5.015 | 0.359 ± 0.187 | 0.385 ± 0.181 |
| R10 BE | 0.8 | 0.514 | 18.75 | 0.15 | 0.096 |
| R11 BE | 1.5 | 0.965 | 32.60 | 0.489 | 0.314 |
| R12 BE | 0.8 | 0.514 | 26.00 | 0.208 | 0.134 |
| R10, R11, R12 average | 1.033 ± 0.311 | 0.664 ± 0.2 | 25.78 ± 4.689 | 0.282 ± 0.138 | 0.181 ± 0.088 |
| R13 RUP | 2.5 | 1.742 | 33.68 | 0.842 | 0.588 |
| R14 RUF | 1.5 | 1.045 | 41.20 | 0.618 | 0.431 |
| R15 RUX | 1.5 | 1.045 | 26.87 | 0.403 | 0.281 |
| R13, R14, R15 average | 1.833 ± 0.444 | 1.277 ± 0.31 | 33.92 ± 4.856 | 0.621 ± 0.147 | 0.433 ± 0.103 |

Note: LR – Lactarius rufus; LT – Lactarius terminosus; AMA – Amanita mappa; AMU – Amanita muscaria; BE – Beletus elegans; RUP – Russula paludosa; RUF – Russula foetens; RUX – Russula xerampelina

The average biogas and methane yields from mushrooms are shown in Fig. 1.

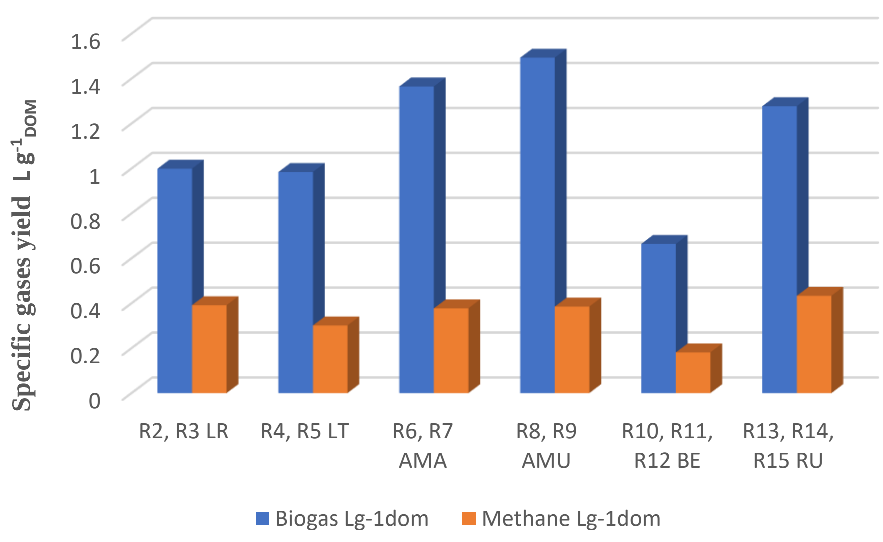


Fig. 1. **Average biogas and methane yields from mushrooms**

Average methane content in yields from mushrooms is shown in Fig. 2.

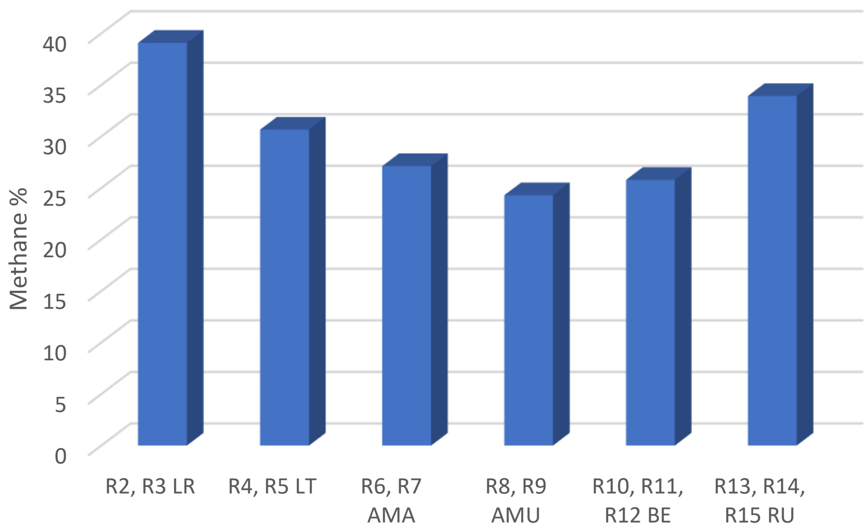


Fig. 2. **Average methane content in biogas yields from mushrooms**

Although the methane content in% of biogas from all groups of mushrooms tested was low, the yield from dry organic matter was relatively high. This can be explained by the low quality of the inoculum and the low content of dry organic matter in the raw materials.

