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The efficiency of organic pollutants degradation in the process of anaerobic digestion of feedstocks with different origin

Andrii Paramonov*

Postgraduate Student
Sumy State University
40007, 116 Kharkivska Str., Sumy, Ukraine
https://orcid.org/0009-0009-2721-757X

Iryna Ablieieva

Doctor of Technical Sciences, Associate Professor
Sumy State University
40007, 116 Kharkivska Str., Sumy, Ukraine

Linköping University
SE-581, 83, Linköping, Sweden
https://orcid.org/0000-0002-2333-0024

Iryna Vaskina

PhD in Technical Sciences, Associate Professor
Poznan University of Life Sciences
60-637, 50 Wojsko Polskie Str., Poznan, Poland

Sumy State University
40007, 116 Kharkivska Str., Sumy, Ukraine
https://orcid.org/0000-0001-5904-9640

Anna Lysytska

Student
Sumy State University
40007, 116 Kharkivska Str., Sumy, Ukraine
https://orcid.org/0009-0001-5533-4660

Nataliia Makarenko

Forensic Expert
Sumy Research Expert Forensic Centre of the Ministry of Internal Affairs of Ukraine
40000, 8 Promyslova Str., Sumy, Ukraine
https://orcid.org/0009-0000-7653-1877

Abstract. The study of the degree of degradation of pollutants in the process of anaerobic digestion and their content in digestate is an urgent scientific issue driven by the necessity to confirm the safety of using this product as a biofertilizer to improve the quality of agricultural land. The aim of the article was to determine the efficiency of the degradation of pollutants in the process of anaerobic digestion with the use of various additional treatment methods. A meta-analysis was conducted to establish the patterns of pollutant degradation during anaerobic digestion under different conditions and with the application of additional process intensification technologies (ultrasound, the use of chemical reagents and carbon

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*Corresponding author

cloth, interspecies electron transfer, immobilisation of microalgae, etc.). The reliability of the selected hypotheses and the statistical significance of the obtained data were determined using statistical analysis methods. It was found that additional treatment methods allow to increase in the efficiency of degradation of organic pollutants in mixed organic waste by 180% (direct interspecies electron transfer), in animal waste by 115% (alkaline pre-treatment), and in wastewater sludge by 55% (by treating them in a thermophilic aerobic reactor together with anaerobic digestion). The efficiency of the immobilisation of microalgae technology, which provides for the removal of pollutants from the liquid fraction of digestate, is 135%. The presence of a synergistic effect of additional methods of feedstock treatment has also been established, which is illustrated by an increase in the amount of methane production by 98.88-261.56%, depending on the type of waste and the treatment method. The results obtained prove the high efficiency of additional treatment technologies. The practical significance of the results obtained lies in the proven synergistic effect of using additional methods of treatment of feedstocks and digestate due to an increase in the level of degradation of organic pollutants with the production of environmentally safe fertilizer and an increase in biogas yield

Keywords: biogas production; digestate; hazardous chemicals; organic waste; pre-treatment; sustainable development; substrate

Introduction

The issue of environmentally safe handling of organic waste is quite relevant in terms of environmental safety and environmental protection technologies. The technology of anaerobic digestion of waste for the production of biogas and digestate is becoming widespread in the world as a whole. Due to its physicochemical and microbiological properties, digestate has found extensive use as a fertilizer in European countries. For Ukraine, with its developed agriculture sector, the use of digestate as an organic or organo-mineral biofertilizer and an alternative to mineral fertilizers is justified from the point of view of environmental protection, conservation of natural resources and the economic effect obtained, which is especially relevant in the conditions of martial law. However, the digestate must meet quality and environmental safety standards, therefore, it is necessary to research the content of pollutants that were originally contained in the feedstock and to study the processes of their transformation during anaerobic digestion.

According to the research of K. Chojnacka & M. Chojnacki (2023), biogas technology is quite effective and promising in terms of solving the problem of waste utilisation with the production of useful products in the form of biogas and digestate, which fits into the framework of the circular economy and ensures the production of renewable energy sources. Biogas can be used for cogeneration of thermal and electricity, as well as in gas networks as a substitute for natural gas, which is quite relevant for Ukraine and is undergoing development. K. Pilarski *et al.* (2023) note that with the help of various biogas post-purification technologies to biomethane, it is used as fuel for various types of transport in Europe. T.M. Cabrita & M.T. Santos (2023) showed in their research that a wide range of organic feedstock can be used for anaerobic digestion. Substrates should be grouped by type according to their origin: animal waste (cattle manure, pig manure, chicken manure), plant waste (corn silage, energy crops, other lignocellulosic materials), food waste, and sewage sludge. Ye. Chernysh *et al.* (2021) analysed the trends regarding prospective feedstock for biogas production in Ukraine, based on the assessment of the actual generated waste volumes and forecasts for the future, it was established that animal husbandry waste together with plant biomass will occupy leading positions. At the same time, the practice of using sewage sludge for anaerobic digestion is not developed in Ukraine, although, as noted in the work of V. Vambol *et al.* (2022), it is a quite promising and environmentally safe way of handling these types of waste in Europe.

Various technological and operational approaches are employed to enhance biogas production, including the regulation of physicochemical parameters of the process with the identification of inhibitory factors. Among these, M. Kumar *et al.* (2023) investigated the pH of the medium, the content of volatile substances, and heavy metals. V. Ripoll *et al.* (2022) noted the effectiveness of using mixed feedstock to improve the quality of digestate. Simultaneously, E. Nordell *et al.* (2022) studied the positive impact of additional digestate treatment technologies, including thermal post-treatment of digestate with simultaneous pasteurisation on the organic matter content in digestate and biogas yield. A valuable by-product of biogas technology is digestate, which is effectively used as a biofertilizer. A.M. García-López *et al.* (2023) demonstrated in their research that digestate can increase crop yields, as well as restore soils and improve their properties. Research involving R. Bouaita *et al.* (2022) indicates that depending on the type of feedstock used for anaerobic digestion, digestate may contain certain pollutants. Therefore, the issue of the environmentally safe use of digestate as a biofertilizer arises, which is regulated by relevant directives and quality standards for organic fertilizers within each state or at the level of the European Union. However, L. Hammer & L. Palmowski (2021) noted that a significant portion of organic pollutants are degraded during anaerobic digestion, which contributes to a decrease in their concentration in digestate compared to the input feedstock. At the same time, the extent of degradation of these or other substances varies depending on factors such as the chemical nature of the substance, its concentration, operational-technological parameters of the process, microbiological activity, and environmental conditions in the bioreactor, the application of additional substrate pre-treatment and digestate post-treatment technologies. Moreover, the process of anaerobic digestion, having significant potential for the degradation or binding of pollutants, can be used for the purification of polluted environments and the environmentally safe disposal of waste.

