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Biotechnological aspects of digestate production during methane fermentation and utilisation

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Abstract. The relevance of the study is conditioned by the rapid development of the bioenergy industry, namely, the biogas sector in Ukraine, which leads to the generation of significant volumes of one of the by-products of anaerobic digestion – digestate, which is valuable in many aspects. The purpose of this study was to investigate the main stages of methane fermentation and factors affecting the composition and quality of digestate, and to review conventional and innovative methods of its utilisation. Analysis of the available literature has shown that current methods of storing the produced digestate are ineffective and act as a temporary way of its accumulation, given the growing scale of its generation every year and in the future. Each of the four stages of anaerobic digestion (hydrolysis, acidogenesis, acetogenesis, methanogenesis) was considered in detail in the paper and specific groups of microorganisms involved in these processes were characterised. The relationship between the composition of the raw material and the final physical and chemical characteristics of digestate was determined. The influence of the initial feedstock on the composition and key characteristics of the resulting digestate were presented. The paper analysed the results of contemporary research in the field of digestate processing and utilisation using biotechnological methods to ensure the main goals of sustainable development and compliance with the principles of the circular economy. Special attention was paid to the key characteristics of the resulting digestate, which have a predominant influence on the choice of further methods and technologies for its utilisation. Based on the analysis, the information was systematised and presented in the form of generalising tables. The results of this review paper are of practical value and can be used for the selection and implementation of digestate processing technologies in existing enterprises

Keywords: fermented residue; anaerobic digestion; processing; biogas; circular economy; sustainable development

INTRODUCTION

The transition to sustainable development models is one of the key strategies of the global economy amid the depletion of natural resources and the accumulation of substantial amounts of waste. Of particular importance is the concept of a circular economy, which aims to minimise waste by

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including it in new production cycles or reusing it. Thus, one of the stages on the way to sustainable development and a circular economy is the introduction of renewable energy sources, which have been increasingly developed over the past decades. The production of biogas and biomethane from organic waste of various origins is a relatively widespread technology that combines energy production with waste management and contributes to the achievement of the Sustainable Development Goals. The introduction of circular economy concepts is promising, which would allow obtaining new valuable products and reducing the accumulation of waste that remains as a result of the technological process of obtaining biogas and biomethane.

As noted by M.-N. Dincă *et al*. (2025), circular economy involves extending the life cycle of products through actions such as reuse, recovery, and recycling. In analytical notes, the non-governmental organisation UABIO (n.d.) noted that the total share of biomethane and biogas produced in Europe was more than 22 billion m^3 in 2024. Biomethane production remains the fastest growing segment (5.2 billion cubic metres, of which 4.3 billion cubic metres are produced in the EU-27), supported by an installed capacity of 7 billion m^3 per year in Europe at the beginning of 2025. In addition, as noted by K. Chojnacka & K. Moustakas (2024), the latest report of the Intergovernmental Panel on Climate Change indicates that global greenhouse gas emissions reached 59.1 gigatonnes of CO_2 equivalent in 2022. Appropriate waste management and utilisation will help to reduce greenhouse gas emissions, conserve resources, and produce valuable new products.

An important by-product of biogas production is digestate, a mixture of non-fermented organic residues. D. Kovačić *et al.* (2022) reported that approximately 180 million tonnes of digestate are produced annually in the EU. About 120 million tonnes are of agricultural origin (usually a mixture of manure and plants, in particular energy crops), 46 million tonnes are produced from the organic fraction of mixed municipal solid waste, at least 7 million tonnes from organic waste, while a smaller amount (about 1.7 million tonnes each) comes from the fermentation of sewage sludge and by-products of the agro- and food industries. According to M.-N. Dincă *et al*. (2025), the European Biogas Organisation estimates that 75 million tonnes of dry matter digestate will be produced in Europe by 2030, and the total potential of digestate produced in Europe by 2050 will be 177 million tonnes of dry matter.

M. Nowak & W. Czekała (2024) noted that the vast majority of digestate is used as fertiliser in liquid form in private enterprises or stored in special non-sealed tanks – lagoons. When digestate is stored for a long time in lagoons, solid particles settle and accumulate at the bottom. In addition, V. Polishchuk *et al*. (2020) indicate that due to non-sealed tanks, greenhouse gas emissions – methane, carbon dioxide, etc. – increase and unpleasant odours spread.

Accordingly, lagoons are only one of the ways to accumulate and temporarily store digestate, but not a solution to the problem of its utilisation, and given the growing scale of biogas production, the amount of digestate generated will continue to increase. Thus, it is important to find new ways to dispose of it and process it in order to obtain valuable products and comply with the principles of a circular economy. It is also important to consider the main characteristics and properties of digestate to choose the most optimal method of its processing. The purpose of this study was a comprehensive analysis of biotechnological aspects of digestate generation as a product of anaerobic digestion and analysis of current methods of its processing to ensure the principles of circular economics.

MATERIALS AND METHODS

To achieve the stated objective and ensure objectivity, a systematic analysis of contemporary scientific literature, primarily by foreign and Ukrainian authors, was conducted. This was a narrative review with elements of a systematic approach to literature search and analysis. The methodology for selecting relevant research was based on determining the search query, forming keywords for its implementation, analysing relevant information sources, and including or discarding them as relevant. Databases such as Scopus, Web of Science, Google Scholar, PubMed, and Science Direct were used for the search. In total, about 200 papers were analysed, the search for which was carried out by keywords mainly in English: biogas technology, substrate for biogas production, digestate characteristics or utilisation, physico-chemical methods or biotechnology of digestate utilisation, composting and vermicomposting, microalgae. The time frame for searching for the main sources was the period during 2022-2026, which was explained by the rapid development of this segment and the development of new technologies. In accordance with this, the results of studies in the specified time range were used and processed to obtain up-to-date information.