## Conclusions

1. Average methane yield from Lactarius rufus is 0.391 $L \cdot g^{-1}_{DOM}$, Lactarius terminosus – 0.301 $L \cdot g^{-1}_{DOM}$, Amanita mappa – 0.377 $L \cdot g^{-1}_{DOM}$, Amanita muscaria – 0.385 $L \cdot g^{-1}_{DOM}$, Boletus elegans – 0.181 $L \cdot g^{-1}_{DOM}$ and Russula – 0.433 $L \cdot g^{-1}_{DOM}$
2. The study shows that mushrooms are a good, except Boletus elegans, raw material for biogas production and there is no need to fear,if they end up in household waste used for biogas production, even Amanita mappa, the yield could not be reduced.
3. None of the mushrooms tested showed catalytic properties of the anaerobic fermentation process.

## Acknowledgements

This work has been supported by the project G4 “Feasibility Study of Biomass Anaerobic Fermentation Process Efficiency”.

## Author contributions:

The contribution of each author. Conceptualization, V.D.; methodology, V.D.; software, V.D. and D.D.; validation, D.D. and V.D; formal analysis, V.D.; investigation, V.D. and I.P.; data curation, V.D. and I.P.; writing – original draft preparation, V.D.; writing – review and editing, D.D. and V.D.; visualization, V.D. ; project administration, V.D. All authors have read and agreed to the published version of the manuscript.

## References

[1] Dubrovskis V. Biogāzes ražošana Latvijā (Biogas production in Latvia) 2018, Jelgava, Latvia, 152 p. (In Latvian).
[2] Dubrovskis V., Adamovics A. Bioenergetikas horizonti (Horizons of Bioenergy) 2012, Jelgava, Latvia, 352 p. (In Latvian).
[3] Williams B., McMullan J., McCahey S. An initial assessment of spent mushroom compost as a potential energy feedstock. Bioresour. Technol. 2001, 79, pp. 227-230.
[4] Xiao Z., Lin M., Fan J., Chen Y., Zhao C., Liu B. Anaerobic digestion of spent mushroom substrate under thermophilic conditions: Performance and microbial community analysis. Appl. Microbiol. Biotechnol. 2018, 102, pp. 499-507.

[5] Müller H. W. & Trösch W. Screening of white-rot fungi for biological pretreatment of wheat straw for biogas production. Applied Microbiology and Biotechnology, 1986, volume 24, pp. 180-185.
[6] Perez-Chavez A. M., Mayer L., Edgardo A. Mushroom cultivation and biogas production: A sustainable reuse of organic resources. Energy for Sustainable Development Volume 50, June 2019, pp. 50-60.
[7] Lin Y., Ge X., Li Y. Solid-state anaerobic co-digestion of spent mushroom substrate with yard trimmings and wheat straw for biogas production. Bioresour. Technol. 2014, 169, pp. 468-474.
[8] Gangulli N. K. and Chanakya H. N. Mushroom cultivation on spent biomass from biogas plants Current Science Vol. 66, No. 1 (10 January 1994), pp. 70-74.
[9] Gupta Virendra Pratap and Kumar Vishnu Evaluation of spent biogas silage as casing soil in mushroom cultivation J. Appl. Hort., 3(2), July-December, 2001, pp. 119-121.
[10] Angelidaki I., Alves M., Bolzonella D., Borzacconi L., Campos J., Guwy A., Kalyuzhnyi S., Jenicek P., Van Lier J. Defining the biomethane potential (BMP) of solid organic wastes and energy crops: a proposed protocol for batch assays. Water Sci.Technol. 2009, 59(5), pp. 927-934.
[11] Thran D. Methodenhandbuch Energetische Biomassenutzung, (Methods Manual Energetic use of biomass), 2010, Leipzig, 161 p. (In German).
[12] VDI 4630. Vergärung organischer Stoffe Substrat charakterisierung, Probenahme, Stoffdatenerhebung, Gärversuche. Vereindeutscher Ingenieure (Fermentation of organic substances Substrate characterization, sampling, substance data collection, fermentation tests. German engineers), Düsseldorf, 2006, 48 p. (In German).