These questions are of scientific interest and relevance to the problem being studied, as the level of environmental safety of digestate and compliance with quality standards will affect its scope of application and the use of its beneficial properties. The aim of the study was to establish the patterns and effectiveness of the degradation of various pollutants in the process of anaerobic digestion of organic feedstock. To achieve this goal, the research tasks were set as follows: to conduct a meta-analysis and systematise information on the content of organic pollutants in the substrate for anaerobic digestion; to investigate the effectiveness of the degradation of various organic pollutants during anaerobic digestion, as well as with the use of various additional technologies; to analyse the additional benefits of using additional technologies and provide appropriate recommendations for the directions of further experimental research. The scientific novelty of the obtained results lies in the determination, on the basis of the conducted meta-analysis, of the high efficiency of the direct interspecies electron transfer method in the process of anaerobic digestion of mixed organic waste and the method of immobilisation of microalgae during the post-treatment of the liquid fraction of digestate in relation to the removal of organic pollutants, as well as the pre-treatment of animal waste with strong alkali and alkaline hydrogen peroxide additionally to intensify their digestion.

Materials and Methods

The methodological basis of the study was chosen as a meta-analysis, which involved searching for scientific articles on the relevant topic, a comparative-descriptive analysis of the results of various studies to establish the patterns of degradation of pollutants in the process of anaerobic digestion under different conditions and with the use of selected additional technologies for process intensification. Statistical analysis of the obtained results was also conducted to determine the reliability of the chosen hypotheses and the statistical significance of the data. At the first stage of the study, a search for relevant scientific literature was conducted, and at the same time, the focus was on articles published in the period 2015-2024 and indexed by the international scientometric database Scopus, based on the following code.

TITLE-ABS-KEY ("organic pollutant*" AND degradation AND "anaerobic digestion")

AND (LIMIT-TO (DOCTYPE, "ar"))

AND (LIMIT-TO (LANGUAGE, "English"))

As a result of the search, 136 publications were found. After analysing each article, clarifying the direction of the research and the availability of the necessary results, the sample for meta-analysis was limited to 50 original articles. The frequency of occurrence of articles on the research topic, according to the search code and the results of the extended analysis, in individual journals ranged from 8 to 1. The number of journals with more than three articles published during the study period from 2015 to 2024 was six, as shown in Figure 1, constructed using the analytical tools of the Scopus database.

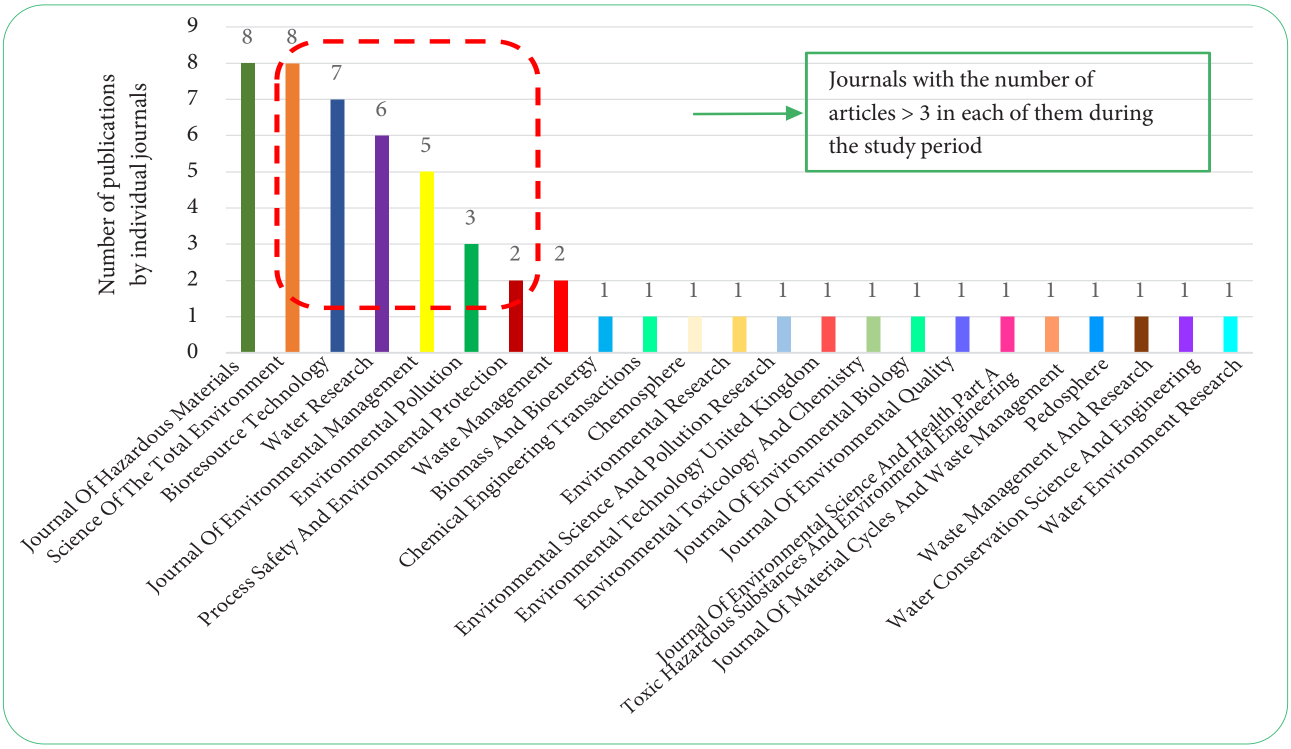

Figure 1. Quantitative distribution of articles by research topic in the context of individual journals indexed by the Scopus database for the period 2015-2024

Source: created by the authors

Based on the selected articles, a bibliometric network was constructed and visualised using the VOSviewer software (version 1.6.20). As a result of the analysis of the obtained network, clusters were identified by keywords describing four research directions (Table 1; Fig. 2). Further meta-analysis was conducted based on the identified clusters.