The key selection criteria were publications from peer-reviewed scientific journals, research focused on the biological and biochemical features of digestate generation in the process of biogas production, the influence of the main factors on this process, and the features of its utilisation, studies that included the experimental part of the real implementation of these technologies, and the presence of certain statistically significant results. The main exclusion criteria were non-peer-reviewed articles and conference abstracts; studies that duplicated previously published results by the same researchers without providing additional findings; and papers focusing primarily on the economic and logistical aspects of the process under study. The analysis of the selected literature was based on methods of systematisation, identification of key results of publications, comparison, and structuring of the selected information in text format and using summary tables. As a result of the analysis and systematisation according to the above criteria, 35 publications were selected, including review papers and experimental studies on the main stages of methane fermentation and the biological agents involved in each of them, the main characteristics of digestate obtained as a result of anaerobic digestion and their influence on processing methods.

RESULTS AND DISCUSSION

Anaerobic digestion is a complex biochemical process of converting organic waste into biogas and a liquid fraction – digestate with the participation of a whole consortium of microorganisms. In their publication, D. Kovačić *et al*. (2022) indicated that this technology is widely used for the treatment of agricultural and food waste, organic fraction of municipal solid waste, and sediments generated as a result of wastewater treatment. As noted by K. Pilarski & A. Pilarska (2025), anaerobic digestion was carried out in hermetically sealed reactors, which ensured stable process conditions and control over operating parameters. The resulting biogas is sent to storage tanks and then most often used in combined heat and electricity generation systems. It can also be burned directly for heat generation or converted to biomethane for injection into the grid or used as a transport fuel, depending on local energy demand profiles and the availability of appropriate infrastructure.

According to M. Jameel *et al*. (2024) biogas is a mixture of gases consisting of 50-70% methane (CH_4), 30-50% of carbon dioxide (CO_2), and residues of other gases, such as water vapour (H_2O) and hydrogen sulphide (H_2S). Depending on further use, the impurities present in the biogas are removed. As indicated by H.G. Katariya & H.P. Patolia (2023), various methods are being developed that various methods are being developed for upgrading biogas to biomethane by removing CO_2, H_2O and other residual gases that may cause equipment and engine damage and reduce their service life. In addition, some of them, namely hydrogen sulphide, are toxic and, if released into the atmosphere, can pose a threat to human health and the state of ecosystems. As mentioned above, in addition to biogas, as a result of anaerobic fermentation of organic waste, a by-product is formed – digestate, consisting of a mixture of solid and liquid fractions that do not decompose during fermentation. Digestate is rich in organic substances and macro- and microelements, and that is why it is quite a valuable product.

To better understand the composition and characteristics of digestate, it is necessary to consider the key stages and conditions of fermentation. D. Kovačić *et al*. (2022) noted that the anaerobic fermentation process is carried out due to the joint action of groups of microorganisms: hydrolytic and acidogenic bacteria, syntrophic acetogenic bacteria, and methanogenic archaea, and is generally divided into 4 stages – hydrolysis, acidogenesis, acetogenesis, and methanogenesis. The syntrophic nature of this process makes each stage important, as indicated by T. Induchoodan *et al*. (2022). For example, acidogenesis is a prerequisite for methanogenesis, but if a large amount of acids is formed during the acidogenesis stage, this will affect the methanogens and interfere with the further process. As noted by K. Pilarski & A. Pilarska (2025), during the first phase of anaerobic digestion, polymers such as starch, proteins, and fats are hydrolysed to monomers – simple sugars, amino acids, glycerol, and fatty acids – using hydrolytic exoenzymes such as cellulases, hemicellulases, and proteases, which are mainly secreted hydrolytic and fermentative bacteria of the genera microorganisms belonging to the phyla *Firmicutes* and *Bacteroidetes* and the genera *Ruminococcus* and *Clostridium*.

Hydrolysis is a relatively slow step, and it can limit the speed of the overall anaerobic digestion process, especially when using large amounts of solid waste as a substrate. According to P. Bajpai (2017), the next step is acidogenesis, during which simple organic substances are converted to higher organic acids (propionic and butyric acids), and acetic acid, hydrogen, and carbon dioxide. This phase is managed by a broad consortium of enzymatic and acidogenic bacteria, as noted by K. Pilarski & A. Pilarska (2025), including representatives of *Clostridium*, *Lactobacillus*, *Enterobacter*, and *Bacteroides* which convert sugars, amino acids, and other hydrolysis products into volatile fatty acids (VFA), alcohols, CO_2, and H_2. In the third phase of acetogenesis, acid-forming bacteria produce carbon dioxide, hydrogen, acetate, hydrogen sulphide, ethanol, and very small amounts of methane, nitrogen, and ammonia (Jameel *et al*., 2024). The key participants at this stage are syntrophic acetogenic bacteria, such as *Syntrophomonas* and *Syntrophobacter*, which oxidise VFA (e.g. propionate and butyrate) to acetate and H_2, as indicated in the publication by K. Pilarski & A. Pilarska (2025). However, according to P. Bajpai (2017), it is difficult to clearly draw the line between the acidogenic and acetogenic stages. Acidogenic and acetogenic bacteria belong to a large and diverse group that includes both facultative and obligate anaerobes. Species isolated from anaerobic reactors include *Clostridium*, *Peptococcus*, *Bifidobacterium*, *Desulfovibrio*, *Corynebacterium*, *Lactobacillus*, *Actinomyces*, *Staphylococcus*, *Streptococcus*, *Micrococcus*, *Bacillus*, *Pseudomonas*, *Selemonas*, *Veillonella*, *Sarcina*, *Desulfobacter*, *Desulfomonas*, and *Escherichia coli*.

