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Multi-phase flow assessment for the fermentation process in mono-substrate reactor with skeleton bed

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Abstract

The selected techniques were reviewed and their technological aspects were characterized in the context of multi-phase flow for biogas production. The conditions of anaerobic fermentation for pig slurry in a mono-substrate reactor with skeleton bed were analysed. The required technical and technological criteria for producing raw biogas were indicated.

Design and construction of the mono-substrate model, biogas flow reactor, developed for cooperation with livestock buildings of various sizes and power from 2.5 kW to 40 kW. The installation has the form of a sealed fermentation tank filled with a skeletal deposit constituting a peculiar spatial system with regular shapes and a rough surface.

Incorporating a plant in such a production cycle that enables the entire slurry stream to be directed from the cowshed or pig house under run channels to the reactor operating in the flow mode, where anaerobic digestion will take place, allows to obtain a biogas.

The paper presents preliminary results of experimental investigations in the field of hydrodynamic substrate mixing system for biogas flow assessment by the adhesive bed in the context of biogas production. The aim of the study was to assess and shows the influence of the Reynolds number on the biogas resistance factor for the fermentation process in mono-substrate reactor with adhesive deposit. The measurement results indicate a clear effect of the Reynolds number in relation to the descending flow resistance coefficient for the adhesive bed.

Key words: *fermentation process, mono-substrate reactor, multi-phase flow, skeleton bed*

INTRODUCTION

FERMENTATION PHASES

Biological processes have been used in the environmental protection for many years and their rapid development dates back to the application of many other disciplines – microbiology, biochemistry, process engineering and genetic engineering. To emphasize qualitative changes made by the knowledge development, this field of the application of biological processes in the environmental protection is considered as environmental biotechnology. Since a fundamental condition that needs to be met to consider a process as biotechnology is to control it on a technical scale, it is also justified to use the environmental engineering term [KOSTER, LETTINGA 1985].

The widespread of fermentation methods [CZEKAŁA *et al.* 2017; MIKOŁAJCZAK *et al.* 2009; SKOWRON *et al.* 2015] is primarily associated with the solution of the problem of the slowness of multiplication of bacteria in this process. This chiefly refers to methanogenic bacteria responsible for the last but most fundamental stage of fermentation. This problem was partially solved through the technological separation of two fundamental stages of fermentation. The first stage involves quick hydrolysis phases – acido and acidogenic, whereas the latter one includes the methane phase. This solution also enabled decreasing a risk to stability of the process as a result of the accumulation of products of the first phases, which act as a brake on the last phase. However, the greatest progress resulted from the effective acquisition of the population of microorganisms

with small sedimentation properties or – thanks to their granular form or the application of media for the microbial immobilization. This allowed to significantly increase their concentration in the bioreactor and to make the process independent from the time of microbial generation. The technique of obtaining active biocenosis by simple selection through subsequent portioning processes was also mastered. However, a certain limitation of the widespread use of fermentation is its high susceptibility to fluctuations of environmental factors:

- inoculation with selected bacterial strains producing enzymes for cellulose hydrolysis,
- immobilization of bacteria to ferment the substrate without leaching, and temperature.

STRUCTURE OF MEDIA MATERIAL

Immobilization from Latin “immobilia”, i.e. immobile, is a partial or complete restriction of the free movement of microorganisms while ensuring that they have access to nutrients and the outflow of transformation products.

The medium should be insoluble in an environment where the microorganisms will be immobilized. Resistance to chemical and microbiological degradation is important. The chemical properties also determine the possibilities of sterilization and purification, the so-called hygienization. In addition, the presence of appropriate function groups may affect the adhesion of bacteria cells to the medium surface. The coating of the matrix by microorganisms may clog pores and significantly reduce its permeability. The usefulness of the material is also affected by its price, availability, industrial-scale regeneration and application possibilities, simplicity and biological parameters during immobilization.