Table 1. Clusters of research areas on the degradation of pollutants during anaerobic digestion of waste according to keywords in articles published in Scopus in 2015-2024

Red cluster	Blue cluster	Green cluster	Yellow cluster
Anaerobic digestion of organic waste as one of the approaches to waste management (sewage sludge) and biotransformation of organic pollutants	The fate of organic pollutants during anaerobic digestion and the influence of various factors at each stage of the process	Factors, parameters and role of microbial activity for biodegradation of organic pollutants; toxicity of pollutants for metagenic bacteria	Mechanisms and patterns of organic pollutants degradation during the process of anaerobic digestion of feedstock

Source: created by the authors

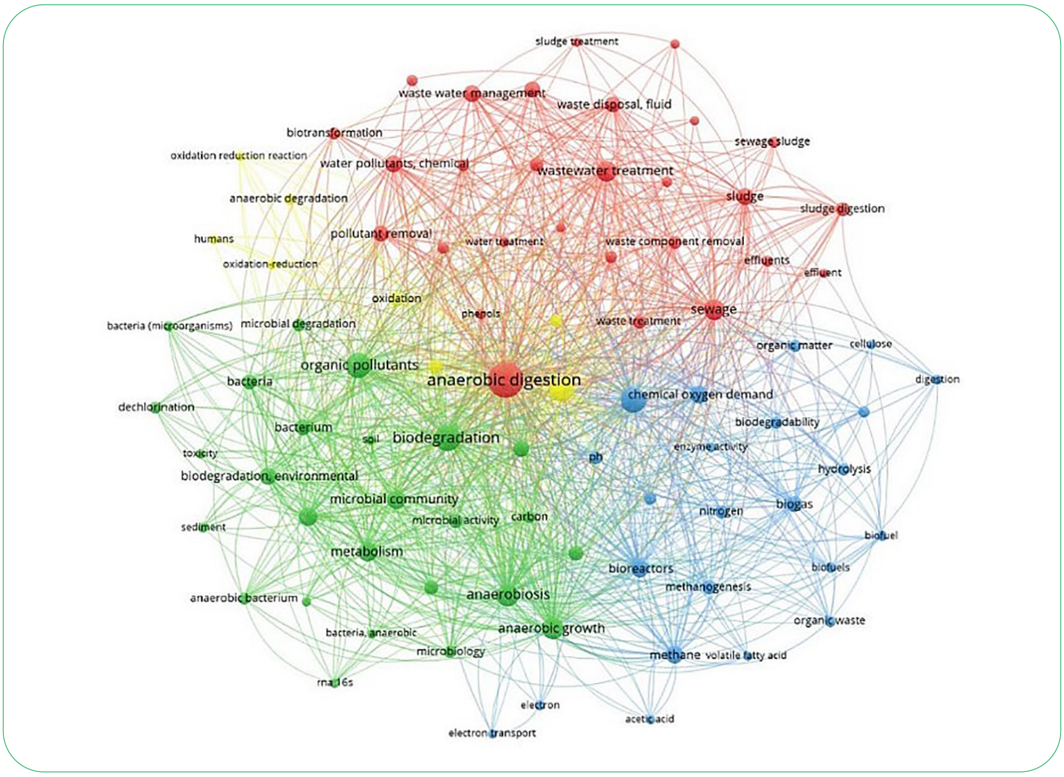

Figure 2. Visualisation of the bibliometric network based on the frequency of occurrence of > 5 keywords in articles on pollutant degradation during anaerobic digestion of organic waste published in Scopus in 2015-2024

Source: created by the authors

In the second stage of the research, based on the dataset obtained from the analysed articles, the level of removal of organic pollutants from the anaerobic digestion system was assessed, which allowed the establishment of the efficiency of substance degradation in anaerobic bioreactors. Within the framework of the analysis of the research results, the following independent factors were identified: substrate type, the chemical nature of the substance, and the peculiarity of the method (technology). The effectiveness of this process was evaluated by the absolute values of concentrations or by such an indicator as chemical oxygen demand (COD). The effectiveness of the removal (degradation) of organic pollutants in the process of anaerobic digestion of the substrate and the effectiveness of the use of additional technologies for the removal of substances in relation to anaerobic digestion (control) were evaluated in accordance with the formulas:

$$E_{rem} = \frac{C_d - C_s}{C_s} \times 100\%; \qquad (1)$$

$$E_t = \frac{C_d - C_{dt}}{C_s - C_{dt}} \times 100\%, \qquad (2)$$

where E_{rem} is the efficiency of removal (degradation) of organic pollutants in the anaerobic digestion process, %; C_d is the concentration of organic pollutants in the digestate, mg/g; C_s is the concentration of organic pollutants in the substrate, mg/g; E_t is the efficiency of applying additional technologies for substance removal relative to anaerobic digestion as a control, %; C_{dt} is the concentration of organic pollutants in the digestate when additional technology is applied, mg/g. The following technologies were subjected to the most thorough investigation. One of the studied technologies for the chemical pre-treatment of lignocellulosic feedstock in animal waste is alkaline treatment using strong alkali, such as NaOH and alkaline hydrogen peroxide. Alkaline pre-treatment ensures the breakdown of cellulose by an average of $14.8 \pm 0.1\%$ and prevents inhibition of anaerobic digestion by furan by-products, according to the research of J.R. Kim & K.G. Karthikeyan (2021). The method of direct interspecies electron transfer as an additional technology in the process of anaerobic digestion of substrates is used with the application of various conductive materials. This approach is based on extracellular electron exchange through physical contact between microorganisms, which replaces chemical indirect interspecies electron transfer (Hassanein & Lansing, 2022).

In the third stage of the research, the additional advantages of using technologies for the anaerobic degradation of organic pollutants were evaluated. In this process, the efficiency of anaerobic digestion for a particular type of substrate was determined based on the volume of methane produced. Theoretical values of the biomethane potential for various substrates, using literature data on the effectiveness of the aforementioned methods, were calculated using the approach described in by S. Weinrich *et al.* (2018). The elemental composition of substrates for anaerobic digestion was calculated as an average based on literature data. Statistical analysis of the dataset obtained from the meta-analysis was performed using the MS Excel and Statistical Package for the Social Sciences (SPSS) software (IBM SPSS Statistics, version 29.0.0.0). Data processing regarding the removal level of various groups of organic pollutants in the anaerobic digestion process of feedstocks of different origins and with the use of additional technologies was carried out using the analysis of variance (ANOVA) (Fu *et al.*, 2019). In this process, differences between data from different categories (substrate type, substance type, and type of additional technology or its absence) were evaluated. For post-hoc analysis, the Tukey honestly significant difference test (HSD) was used to establish a statistically significant difference between the data ($p < 0.05$). If the significance level value was $p < 0.05$, the null hypothesis, which assumed the absence of differences between the data, was rejected, and the difference between the results was accepted as statistically significant. For each value of the additional technology's efficiency compared to anaerobic digestion (control), the standard deviation was calculated.