The next stage is methanogenesis – a complex phase carried out due to the metabolic action of a complex of methanogenic archaea. They grow slowly, are very sensitive to changing environmental conditions, and are strict anaerobes, as indicated by M. Jameel *et al.* (2024). As noted by K. Pilarski & A. Pilarska (2025), there are several pathways of methane generation by methanogens: acetoclastic and hydrogenotrophic. Acetoclastic methanogenesis is carried out mainly by archaea of genera *Methanosaeta* and *Methanosarcina*, which convert acetate to CH_4 and CO_2, whereas hydrogenotrophic methanogenesis is carried out by genera such as *Methanobacterium*, *Methanospirillum* and *Methanococcus*, which reduce CO_2 to methane using H_2. In addition, there is also a pathway for methane production from methanol and methylated compounds, but it is less common. According to P. Bajpai (2017), some of the species that have been isolated and classified include *Methanobacterium formicicum*, *M. bryantic* and *M. Thermoautotrophicum*; *Methanobrevibacter ruminantium*, *M. arboriphilus*, and *M. smithii*; *Methanococcus vannielli* and *M. voltae*; *Methanomicrobium mobile*; *Methanogenium cariaci* and *M. marinsnigri*, *Methanospirilum hungatei*, and *Methanosarcina barkei.*

In general, according to V. Polishchuk *et al.* (2020), digestate is a product of bioconversion of organic materials during methane fermentation, causing complex organic matter to break down into simpler organic compounds, inorganic substances, microbial biomass, and biogas. The amount of digestate obtained is usually 70-90% of the volume of incoming raw materials, as indicated by M.-N. Dincă *et al.* (2025). It is biologically stable and rich in nitrogen, phosphorus, and potassium. Due to the decomposition of organic matter, mineralisation processes and the generation of biogas, in comparison with the input raw materials, the content of dry matter in the resulting digestate decreases, and moisture content increases accordingly (it can be in the range of 92-99%, but, as a rule, it is 94-96%) and viscosity decreases. Digestate has a high content of easily accessible ammonium nitrogen for plants (about 60-80% of the total nitrogen content); has a balanced C/N ratio (20-30), and a pH value close to neutral (6.5-8.0); almost does not contain viable weed seeds and pathogenic microflora (provided that the required duration and temperature of the process is observed, which mainly provides a thermophilic fermentation regime), which was indicated by V. Polishchuk *et al.* (2020). For these reasons, digestate is often used in agriculture as a substitute for synthetic fertilisers.

According to R. Feiz *et al.* (2022), the most common method of further digestate treatment is separation into two fractions, solid and liquid, with distinct nutrient profiles, i.e., different N:P:K concentrations and ratios. The liquid fraction can be used as fertiliser, and the solid fraction can be used as a soil improver. The solid fraction consists of a certain amount of organic matter, but contains a large amount of cellulose and lignin, which affects its sorption properties. Its use in agriculture allows increasing the moisture capacity of the soil and content of organic matter. In addition, the vast majority of phosphorus passes from digestate to the solid fraction. The liquid fraction contains more soluble forms of nitrogen, phosphorus, and potassium, which have a higher level of assimilation by plants, compared to the solid form. The liquid phase is characterised by a high content of nitrogen and potassium, with 80% nitrogen in the form of ammonia ions, as noted by M. Nowak & W. Czekała (2024). It was also proved, as indicated by M.-N. Dincă *et al.* (2025), that digestate contains beneficial microorganisms such as nitrogen-fixing, denitrifying, and nitrifying bacteria, saprophytic fungi, and soil methanogens. In addition, digestate-derived microorganisms can also be used in the soil bioremediation process due to their ability to decompose aliphatic hydrocarbons from oil-contaminated soils (Table 1).

Table 1. Characteristics of the biogas generation process

Stage	Process	Substrate	Reaction products	Microorganisms
Hydrolysis	Hydrolysis of biopolymers to monomers	Blend of protein, fat, and carbohydrates	Amino acids, fatty acids, mono- and oligosaccharides	Cellulolytic fungi, hydrolytic microorganisms: *Bacillus subtilis*, *Pseudomonas putida*, *Aspergillus niger*
Acidogenesis	Organic compounds obtained as a result of hydrolysis are converted into organic acids with the participation of bacteria	Amino acids, fatty acids, mono- and oligosaccharides	Short-chain organic acids (butyric, propionic, etc.)	Acidogenic bacteria: *Clostridium acetobutylicum*, *Bacteroides fragilis*, *Enterobacter aerogenes*
Acetogenesis	Acetate is formed in two ways: from the acids of the previous stage, with the release of hydrogen, and by reduction of CO_2 using hydrogen	Short chain organic acids (butyric, propionic, etc.)	Hydrogen, carbon dioxide, acetic acid	Homoacetogenic bacteria: *Butyribacterium methylotrophicum*, *Acetobacterium woodii*, *Syntrophomonas wolfei*
Methanogenesis	Methane is formed in two ways: by splitting acetate and carbon dioxide reduction	Acetic acid, carbon dioxide	Biogas, digestate	Hydrogenotrophic and acetotrophic methanogens: *Methanobrevibacter smithii*, *Methanosarcina barkeri, Methanogenum thermautotrophicus*, *Methanosarcina thermophila*
Sulphate reduction	Generation of hydrogen sulphide and carbon dioxide	Substrates from previous stages	Hydrogen sulphide, carbon dioxide	*Desulfovibrio vulgaris*, *Desulfobacter postgatei*

Source: created by the authors based on the analysis performed

As mentioned above, the composition of the initial substrate used for anaerobic digestion significantly affects the further composition and physico-chemical characteristics of the digestate. In addition, the features and parameters of fermentation are important, including the operating conditions of the biogas plant and the physico-chemical parameters of the fermentation process (temperature, pH, etc.). For consideration, it is customary to take such a division of types of input substrates into 5 categories: agricultural waste and by-products (animal manure, silage), crop residues (plant residues, straw), sewage sludge (municipal and household), organic fraction of municipal solid waste, and food production waste.