The pore structure is determined by its size, distribution and depth, which in the medium determines the range of interaction between immobilized bacterial cells and the external environment – the area inside the bioreactor. According to the International Union of Pure and Applied Chemistry (IUPAC) classification, pore media are defined as microporous (diameter of pores below 2 nm), mesoporous (diameter between 2 and 50 nm) and macroporous (diameter above 50 nm). Only small particles such as oxygen can penetrate microporous materials. However, many of the materials used have an irregular structure and pore size, and in addition their properties may change in the environment. Pores should allow the free movement of nutrients and transformation products, but not the bacteria cells themselves. Transport of the substance along the matrix of the media is possible due to a gradient of concentrations [SCHREZENMEIR *et al.* 1994].

Bacterial cells with higher nutritional requirements should be found in very well permeable materials [LANZA *et al.* 1999].

Suitable bacterial strains or cell lines that produce particles for a long period of time or are able to perform specific functions in a continuous process, are suitable as biological material for immobilization. There may be used primary, stable, allogenic, and xenogenic lines.

When selecting the appropriate biological material, consideration should be given to its nutritional requirements, stress resistance and morphological variability. It is important to make sure that the bacteria cells that we apply have an advantage over eukaryotic cell lines. They are usually cheaper in breeding, more resistant to stress, less demanding in outdoor conditions, and can be active for a longer period. They are also simpler models for modification. Genetically modified microorganisms, designed to produce the desired agents, can become a future. Bacteriophages may also be irreplaceable for certain types of processes. Biosafety is a very important aspect. As for genetically modified organisms, the gene expression should be regulated on a tiered basis and the strains themselves should be highly genetically stable [BAKUŁA *et al.* 2013].

METHODS AND TECHNIQUES OF IMMOBILIZATION

Microbial of immobilization may be classified as follows (Tab. 1).

Table 1. Immobilization of microorganisms

Methods	On surface carrier	Inside carrier	Without carrier
Techniques	adsorption	inclusion (trapping)	spatial networking
	adhesion	encapsulation: (i) nano and micro (capsule) (ii) macro (capillary)	flocculation: (i) induced, (ii) natural (self-aggregation)
	covalent bonds		

Source: own elaboration.

Adsorption and adhesion as well as covalent bonding are distinguished within the immobilization methods on the media surface. Adsorption and adhesion are based on ionic, hydrogen, hydrophobic, electrostatic, van der Waals forces or a combination of these forces.

The material is immobilized inside the medium by physically closing the cells in the matrix. Microorganisms can be sealed inside a semi-permeable membrane in the form of a capsule (nano-encapsulation and micro-encapsulation) or a capillary (macro-encapsulation) or trapped in a porous medium [ULUDAG *et al.* 2000].

Trapping (inclusion) involves entrapment of cells in a three-dimensional matrix whose size is significantly larger than the size of bacterial cells – these are usually balls with a diameter of 0.3–3.0 mm [BONIN 2008].

The encapsulation consists in covering the core with walls formed from one or more covering substances [ARSHADY 1993]. The core usually represents (10–90)% of the total capsule mass, which may be a solid, liquid or gaseous substance or mixture. Microcapsules are spherical and have a diameter of 0.2–5000 μm . The most common methods used for their production are those involving the modification of full gel capsules. Macrocapsules also called capillary membranes have a cylindrical shape, internal diameters of 0.5–1.5 mm and length of 1–10 cm [JASIŃSKI *et al.* 2006]. Capillary membranes are manufactured, among others, from polypropylene, polymer from polyolefins group.

The immobilization without the medium is based on either natural (self-aggregation) or induced cell ability to form clusters (induced cell flocculation and spatial cross-linking). Spatial crosslinking is the binding of bacterial cells with different substances that may react with functional groups of bacterial cell shields. Mutual crosslinking usually results in relatively stable biomaterials, but it may lead to a partial loss of microbial activity and hinder the diffusion of substrates. Self-aggregation is possible due to the secretion of compounds by cells, which allow them to grow in the form of flakes or granules (e.g. polymucosaccharides), and the formation of clusters is facilitated by a high concentration of biomass. The ability of the cells to interconnect may increase, e.g. by adjusting pH, temperature and by adding polyelectrolytes.

Manipulation of the aforesaid factors to enable the formation of clusters and conglomerates is called the induced flocculation. However, the biological material obtained in both self-aggregation and flocculation processes is characterized by low mechanical strength, which limits the possibilities of its application.