Statistical processing of the data regarding the effectiveness of additional technologies in terms of biomethane potential was carried out using the chi-square method, which involved calculating the chi-square statistic with the consideration of the sample size and degrees of freedom (Aslam & Smarandache, 2023). The chi-square goodness-of-fit test uses sample data to test the hypothesis about the necessary proportions of the distribution of different values, and this test determines how well the obtained values within the sample correspond to the necessary proportions of values specified in the null hypothesis. In this process, the hypothesis about the distribution of technology efficiency over time depending on the stage of anaerobic digestion during the biomethane potential test was tested. The null hypothesis assumed that the distribution of effectiveness for the studied technologies over time is not uniform (normal). Calculations were performed based on the data obtained during the meta-analysis. The authors conducted scientific research within the framework of the Research Plan of the Department of Ecology and Environmental Protection Technologies of Sumy State University, Sumy, Ukraine, on the topics "Assessment of technogenic load in the region during changing the industrial infrastructure" (state registration No. 0121U114478) and "Reduction of technogenic load on the environment from oil production facilities: prospects for the use of biotechnology" (state registration No. 0121U114460) in accordance with the scientific and technical programme of the Ministry of Education and Science of Ukraine.

Results and Discussion

Characterisation of organic pollutants in groups of substrates used for anaerobic digestion

Substrates used for biogas production and derived from various waste origin contain certain contaminants that enter the feedstock through natural and anthropogenic pathways. Pollutants transfer into animal wastes from food products, including heavy metals and polycyclic aromatic hydrocarbons (PAHs) (Liu *et al.*, 2023b), or from animal husbandry practices (antibiotics, hormones, pharmaceuticals) as demonstrated in studies by X. Zhou *et al.* (2020) and L.G. Marutescu *et al.* (2022). Lignocellulosic materials can contain pesticides, heavy metals, and radionuclides due to the application of agrochemicals and mineral fertilizers. Additionally, during cultivation, transportation, processing, and other agricultural activities, films, woven bags, mesh covers, and some plastic pipes and plates are often used, and some of these may remain in agricultural waste. The study by O. Golovko *et al.* (2022) found that food waste contains heavy metals, pathogens, and organic micropollutants, including chemicals such as pharmaceuticals and pesticides, which raises concerns. Sewage sludge is the most contaminated feedstock for anaerobic digestion among other types of substrates (Gizaw *et al.*, 2024), as confirmed by the authors' literature search in the Scopus database. At the same time, this type of substrate is most often used in combination with other types of organic substrates of animal or plant origin (Malovanyy *et al.*, 2022) to balance the chemical composition of the feedstock and create favourable conditions for bacterial metabolism. The most toxic are persistent organic pollutants (Košnář *et al.*, 2023), which pose a high risk to human health (Beduk *et al.*, 2023). However, as mentioned above, pollutants are degraded in the process of anaerobic digestion, which completely or partially prevents their transition to digestate (Table 2).

Table 2. Characteristics of pollutants in feedstocks for anaerobic digestion of different origin

Type of substrate (feedstock for anaerobic digestion)	Pollutants	Presence in the digestate (yes/no)	Reference to the source of literature
Animal waste	Tetracycline, oxytetracycline, benzene, toluene	yes	Z. Liu *et al.* (2023b)
	Antibiotics	yes (10%)	W. Liu *et al.* (2023a)
	Sulfamethazine	yes	B.M. Oliveira *et al.* (2019)
	Tylosin	yes (10%)	A.G.D.O. Paranhos *et al.* (2022)
	Polychlorinated biphenyls (PCBs)	no	I. Višniauskė *et al.* (2018)
Sewage sludge	PAHs, PCBs, organochlorine pesticides (OCPs)	yes	Z. Košnář *et al.* (2023)
	Phenolic and fatty compounds from an oil	no	Y. Bouhia *et al.* (2023)
	Personal care products (fragrances, UV filters, antimicrobials, surfactants), PAHs and PCBs	yes	M. Biel-Maeso *et al.* (2019)
	Alkylphenols, phthalates, PAHs, pharmaceutical waste, hormones, perfluorinated acids, linear alkylbenzene sulfonates and PCBs	yes	D. Patureau *et al.* (2021)
	Pharmaceuticals, stimulants, antibiotics	yes	I. Gonzalez-Salgado *et al.* (2020)
	Pharmaceutical products	yes	A. Barreiro *et al.* (2022)
	PCBs and OCPs	yes	Z. Košnář *et al.* (2023)
	PCBs	yes	I. Višniauskė *et al.* (2018)
	Octocrylene	no	D. Fu *et al.* (2019)
	Microplastic (di(2-ethylhexyl) phthalate and phthalate)	no	N. Estoppey *et al.* (2024)
Agricultural residues and crops	PCBs	yes	K. Barcauskaitė (2019)
	Chlorophenol	yes	S.S. Ali *et al.* (2020)
	Hexachlorobenzene, humic acid	yes	C. Liu *et al.* (2021)
	Microplastic (di(2-ethylhexyl) phthalate and phthalate)	no	N. Estoppey *et al.* (2024)
Food waste	PLA plastic	yes	K.K. Porterfield *et al.* (2023)
	Microplastic (di(2-ethylhexyl) phthalate and phthalate)	yes	N. Estoppey *et al.* (2024)
	Bioplastics	yes	A. Kosheleva *et al.* (2023)