Thus, according to, P. Ragályi *et al.* (2025), animal manure is one of the most common substrates used for biogas production. During anaerobic fermentation, approximately 70% of the total nitrogen content in raw animal manure is mineralised to form ammonium ions (NH_4^+) or free ammonia (NH_3). Digestate obtained from animal manure usually contains 0.8-5.0 g/L of ammonium nitrogen. Phosphorus (P) comes partly from feed and partly from inorganic additives, and its concentration can reach 1.2 g/L in the resulting digestate. Dissolved P can be detected mainly in the form of orthophosphate (PO_4^{3-}), and its presence strongly depends on the pH indicator. Approximately 90% of the phosphorus content in the digestate forms a precipitate in the form of compounds with metals, while the dissolved fraction is only about 10%. In addition, poultry manure and cattle manure contain a higher proportion of solid components and lead to slower decomposition than in the case of pig manure. Compared to others, poultry manure has a higher content of ammonium nitrogen and total nitrogen, which accordingly affects the nitrogen content in the digestate formed from it. Digestate obtained from pig manure contains approximately 80% of the total nitrogen content in the form of NH_4^+.

As noted by J. Ries *et al.* (2023), digestate produced by the fermentation of food waste is usually rich in nitrogen, chlorine, and sodium, along with lower concentrations of phosphorus, sulphur, boron, magnesium, manganese, and molybdenum. The total percentage of nitrogen is usually between 4-5%, but there is evidence that it can be 1.1-9.6%. Common elements found in excess include Na and Cl, which can be explained by the high salt content of food waste. Recorded Na^+ concentration ranges from 36-319 mg/L, while the Cl^- concentration is in the range of 87-2,100 mg/L. The pH value is mostly alkaline. Due to the fermentation of raw materials with a high protein content, which prevails in the composition of food waste, the resulting digestate is characterised by an increased content of ammonium nitrogen, as noted by F. Rivera *et al.* (2022).

P. Ragályi *et al.* (2025) indicated that digestate formed from the organic fraction of municipal solid waste usually has a slightly alkaline pH and contains both macro- (N, P, K, Ca, S, and Mg) and microelements (B, Cl, Mn, Fe, Zn, Cu, Mo, and Ni). In some cases, the N, P, and K content may be high, but in general, the nutrient concentration is usually lower compared to digestate from manure or sewage sludge. High (about 30-50%) total solid content, high C:N ratio and increased content of non-fermented components (plastic, metal, rubber, glass, ceramics, sand, and stones) in the organic SHW fraction can significantly complicate the anaerobic digestion process and, accordingly, affect the composition of digestate. Crop residues (plant residues left after harvesting: roots, straw, etc.) are difficult to decompose due to the suboptimal C:N ratio, so they are usually used as a co-substrate in combination with animal waste. Plant tissues have a high content of lignocellulose, and therefore, a complex structure that makes them resistant to decomposition, so the resulting digestate is characterised by a high content of cellulose. A higher yield of digestate is formed from plants with a high fibre content, and, conversely, with a lower fibre content – less digestate is formed. In addition, this digestate does not contain pathogenic microorganisms and heavy metals.

Classical wastewater treatment plants form sediments that differ in organic matter content and humidity. Primary sludge is formed as a result of the process of precipitation of wastewater in primary settling tanks and consists mainly of organic substances, with a solid content in the range of 2-8% and with a moisture content of 96%. Sediment from secondary sedimentation tanks contains mainly microbial biomass after biological treatment facilities (excess activated sludge from aeration tanks, biofilm from biofilters, etc.). It has a lower dry matter content than primary silt, about 1-2%, and a humidity of about 98-99%. A mixture of such sediments, according to A. Elgarahy *et al.* (2024), contains about 50% organic residue, 1-6% total nitrogen and other macro- (mainly phosphorus, sulphur) and trace elements. The possible presence of surfactants, pharmaceuticals and antibiotics, cosmetics, heavy metals (Ni, Pb, Cr, and Zn) and pathogenic bacteria is dangerous. In general, according to D. Yang *et al.* (2022), anaerobic digestate from various sources has the common characteristics of high ammonium nitrogen content, alkaline pH (6.5-8.0), low C/N ratio, and different content of valuable macronutrients. Thus, digestate from food waste usually contains 1 kg N/t and 0.25 kg P/t, from sewage sludge – 2 kg N/t and 0.5 kg P/t, and cattle and chicken manure – 5-15 kg N/t and 0.1-1 kg P/t.

In addition, during anaerobic digestion, about 70% of the total nitrogen in organic waste is converted to its ammonium form, which leads to its high content in digestate. This factor is one of the obstacles in the processing of digestate, especially by biological methods. As mentioned at the beginning, the operating parameters and operating mode of the biogas plant during the fermentation process are important. Operating parameters include temperature, fermentation duration, C/N ratio, and pH. Thus, the operation of a thermophilic (~55°C) plant requires greater energy input, but the amount of biogas produced will be higher, and the digestate will be of high quality and free of pathogens. However, the decomposition process is often unstable, and plants operating in a higher temperature range are more expensive to maintain than mesophilic (~35°C) systems, so most commercial biogas plants operate in mesophilic mode.