BIOFILM AS UNIVERSAL MEDIUM

Environmental micro-organisms rarely occur in the form of single dispersed cells, i.e. the so-called "plankton" [DONLAN 2002]. They rather show a tendency to adsorption at the phase boundary between solid-liquid, liquid-gas, or liquid-liquid. Most often, they are aggregates called biofilm (or a biological membrane) adhering to solid surfaces or surfaces of cells of other organisms [COSTERTON *et al.* 1994; 1995]. The biofilm is a multi-cellular creature composed of micro-organisms of one or more species or genera [CHANDRA *et al.* 2005].

Both autotrophic and heterotrophic microorganisms, including saprophyte and pathogenic microorganisms, have the ability to form the biofilm. In addition to bacteria, it may also include fungi, algae or protozoans [CURRIE 2001]. The colonization of various surfaces by microorganisms is possible due to their adhesion properties, and the structure of the resulting biofilm is stabilized by polymeric substances secreted extracellularly, the so-called EPS (extracellular polymeric substances). The biofilm comprises complex, multicellular structures in which numerous microbial cells are surrounded by a mucous layer [MONDS, O'TOOL 2009]. The cells of microorganisms included in the biofilm are characterized by specialization to perform various functions and show different characteristics than the cells living in free form. The design of these aggregates protects micro-organisms from the adverse effects of external factors and creates opportunities for easier nutrient availability. Therefore, the biofilm may function under conditions where the survival of individual cells would be difficult and in many cases even impossible [FURWICZ *et al.* 2010].

Microorganisms have developed mechanisms to be able to utilize hydrocarbons as a digestive substrate [KOŁWZAN 2008; KOŁWZAN *et al.* 2005]. These mechanisms allow microorganisms to decompose suspended hydrocarbons in the form of fine drops (micelles) in aqueous

solutions. Bio-surfactants produced by microorganisms increase the availability of water-insoluble substrates and in their presence these substances are transferred to micelles and distributed on the cell surface. Substrates that are solids are moistened and dispersed, increasing their surface area. This, in turn, enables their colonization by microorganisms. The biodegradation of hydrocarbon leads numerous species of bacteria and fungi. The consequence of the performance of microorganisms degrading hydrocarbons is problems with the operation of multiple devices as a result of the change in the properties of physical-chemical substrates.

The aim of the study was to assessment and shows the influence of the Reynolds number on the biogas resistance factor for the fermentation process in mono-substrate reactor with adhesive deposit.

MATERIALS AND METHODS

EXPERIMENTS

In Institute of Technology and Life Sciences, Department of Renewable Energy Sources, Poznan Branch (Pol. Instytut Technologiczno-Przyrodniczy, Zakład Odnawialnych Źródeł Energii, Oddział w Poznaniu) has developed system of installations with a power of 3–40 kW. It is a pilot installation with an active capacity of 15 m³ of fermenter (Photo 1) was located in a farm with 1100 fatteners (pigs) kept in a grate system.



Photo 1. Pilot installation – examples of techniques of the anaerobic fermentation of waste – mono-substrate reactor (phot. G. Wałowski)

The raw biogas production unit is a biogas transport system produced in the fermentation tank with its equipment and enables the fermentation process to be carried out, controlled and adjusted (Fig. 1). The fermentation tank is the main and key component of the system for achieving the maximum efficiency of the fermentation technology.