Source: created by the authors

Based on the data in Table 2, it was determined that there is a certain specificity regarding the content of various pollutants in digestate depending on the type of feedstock. Six groups of organic pollutants have been identified: antibiotics (sulfamethoxazole, trimethoprim, erythromycin, roxithromycin, cephalexin, ciprofloxacin, metronidazole, trimethoprim, sulphadoxine); personal care products (fragrances, UV filters, antimicrobial agents, surfactants); industrial chemicals (perfluorinated acids, linear alkylbenzene sulfonates, polycyclic aromatic hydrocarbons, polychlorinated biphenyls, bisphenol A, alkylphenols, phthalates, organochlorine compounds, azo dyes); pesticides (chlordane, dieldrin, diuron, endrin, heptachlor, hexachlorobenzene, pentachlorophenol); stimulants (caffeine); pharmaceuticals (anti-inflammatory drugs (ibuprofen, naproxen, and diclofenac), neuropsychiatric drugs (fluoxetine, carbamazepine, citalopram, and diazepam)); hormones (estrone, 17β-estradiol, and 17α-ethinylestradiol]). Moreover, a separate research area has established the application of the anaerobic digestion process for the purification of industrial wastewater contaminated with a wide range of organic compounds according to the specifics of the technological process. Given the wide range of pollutants present in the most common feedstock for anaerobic digestion in bioreactors aimed at biogas production, and the importance of environmentally safe valorisation of digestate in agriculture, there is an urgent necessity to research the degradation efficiency of these substances and assess their content levels in the final product. In addition, in the context of solving the problem of transformation of such a group of substances as persistent organic pollutants, in particular, pesticides, which have been found mainly in sewage sludge and agricultural feedstock (Table 2), the study of the possibility of using additional technologies is seen as necessary.

Removal and degradation of organic pollutants in the process of anaerobic digestion and with the use of additional technologies

Based on the conducted meta-analysis, anaerobic digestion was evaluated in terms of the efficiency of degrading various pollutants within specific substrates, which accordingly contributes to the purification of the feedstock and enables the production of environmentally safe fertilizers from digestate. Thus, a statistically significant high potential of anaerobic digestion was established for the removal of stimulants at the level of 100% and antibiotics (60%), an average level for pesticides and pharmaceuticals (30-50%), but a low level for personal care products and industrial chemicals (0-25%) on average for all types of substrates. Therefore, the challenge arises in finding additional technologies to intensify the degradation process of organic pollutants to reduce their concentration in the resulting digestate.

J.R. Kim & K.G. Karthikeyan (2021) found that strict pre-treatment conditions with strong alkali and alkaline hydrogen peroxide, as well as the impact of furane-derived lignocellulose by-products on cow manure, increased the anaerobic digestion efficiency of cow manure itself by 286%. At the same time, A.K. Rathankumar (2022) evaluated the high efficiency (up to 94%) of alkaline treatment of solid organic substrates contaminated with a wide range of concerning pollutants, in terms of removing 10 pesticides and 13 pharmaceuticals. Meanwhile, L. Wu *et al.* (2023) found that the use of direct interspecies electron transfer using various conductive materials increases the efficiency of anaerobic degradation of various persistent organic compounds in mixed organic waste by approximately 97%. At the same time, S. Mumtaz *et al.* (2023) determined the level of degradation of organic matter using the direct electron transfer method in a mixed substrate based on buffalo manure as the main co-substrate and plant waste as the additional one, and the efficiency of this process, evaluated as the level of COD removal, was 31.18%. In contrast, X. Lu *et al.* (2018) noted the positive effect of iron oxide-zeolite on the anaerobic digestion of complex organic waste, as a result of which the degradation efficiency increases by 74.8%. In addition, B.M. Oliveira *et al.* (2019) showed that the use of an additional carbon source in the form of glucose, fructose, sucrose, and meat extract during anaerobic digestion of waste contributes to an increase in biodegradation of the veterinary antimicrobial agent sulfamethazine from 39% to 54-61%, respectively.

It is worth noting that in the case of using anaerobic digestion for wastewater treatment, the effective application of additional technologies alongside the digestion process has been demonstrated. Y. Xiao *et al.* (2023) demonstrated the effectiveness of using titanium nanofibers for the degradation of organic pollutants in feedstock based on chicken manure and wheat straw waste, using the dynamics of volatile fatty acid indicators, COD, and decolourisation rate, the removal levels of which were 72.9%, 59.1%, and 66.8%, respectively. D. Feng *et al.* (2022) noted that the use of carbon cloth has a positive effect on the anaerobic digestion of high-concentration organic wastewater under different mixing conditions, increasing the efficiency of this process by 23%. K.-H. Phan *et al.* (2024) noted the high efficiency of anaerobic degradation of various azo dyes under thermophilic conditions, in particular, the removal of the azo dye mixture under thermophilic conditions was 6.7% higher than under mesophilic conditions. Instead, C. Liu *et al.* (2021) argue that the use of hydragric acrisol together with humic acid and urea increases the efficiency of anaerobic degradation of hexachlorobenzene by 71.3%. W. Gong *et al.* (2019) demonstrated the high efficiency (94.05%) of purification of the liquid fraction of digestate obtained after anaerobic digestion of waste using various technologies that involve the immobilisation of microalgae. In the study involving S.S. Ali *et al.* (2020), it was found that bacteria from the *Methanosataceae* family can have a positive effect on the anaerobic breakdown of woody biomass, as well as the detoxification of chlorophenols, increasing the efficiency of these processes by 113.7%.

Thus, the above-mentioned studies noted the positive role of applying various technologies in relation to increasing the efficiency of anaerobic degradation of a specific organic compound. Accordingly, there is a task of systematising and determining effective pre-treatment or post-treatment technologies depending on the type of feedstock used for anaerobic digestion. In the process of studying the effectiveness of degradation (removal) of organic pollutants, additional technologies are divided into 3 categories in relation to anaerobic digestion: pre-treatment of the substrate, compatible with digestion, and post-treatment of digestate (Fig. 3). When evaluating digestate post-treatment technologies, differences were found depending on the processed fraction.