P. Ragályi *et al*. (2025) indicate that the fermentation time and organic load factor should be sufficient to form biogas from the organic substrate available in the raw material. Insufficient duration along with a high organic load can lead to a high content of unprocessed organic substances in the digestate and, accordingly, vice versa. The optimal C/N ratio for most anaerobic digestion processes is between 15 and 30, depending on the characteristics of the substrate. A low C/N ratio means a higher concentration of N, which will eventually turn into excess ammonia. Excess ammonia increases the alkalinity of the reactor and slows down the fermentation process, which leads to a decrease in the yield of biogas and an increased content of ammonia in the digestate. On the other hand, a high C/N ratio means a lack of N in the substrate. Microorganisms quickly consume N, and it is not enough for their metabolism.

According to M. Uddin & M. Wright (2023), most microorganisms prefer a neutral pH value. Methanogens are very sensitive to pH and require a pH of 7 for maximum performance. Hydrolytic and acid-forming bacteria work better at pH values between 5.5 and 6.5. Acid-forming bacteria, on the other hand, are more tolerant of a larger pH range. Maintaining an optimal pH for all microorganisms in a single reactor is a challenge, especially for substrates with uneven composition, such as sewage sludge or organic SHW residues. Therefore, Table 2 summarises the data on the effect of substrate type and conditions on the fermentation process and digestate characteristics.

Table 2. Influence of the input substrate composition on the fermentation process and digestate characteristics

Substrate	Features	Influence on the fermentation process and digestate composition
Animal excreta	High content of ammonium nitrogen and total nitrogen in poultry manure; presence of methanogens in cattle manure	Gradual accumulation of nitrogen compounds causes an increase in pH and cessation of fermentation
Crop residues (plant remains)	High fibre content, high C:N ratio, no methanogens; no heavy metals	Fermentation without mixing with other types of substrates is ineffective due to the high C:N ratio and the absence of methanogens
Organic part of SHW (food scraps, food waste, organic waste)	High total solid content; unstable composition throughout the year; presence of mechanical impurities and hazardous chemicals: lower nutrient content compared to other substrates	Mechanical impurities can cause disruption of the fermentation process and damage to equipment; instability of the composition requires frequent formulation changes and affects the final composition of digestate
Sediments formed as a result of wastewater treatment in cities or private farms	High content of phosphorus and trace elements; possible exposure to surfactants, medicines, antibiotics, cosmetics, and other chemicals; low fibre content	Negative impact of chemicals on the fermentation process and the composition of digestate; the need to combine with other types of substrates in order to increase the fibre content

Table 2. Continued

Substrate	Features	Influence on the fermentation process and digestate composition
Waste from food production and catering establishments	Increased content of Na^+ and Cl^-; variable composition depending on the type of enterprise	A higher proportion of animal-derived products results in a higher nitrogen content in digestate; a higher proportion of plant-derived products results in a lower nitrogen content

Source: created by the authors based on the analysis performed

The use of mechanical methods for the processing and utilisation of digestate is more widespread today. The purpose of mechanical processing is to separate digestate into 2 fractions: liquid and solid, which is one of the most common methods of processing digestate. For this purpose, as noted by A. Cathcart *et al.* (2023), the screw press and decanter centrifuge are the most used. Screw presses separate the particles by size by pressing the digestate against the filter material with a screw, which allows liquid and solid particles smaller than the filter pores to pass through them and form a liquid fraction. The dehydrated solid fraction moves along the screw, the diameter of which gradually increases, which leads to an increase in pressure. The final stage of the process is the removal of digestate to the outside, as indicated by M. Nowak & W. Czekała (2024).

A. Cathcart *et al*. (2023) wrote that decanter centrifuges are divided by density; the digestate rotates in a rotating bowl, and an internal auger pushes the solids stuck in the bowl into the solid release hole. With centrifuges, a higher separation quality is achieved, since the process is not limited by the pore size. The predominant application of the two resulting digestate fractions is in agriculture, where they are used as fertilisers, soil amendments, and growing media, providing a relatively simple and economically viable means of digestate utilisation. The dry matter, nitrogen, phosphorus and potassium contents, and pH, are the most important parameters determining the suitability of digestate as a fertiliser, according to W. Czekała *et al.* (2020). The dry matter content, which usually does not exceed 8%, makes the application of digestate similar to manure. After preliminary separation of digestate by mechanical means, its solid fraction consists of a certain amount of organic matter, contains a large amount of cellulose and lignin, about 20-30% of dry substances, and a higher phosphorus content. The solid fraction is used for composting and subsequent application to the soil. In their publications, M. Nowak & W. Czekała (2024), described that the liquid fraction contains more soluble forms of nitrogen, phosphorus and potassium, which are better absorbed by plants. One of the ways to use it is liquid fertiliser, due to the high content of more readily assimilable forms of nitrogen, potassium, and phosphorus. The second method is to partially return the part from the filtered digestate back to the reactor to replenish the liquid part level and reduce water use. However, this pathway requires strict control of the nitrogen content, since it can return together with the digestate and accumulate in the reactor, which leads to inhibition of the fermentation process. However, despite the common processing method, digestate does not meet several requirements and standards that allow it to be used as fertiliser without pretreatment and careful analysis. Digestate, depending on the composition of the substrate used for fermentation, has a rather unstable composition and nutrient ratio. This, in turn, was indicated by M. Zielińska & K. Bułkowska (2024). Accordingly, the application of such fertiliser may have an unknown effect on the state of the soil and the ecosystem as a whole. Excess nutrients can lead to their leaching and contamination of surface and ground water.

The next reason, according to M. Malhotra *et al*. (2022), are pathogens, toxic substances, and heavy metals that can accumulate in the field where fertilisers are applied. Biogas production mainly uses waste that may be biologically contaminated and contain heavy metals, including Cd, Cr, Cu, Pb, Ni, Zn and As, or other toxic substances. In addition, some of the microorganisms involved in fermentation are dangerous for humans and animals. When they enter the soil, they can pass through the food chain to humans, causing several diseases. Such microorganisms, according to D. Kovačić *et al*. (2022) include

Escherichia coli, *Salmonella* spp., *Listeria monocytogenes*, *Clostridium perfringens*, *Campylobacter jejuni*, *Cryptosporidium parvum*, *Giardia intestinalis*, *Clostridium botulinum*.

The economic component is also important: the cost of transporting fertilisers from digestate is often quite high and unprofitable. In addition, many farmers refuse to use such fertilisers, since synthetic fertilisers are cheaper and give a more predictable result. There are also a number of legal restrictions that prevent the use of digestate as fertiliser. This industry is at the stage of development and requires the establishment of clear norms and legislative documents. One of the reasons may also be the lack of agricultural land due to weather and climatic conditions. There are a number of countries that have almost no developed agricultural sector. Accordingly, the processing of digestate for fertilisers is economically impractical due to the rather high cost of transportation and the lack of places for direct use. Thus, the above list of reasons that prevent the processing of digestate into fertilisers, and the rapid development of the bioenergy industry, proves the need to find new methods of its utilisation to obtain valuable products and achieve the sustainable development goals.

A more promising way, according to M. Zielińska & K. Bułkowska (2024) the recovery and reuse of valuable elements such as N, P, and K, which are essential for plant growth, can reduce dependence on synthetic fertilisers and the impact associated with direct application of digestate to the soil. Restoring nutrients and macro- and microelements from digestate instead of direct application to the soil helps to control the dosage of nutrients, prevents water pollution and soil depletion, and reduces the risk of infection with pathogens or contamination with other toxic compounds. However, this technology requires further research to find effective methods of isolation.

One of the promising ways to process the liquid fraction of digestate is to extract valuable macro- and microelements using physical methods. The recovery of nutrients from digestate follows the principles of a circular economy by converting waste into valuable products such as fertilisers and water. These methods include micro- and ultrafiltration, reverse osmosis, and membrane technologies. As indicated by V. Proskynitopoulou *et al*. (2024), micro- and ultrafiltration allowed minimising the solid content of liquid digestate, and the use of reverse osmosis gave almost deionised water. Solids trapped by filters can be dried and used as soil improvers. However, M. Zielińska & K. Bułkowska (2025) noted that the disadvantages of using these technologies are significant economic costs, scale-up limitations, and gradual contamination of membranes and filters, which as a result require constant cleaning or even disposal.

According to M. Lee *et al*. (2021), biochar is a renewable soil improver that is produced by pyrolysis of organic matter under anaerobic conditions, resulting in a substance that preserves nutrients in soil ecosystems and stabilises soil structure. After treatment, biochar has a large surface area, which increases microbial activity and water absorption, and reduces nutrient leaching and ultimately improves plant growth when used as an additive to substrates, as indicated by J. Ries *et al*. (2023). Due to its special properties, biochar is suitable for various agricultural applications. It helps to improve the physical properties of the soil and increases the availability of nutrients. When added to compost, biochar not only improves the composting process, but also improves the quality of the resulting product. Despite these advantages, M. Zielińska & K. Bułkowska (2024), reported that the biochar production process faces difficulties, including the initial cost of pyrolysis equipment, the need for specialised technical knowledge, and the unstable quality of the resulting finished product due to the unstable composition of digestate.

Composting is one of the ways to process the solid fraction of digestate. In general, this is the biodegradation of biomass under controlled aerobic conditions, with the production of the final product – compost, which is stable enough for safe storage and processing, and mature enough for safe use in agriculture (Tambone *et al*., 2015). As indicated by D. Kovačić *et al*. (2022) during composting, organic matter from the feedstock is converted by microorganisms into a stable humus-like material with the release of carbon dioxide and thermal energy. The composting process includes several phases: mesophilic, thermophilic, cooling, and maturation phases. In the beginning, biomass has a slightly acidic pH and room temperature. Further, microorganisms use monosaccharides, starch and lipids to form organic acids, which reduces the pH. In the next step, microorganisms begin to break down proteins, and then more complex compounds such as cellulose, hemicellulose, and lignin are partially converted to humus, which is the product of the humification process.

According to A. Aguilar-Paredes *et al*. (2023), the dominant phyla of bacteria in the composting process are: *Firmicutes*, *Actinobacteria*, *Proteobacteria*, *Bacteroidetes*, and *Chloroflexi*. Members of the phyla Firmicutes and Actinobacteria play an important role in lignocellulose decomposition, while members of the phylum Bacteroidetes are involved in the degradation of a wide range of complex carbohydrates. As a result of composting, a valuable soil amendment is obtained, rich in humic and fulvic acids, which contribute to increased soil fertility. In addition, the application of such compost improves the structure of the soil and reduces dependence on synthetic fertilisers. M. Zielińska & K. Bułkowska, (2024), noted that during composting, moisture evaporates from the digestate, reducing its volume and facilitating transportation. Due to the increase in temperature as a result of microbial activity to almost 60°C, neutralisation of pathogenic microorganisms that may be present in the digestate is ensured. However, D. Kovačić *et al.* (2022) reported that, because digestate is characterised by a high ammonium content, NH_4^+ can be converted into toxic and volatile NH_3 during composting through a series of reactions, potentially entering the atmosphere and contributing to acid rain and subsequent eutrophication of local ecosystems. In addition, raw digestate is quite moist and dense, which can reduce air access to the compost system. Therefore, in order to prevent this, it is often necessary to add fillers (usually lignocellulose waste), which increases operating costs.