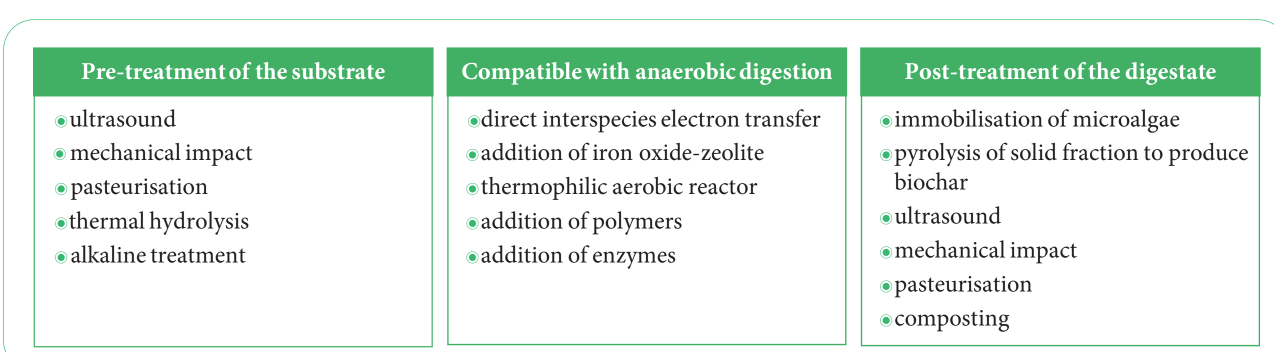

Figure 3. Systematisation of methods to intensify the process of pollutant degradation during anaerobic digestion of organic waste

Source: created by the authors

Based on the meta-analysis, the average efficiency of additional technologies for the removal of substances during the anaerobic digestion process itself was calculated. All the investigated methods and approaches were systematised within three types of fermentable substrates: mixed organic waste, waste of animal origin, and sewage sludge. The efficiency of specific methods was investigated for each type of substrate with the identification of the technology type, as per Figure 3, for the identified groups of organic pollutants: antibiotics; personal care products; industrial chemicals; pesticides; stimulants; and pharmaceuticals. Table 4 presents the results of the study on the identified methods and provides the effectiveness values of the relevant technologies in addressing the problem of anaerobic degradation of a complex of various organic compounds.

Table 4. Systematisation of methods that increase the efficiency of anaerobic degradation of a complex of different organic compounds

Type of substrate	Group of pollutants	Name of the applied technology	Type of technology	Technology efficiency*, %
Mixed organic waste	AB, IC	Direct interspecies electron transfer	Compatible with AD	180.00 ± 43.2
	P	Addition of iron oxide-zeolite		14.00 ± 4.3
		Immobilisation of microalgae	Post-treatment	135.00 ± 14.6
		Composting		31.15 ± 6.8
Waste of animal origin	PP, AB	Alkaline treatment	Pre-treatment	114.65 ± 28.7
		Pasteurisation	Pre- and post-treatment	−6.00 ± 5.5
Sewage sludge	PP, AB	Thermal hydrolysis	Pre-treatment	50.62 ± 12.2
		Mechanical impact		21.62 ± 6.9
		Ultrasound		20.33 ± 3.6
	PP, AB, S	Thermophilic aerobic reactor	Compatible with AD	54.76 ± 10.7
	PCP, IC, P, PP, S	Adding polymer		40.00 ± 4.8
	PP	Adding enzymes		5.03 ± 0.5
		Addition of enzymes + mechanical impact	Post-treatment	38.35 ± 4.7
		Mechanical impact		12.16 ± 3.5
		Ultrasound		10.36 ± 4.6

Notes: **M* ± *SD*; AB – antibiotics; PCP – personal care products; IC – industrial chemicals; P – pesticides; S – stimulants; PP – pharmaceutical products; AD – anaerobic digestion
Source: created by the authors

Thus, based on the results obtained (Table 4), the most effective additional technologies for the anaerobic degradation of various organic compounds were direct interspecies electron transfer using various conductive materials and immobilisation of microalgae, which are used for the degradation of mixed organic waste, and pre-treatment with strong alkali and alkaline hydrogen peroxide for waste of animal origin.

Synergistic effect of anaerobic digestion intensification of organic feedstock for pollutant degradation and increasing biogas yield

Among the studied methods for substance degradation, some also showed effectiveness in increasing biogas yield, particularly direct interspecies electron transfer using various conductive materials and alkaline pre-treatment. Based on a theoretical calculation using averaged literature data, it was established that 1 kg of mixed organic waste produces an average of 400 l of biomethane during normal anaerobic digestion. Whereas, with the application of the direct interspecies electron transfer method, the biomethane yield from 1 kg of this waste increases to an average of 790 l, i.e., by 98.88%, which almost corresponds to the data in the study by L. Wu *et al.* (2023). Meanwhile, 1 kg of cow manure typically yields an average of 295 l of biomethane. However, with the alkaline pre-treatment of waste of animal origin, the biomethane yield from 1 kg of cow manure increases to an average of 1,140 l, i.e., by 261.56%, which is consistent with the data presented in the study by J.R. Kim & K.G. Karthikeyan (2021). Based on the literature data within the conducted meta-analysis regarding the cumulative biomethane potential for anaerobic digestion alone (control) and with the application of direct interspecies electron transfer using various conductive materials and alkaline pre-treatment, graphs of the dependence of technology efficiency on time were constructed (Fig. 4-5).

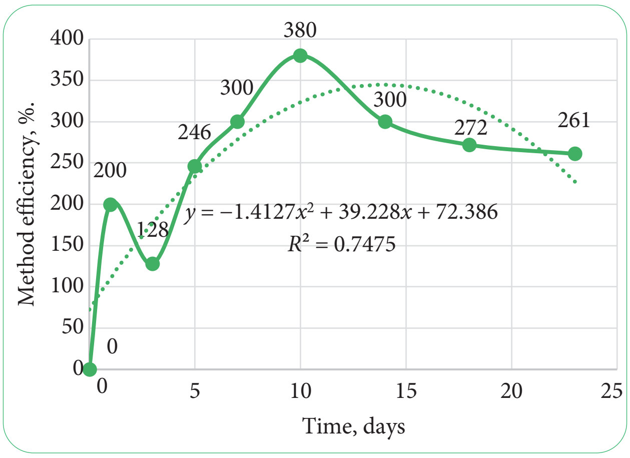

Figure 4. The efficiency of the method of pre-treatment with strong alkali and alkaline hydrogen peroxide in anaerobic digestion of cow manure in relation to time
Source: created by the authors