According to A. Vuković *et al.* (2021), vermicomposting is the process of composting a wide range of organic waste, which involves certain types of earthworms that improve the process of converting waste into a high-quality final product, known as vermicompost. The principle of the process is similar to composting, but it is carried out with the help of earthworms and a whole association of microorganisms. M. Sani *et al*. (2025) reported that earthworms provide substrate grinding, improve aeration through constant movement, and secrete specific mucus, which is a source of nutrients for microorganisms. The following types of earthworms are mainly used for vermicomposting: *Eisenia fetida*, *Eisenia andrei*, and *Perionyx excavatus*. The association of microorganisms consists mainly of genera *Bacillus* (*Firmicutes*), *Streptomyces* (*Actinobacteria*), *Pseudomonas* sp. (*Proteobacteria*) and *Azotobacter* (*Proteobacteria*). Microorganisms provide biochemical conversion of nutrients into a form accessible to plants and secrete specific phytohormones – auxins, gibberellins, etc.

Compost formed as a result of vermicomposting is a balanced granular organic fertiliser containing approximately 30% humus, 0.8-3.0% nitrogen, 0.8-5% phosphorus, 1-2% potassium, 2-5% calcium (in terms of absolutely dry matter). For one development cycle (180 days) 0.5 kg of worms per 1 m^2 produce 400-500 kg of humus with a moisture content of 50% from 1 tonne of compost mass and increase their biomass to 8 kg. Using digestate as a substrate for vermicomposting has the same advantages and disadvantages as conventional composting. This includes increasing the content of nutrients in compost and increasing their availability to plants. One of the main disadvantages, in addition to those listed above, is the high content of ammonia in the digestate, which can lead to the death of worms.

Nowadays, considerable attention is being paid to the use of microalgae as a source of biomass, valuable products and energy, and a way to recover nutrients from waste. One of the ways to process digestate and produce useful products is to cultivate microalgae in a liquid fraction, as noted by R. Rasheed *et al*. (2023). Microalgae are photosynthetic microorganisms that use atmospheric carbon dioxide and solar energy to produce a variety of proteins, carbohydrates, lipids, minerals, vitamins, polyphenols, flavonoids, and carotenoids. Products based on them can be used in various industries, including food processing, agriculture (as animal feed and feed additives), fisheries, cosmetics, bioenergy, etc. as noted by A. Ahmad *et al*. (2022).

Microalgae occur as individual cells or in groups of several cells and can vary considerably in shape and size. As indicated by E. Thoré *et al*. (2023), more than 30,000 microalgae species have already been described, many of which have found industrial applications. For example, *Dunaliella salina* can grow in very salty water and is grown as a source of beta-carotene; *Haematococcus pluvialis* accumulates high concentrations of the carotenoid astaxanthin, which is used as an antioxidant; the genus *Arthrospira* accumulates the blue pigment phycocyanin; *Phaeodactylum tricornutum* synthesises the orange-brown pigment fucoxanthin. In addition, many microalgae are high in omega-3 fatty acids, such as eicosapentaenoic acid and docosahexaenoic acid. J. de Carvalho *et al*. (2022) noted that microalgae can grow on fairly simple sources of carbon and other nutrients, including digestate and wastewater. Algae grown on digestate can absorb dissolved carbon dioxide and nutrients such as ammonia, nitrates, nitrites, and phosphates. These characteristics make them very effective for processing liquid digestate for the purpose of accumulating algae biomass and further use for other purposes.

In addition, according to G. Geletukha *et al*. (2024), the possibility of creating a closed cycle of growing microalgae on digestate with the return of unseparated microalgae suspensions to the bioreactor for renewed biogas production is promising. The idea is to effectively grow algae on a nutrient medium from digestate, which contains a large amount of nutrients, macro- and microelements necessary for their growth, and with the addition of carbon dioxide obtained as a result of biogas enrichment. The cultured algae biomass can be returned to the reactor for fermentation or used to produce biodiesel or other valuable products. An example of this technology is a pilot plant for growing microalgae on a liquid fraction of digestate at a wastewater treatment plant in Milan, Italy. Digestate is obtained as a result of anaerobic digestion of sediments formed after urban wastewater treatment. The dominant algae species during cultivation were *Chlorella* and *Scenedesmus* spp. with an average growth rate of 9.5 ± 6.4 g dry matter/m^2·day during the entire cultivation period. The plant saved about 20% of nitrification energy, while producing valuable microalgae biomass that can be used in many areas, as indicated by S. Rossi *et al.* (2023). A generalised table of biological and physico-chemical methods of digestate processing is shown in Table 3.

Table 3. Comparative characteristics of digestate processing methods

Method	Method principle	Advantages	Disadvantages
Biological methods			
Composting	Composting of the solid fraction of digestate to obtain a valuable fertiliser	Low cost; high content of nutrients, macro- and microelements; partial destruction of pathogens due to increased temperature during composting	Need to add fillers; unknown composition; risk of pathogens and heavy metals entering the soil; insufficient regulation by legislation
Vermicomposting	Composting of the solid fraction of digestate to obtain valuable fertiliser with the help of worms of the genus *Eisenia* and others	Low cost; high content of nutrients, macro- and microelements	Need to add fillers; the risk of worm death due to the high content of nutrients and unknown composition; the risk of pathogens and heavy metals entering the soil
Cultivation of microalgae	Cultivation of microalgae on a liquid fraction of digestate to obtain valuable food additives	Obtaining a valuable food additive or fertiliser from microalgae and purified liquid digestate fraction; the possibility of using the resulting biomass for re-fermentation	Inhibition of microalgae growth due to the high content of ammonium and other nutrients in the digestate; possible crop death due to the presence of pathogens and heavy metals in the digestate; risks of use as a food additive
Physical methods			
Mechanical separation	Separation of digestate into 2 fractions: solid and liquid, mainly for use as fertilisers	Simplicity and relatively low cost; reduction of the volume of digestate generated; low cost; high content of nutrients, macro- and microelements	The need for further processing of the formed 2 fractions; uneven distribution of nutrients; the risk of ingress of increased volumes of methane and ammonia into the atmosphere

Table 3. Continued

Method	Method principle	Advantages	Disadvantages
Nutrient recovery	Use of ultra- and microfiltration methods, reverse osmosis, membrane technologies for concentrating nutrients from the liquid fraction and obtaining purified water	Obtaining a valuable concentrate of nutrients; obtaining purified water that can be reused; reducing the volume of digestate formed	Expensive equipment; the need for constant cleaning of filter materials and membranes; difficulty in scaling; unknown composition of the resulting concentrate
Biochar	Pyrolysis of the solid fraction of digestate to produce biochar as a soil improver	Reduced digestate volumes; lower transportation costs; high content of valuable macro- and microelements; destruction of pathogens	Expensive equipment and its maintenance; risk of equipment damage due to excessive ash accumulation; risk of heavy metals entering the soil

Note: unknown composition – the need for constant thorough analysis of each batch of digestate to determine the specific content of nutrients and other macro- and microelements, heavy metals, and the presence of pathogenic microorganisms
Source: created by the authors as a summary of the above information

The analysis of the literature showed that biological methods of digestate processing are the most environmentally safe and promising technologies that ensure compliance with the principles of a circular economy with the possibility of simultaneously obtaining valuable by-products.

CONCLUSIONS

Due to the growing sector of biogas and biomethane production in Europe and Ukraine, there is a problem in processing large volumes of the resulting digestate to meet the Sustainable Development Goals and maintain a circular economy. Digestate is a by-product of anaerobic digestion and can serve as a valuable source of nutrients, and micro- and macronutrients. It was established that its composition and properties are significantly influenced by the raw material and key parameters of anaerobic digestion, namely, the temperature regime, pH, process duration, organic load coefficient, and C/N ratio. In order to choose the most effective method of its processing, it is necessary to consider the above factors. The choice of processing methods was determined by the physical and chemical characteristics of digestate, the scale of production, and economic requirements. Physical processing methods are the most common, but biological methods are quite promising and require further research. Physical methods include mechanical dewatering with division into 2 fractions for use mainly as fertilisers in agriculture, biochar production, and extraction of nutrients from digestate. The main obstacle to the implementation of these methods is the unstable composition of digestate, its uncontrolled application as fertiliser, and more expensive and more difficult-to-operate equipment for the two extreme methods.

Biological processing methods include microalgae cultivation on liquid digestate fraction, composting, and vermicomposting. These methods make it possible to obtain valuable by-products such as fertilisers, food and feed additives, and co-substrates for re-fermentation. In addition, as a result of biological processing, the composition of digestate becomes more stable. The disadvantage of such methods is mainly the instability of the composition of digestate and the content of compounds in it in high concentrations, which become toxic to living organisms, complicating the process or completely inhibiting the processing. It is worth noting the combination of several technologies at the same time in order to create a closed production cycle with minimising the resulting waste. Thus, efficient processing of digestate is an important component of circular bioeconomics, contributes to the rational use of resources and reduces the environmental burden. Based on the analysis, further scientific research should be aimed at gradually integrating biological methods of digestate processing with a simultaneous combination of several technologies. The development of legislative regulatory mechanisms and legal instruments aimed at controlling the methods of digestate utilisation and processing is also important.

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Біотехнологічні аспекти утворення дигестату при метановому зброджуванні та його утилізації

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Анотація. Актуальність дослідження зумовлена стрімким розвитком біоенергетичної галузі, а саме біогазової сфери в Україні, що зумовлює утворення значних обсягів одного з побічних продуктів анаеробного зброджування – дигестату, що є цінним в багатьох аспектах. Метою статті було дослідження основних етапів метанового бродіння та чинників, що впливають на склад та якість дигестату, а також огляд традиційних та новітніх методів його утилізації. Аналіз наявної літератури показав, що сучасні способи зберігання утвореного дигестату є неефективними та виступають як тимчасовий спосіб його накопичення, враховуючи зростаючі з кожним роком та в подальшому майбутньому масштаби його утворення. У статті детально розглянуто кожен із чотирьох етапів анаеробного розщеплення (гідроліз, ацидогенез, ацетогенез, метаногенез) та охарактеризовано специфічні групи мікроорганізмів, які беруть участь у цих процесах. Визначено взаємозв'язок між складом вихідної сировини та кінцевими фізико-хімічними характеристиками утвореного дигестату. Представлено вплив складу вихідної сировини на особливості складу та основних характеристик утвореного дигестату. В роботі проаналізовано результати сучасних досліджень в галузі переробки та утилізації дигестату за допомогою біотехнологічних методів з метою забезпечення основних цілей сталого розвитку та дотримання принципів циркулярної економіки. Особлива увага приділялася ключовим характеристикам утвореного дигестату, що чинять переважний вплив на вибір подальших методів та технологій його утилізації. На основі проведеного аналізу інформацію було систематизовано та представлено у вигляді узагальнюючих таблиць. Результати цієї оглядової статті мають практичну цінність і можуть бути використані для підбору та впровадження технологій переробки дигестату на існуючих підприємствах

Ключові слова: зброджений залишок; анаеробне зброджування; переробка; біогаз; циркулярна економіка; сталий розвиток