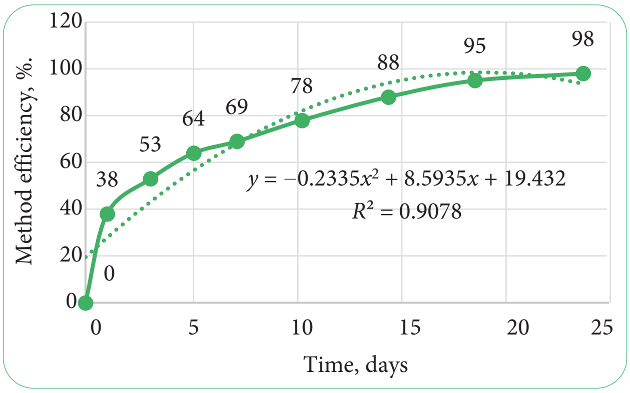

Figure 5. Efficiency of the method of direct interspecies electron transfer in the anaerobic digestion of mixed organic waste in relation to time
Source: created by the authors

During the anaerobic digestion of animal waste with alkaline treatment (Fig. 4), the cellulose in the lignocellulosic biomass of cow manure is broken down. Accordingly, a higher biomethane potential is observed. However, a sharp increase in efficiency is noted on Day 1 followed by a decrease on Day 3. This may be associated with the formation of a bacterial consortium involved in hydrolysis and the facilitation of lignocellulosic biomass hydrolysis treated with alkali. This is followed by a gradual increase in efficiency up to Day 10, followed by a decrease to Day 23. This pattern may be due to the increased availability of nutrients for bacteria, followed by gradual inhibition by volatile compounds and a change in the pH of the medium. The application of the direct interspecies electron transfer method during anaerobic digestion of mixed organic waste (Fig. 5) is implemented simultaneously with anaerobic digestion, unlike the previous one. Therefore, the patterns of efficiency distribution over time differ. There is a gradual increase in the efficiency of the technology in terms of biomethane potential without significant fluctuations, as the method's impact is manifested at all stages of digestion in a cumulative manner. However, the efficiency in terms of biomethane yield is lower compared to alkaline pre-treatment, although the results were the opposite for the degradation of organic pollutants.

By performing statistical analysis using the chi-square method, it was found that all the results obtained were reliable and accurate. The chi-square value for the increase in methane yield when using the direct interspecies electron transfer method during anaerobic digestion of mixed organic waste is 0.13, which is less than the chi-square value of 16.919 ($p > 0.05$) and indicates that the null hypothesis is accepted and the deviations are quite small. At the same time, the chi-square value for the increase in methane yield during anaerobic digestion of cow manure, pre-treated with strong alkali and alkaline hydrogen peroxide, is 0.15, which is also less than the chi-square value of 16.919, so the null hypothesis is accepted and the deviations are quite small. Thus, the results are reliable and quite accurate and indicate that the use of different additional technologies during the anaerobic digestion of different substrates does indeed increase the efficiency of the anaerobic digestion process itself for these substrates.

Based on the conducted meta-analysis, it was established that the application of additional technologies allows not only to increase in the efficiency of pollutant degradation during the anaerobic digestion of feedstock but also to increase in biogas yield. This pattern is associated with the intensification of anaerobic degradation of organic feedstock. As shown in the study by E. Nordell *et al.* (2022), thermal post-treatment of digestate with simultaneous pasteurisation can achieve more complete conversion and mineralisation of organic matter, thereby increasing the volume of biogas produced. Physical-mechanical pre-treatment methods such as grinding (Karuppiah & Azariah, 2019) or extrusion (Olatunji *et al.*, 2021) improve the porosity, flowability, and bulk density of the substrate. All this increases the surface area of fermentable particles, promoting a more efficient breakdown of organic components while simultaneously boosting biogas yield, as noted in the study by K. Obileke *et al.* (2024). The principle behind these studied methods involves biochemical or physical impacts on the structure of organic compounds, leading to the breakdown of chemical bonds and the formation of new substances, including inorganic ones, as highlighted by B. Lee *et al.* (2019) and T. Llano *et al.* (2021). These processes form the basis for the mineralisation of organic matter, as a result of which methane and carbon dioxide are formed in anaerobic conditions as the main components of biogas. Therefore, the transformation of organic matter leads to a synergistic effect in relation to the transformation of pollutants and biogas yield.

Another positive effect of applying digestate post-treatment to intensify the degradation of polluting organic matter is the production of other useful products.

It has been established that the most promising and environmentally safe product is biochar, obtained as a result of pyrolysis of the solid fraction of digestate, which is consistent with the results of research by S. Ghysels *et al.* (2022) on an integrated system of anaerobic digestion and slow pyrolysis of mixed feedstock of plant and animal origin. A systematic approach to the use of biochar involves the purification of the liquid fraction of digestate using biochar, in which the content of pollutants is reduced by more than 95% due to pyrolysis. This approach is justified, since according to the results of the study by A.M. Ali *et al.* (2019) on the content of new organic pollutants in feedstock of various origins (mainly sewage sludge, food waste, and mixed feedstock based on food waste) and two fractions of digestate, sun protective octocrylene and acetaminophen (paracetamol) were found in the highest concentrations in the liquid digestate, while octocrylene and antipyrine TCPP (tris(1-chloro-2-propyl) phosphate) were identified in the highest concentrations in the solid digestate. In addition, biochar can be used for bioremediation of soils contaminated with heavy metals due to the sorbing properties of the porous structure of biochar. On the other hand, digestate binds heavy metals in the soil, based on the complexing properties of organic matter, depending on the ratio of fluorescent components, as shown by the authors P. Skvortsova *et al.* (2024).

It is worth noting that along with traditional, widely used substrates for anaerobic digestion, grouped according to different categories of organic waste, unexplored feedstock, such as coal, also has significant biomethane potential, as shown in the study by P.H. Fallgren *et al.* (2021). In addition, understudied and local feedstock is of particular interest, as its use for biogas and digestate production is a promising way to handle specific types of organic waste, such as sugarcane and solid household waste (Tshemese *et al.*, 2023). The high potential of the fertilizing properties of digestate from local plant feedstock was noted by the authors W. Czekała *et al.* (2020), which ensures compliance with the principle of nutrient recovery in the circular economy and has a positive impact on greenhouse gas emissions (Kowalczyk-Juśko *et al.*, 2023). In addition, the production of biogas from solid household waste landfills is becoming promising due to the installation of degassing systems, as shown in a recent study by I. Vaskina *et al.* (2024). Since this type of waste also contains a wide range of pollutants, the possibility of using the developed technologies increases the scientific interest and practical significance of the research results in this direction. Moreover, mixtures of different wastes used for co-anaerobic digestion, as shown above, differ in the content of pollutants compared to mono-feedstock. Accordingly, the processes of pollutant degradation during anaerobic digestion will have their own specifics due to different microbiological and physicochemical conditions, which require further research and may be one of the directions for further research.

Thus, as a result of the meta-analysis conducted, it was established that the feedstock used for anaerobic digestion to obtain biogas is usually formed from waste with an organic component, which determines the presence of a wide range of pollutants in it. The anaerobic digestion process itself rarely ensures complete degradation of the corresponding groups of pollutants according to their biodegradability and resistance, therefore additional methods of intensification of anaerobic degradation are quite relevant. The maximum synergistic effect for the degradation of pollutants and the increase in biomethane yield was established in the case of the use of direct interspecies electron transfer methods on the example of feedstock from mixed organic waste and alkaline pre-treatment of animal waste. However, these methods require testing on other types of substrates with different properties, chemical composition, and the nature of the impact on the process parameters, including microbiological ones.

Conclusions

The analysed methods for improving the efficiency of anaerobic digestion of different substrates have shown their effectiveness, indicating their great importance and potential for application in biotechnology. The main substrates that are most often contaminated with various organic substances and require the application of effective technologies to improve the result of their anaerobic digestion are sewage sludge, animal waste, and mixed organic waste. As a result of the conducted research, it was found that such a method as direct interspecies electron transfer allows increasing the efficiency of pollutant degradation in mixed organic waste by 180%. In the case of animal waste, it is worth applying alkaline treatment, which will increase the efficiency of degradation of organic pollutants by almost 115%. The degradation of organic compounds in sewage sludge can be increased by 55% by treating it in a thermophilic aerobic reactor in combination with anaerobic digestion. Additional treatment technologies allow not only to ensure the effective degradation of pollutants during fermentation but also to increase the overall efficiency of anaerobic digestion by increasing methane yield. The results obtained indicate that when using direct interspecies electron transfer from mixed organic waste, it is possible to obtain 98.88% more methane, while pre-treatment of animal waste with strong alkali and alkaline hydrogen peroxide makes it possible to obtain 261.56% more methane from this waste. This significantly increases not only the environmental but also the economic component of the application of these treatment technologies.

The results of the study have scientific and practical significance, as they demonstrate the high efficiency and synergistic effect of additional technologies for improving the process of anaerobic digestion and increasing the degree of degradation of organic matter contained in the three most common substrates (mixed organic waste, animal husbandry waste, and sewage sludge). At the same time, this allows for identifying directions for further research aimed at evaluating the efficiency of the studied technologies for environmentally safe management of local feedstock in the context of waste utilisation for bioenergy purposes with the production of biogas and in agroecological purposes through the use of digestate as biofertilizer. Further research will be aimed at developing a laboratory stand and testing the investigated methods on local feedstock with the determination of the efficiency of degradation of organic pollutants for the production of environmentally safe fertilizer from digestate, which will ensure the prevention of chemical pollution of environmental components, including soils.

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Conflict of Interest

None.

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Ефективність руйнування органічних забруднювальних речовин у процесі анаеробного зброджування сировини різного генезису

Андрій Парамонов

Аспірант
Сумський державний університет
40007, вул. Харківська, 116, м. Суми, Україна
https://orcid.org/0009-0009-2721-757X

Ірина Аблєєва

Доктор технічних наук, доцент
Сумський державний університет
40007, вул. Харківська, 116, м. Суми, Україна

Університет Лінчепінгу
SE-581, 83, м. Лінчепінг, Швеція
https://orcid.org/0000-0002-2333-0024

Ірина Васькіна

Кандидат технічних наук, доцент
Природничий університет у Познані
60-637, вул. Війська Польського, 50, м. Познань, Польща

Сумський державний університет
40007, вул. Харківська, 116, м. Суми, Україна
https://orcid.org/0000-0001-5904-9640

Анна Лисицька

Студент
Сумський державний університет
40007, вул. Харківська, 116, м. Суми, Україна
https://orcid.org/0009-0001-5533-4660

Наталія Макаренко

Судовий експерт
Сумський науково-дослідний експертно-криміналістичний центр Міністерства внутрішніх справ України
40000, вул. Промислова, 8, м. Суми, Україна
https://orcid.org/0009-0000-7653-1877

Анотація. Дослідження ступеня деградації забруднювальних речовин у процесі анаеробного зброджування та їх вмісту в дигестаті є актуальним науковим питанням, яке продиктовано необхідністю підтвердження безпечності застосування цього продукту як біодобрива для покращення якості сільськогосподарських земель. Мета статті – визначення ефективності руйнування забруднювальних речовин у процесі анаеробного зброджування із застосуванням різних методів додаткової обробки. Для встановлення закономірностей деградації забруднювальних речовин у процесі анаеробного зброджування за різних умов та зі застосуванням додаткових технологій інтенсифікації процесу (ультразвук, застосування хімічних реагентів та вугільної тканини, міжвидове перенесення електронів, іммобілізація мікроводоростей тощо) було застосовано мета-аналіз. Визначення достовірності обраних гіпотез та статистичної значущості отриманих даних було проведено методами статистичного аналізу. Було визначено, що додаткові методи обробки дозволяють підвищити ефективність деградації органічних забруднювачів у змішаних органічних відходах на 180 % (пряме міжвидове перенесення електронів), у відходах тваринного походження – на 115 % (лужне попереднє оброблення), а в осадах стічних вод – на 55 % (шляхом їх обробки в термофільному аеробному реакторі сумісно з анаеробним зброджуванням). Ефективність технології іммобілізації мікроводоростей, що забезпечує видалення забруднювальних речовин із рідкої фракції дигестату, становить 135 %. Також було встановлено наявність синергетичного ефекту додаткових методів оброблення сировини, що ілюструється зростанням кількості виділеного метану на 98,88-261,56 % залежно від виду відходів і методу обробки. Отримані результати доводять високу ефективність додаткових технологій обробки. Практичне значення одержаних результатів полягає в доведеному синергетичному ефекті від застосування додаткових методів обробки сировини та дигестату за рахунок підвищення рівня деградації органічних забруднювачів з одержанням екологічно безпечного добрива та збільшенням виходу біогазу

Ключові слова: виробництво біогазу; дигестат; небезпечні хімічні речовини; органічні відходи; попередня обробка; стійкий розвиток; субстрат