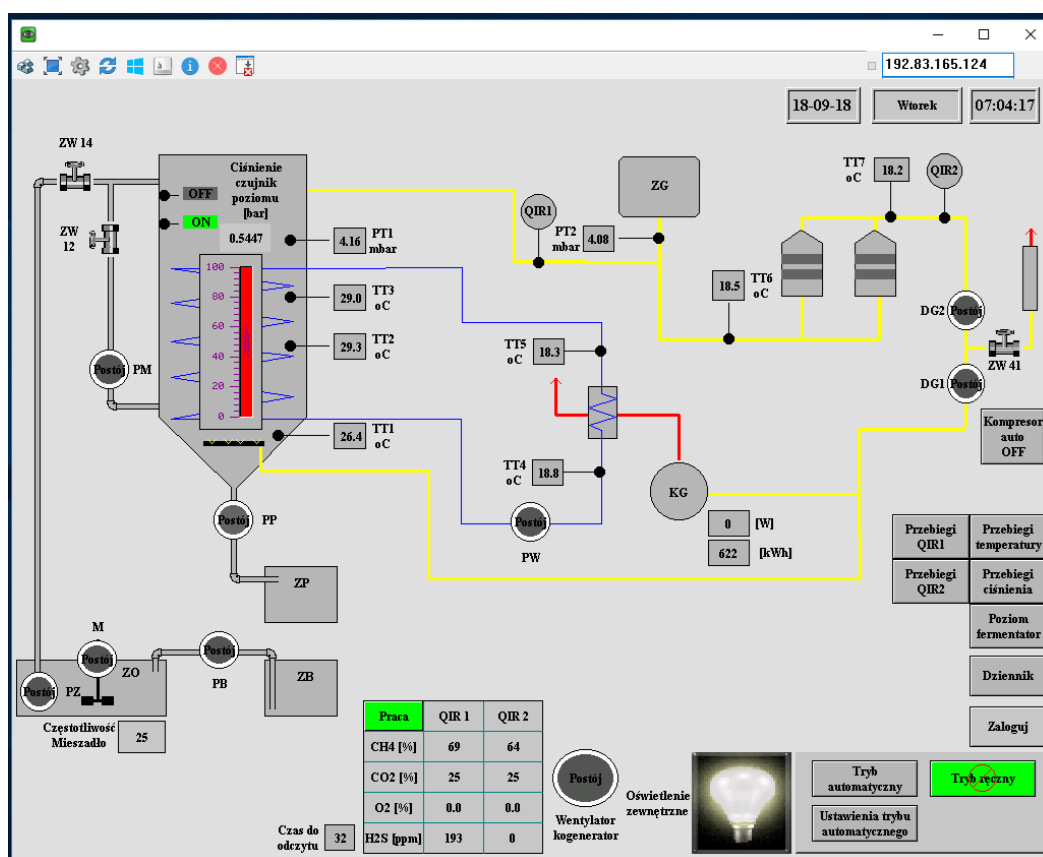


Fig. 1. Synoptic board of the biogas plant control and visualization system (Ultra VNC software): DG = gas blower, KG = co-generator, M = agitator, PB = biomass pump, PM = mixing pump, PP = digestate pump, PW = water pump, PT = pressure, PZ = submersible pump, QIR = gas analyser, TT = temperature, ZB = biomass tank, ZG = gas tank, ZO = operating tank, ZP = digestate tank, ZW = valve; the meaning of the descriptions in the diagram: “Postój” = “Stop”; “Częstotliwość Mieszadło” = “Frequency Stirrer”; “Ciśnienie czujnik poziomu [bar]” = “Pressure level sensor [bar]”; “Czas do odczytu” = “Time to read”; “Praca” = “Job”; “Wentylator kogenerator” = “Co-generator fan”; “Oświetlenie zewnętrzne” = “External lighting”; “Tryb automatyczny” = “Automatic mode”; “Ustawienia trybu automatycznego” = “Automatic mode settings”; “Wtorek” = “Tuesday”; “Przebiegi QIR1” = “Waveforms QIR1”; “Przebiegi QIR2” = “Waveforms QIR2”; “Tryb ręczny” = “Manual mode”; “Kompresor auto OFF” = “Auto compressor OFF”; “Przebiegi temperatury” = “Temperature waveforms”; “Przebiegi ciśnienia” = “Pressure waveforms”; “Poziom fermentator” = “Fermenter level”; “Dziennik” = “Diary”; “Zaloguj” = “Log in”; source: own elaboration

The experimental research concerned a measurement system for the assessment of the amount of biogas under the conditions of the biogas production process. The research was carried out in the field of biogas flow rate measurement resulting from the reference pressure in the fermenter. An independent assessment of the amount of biogas and the pressure drop on the skeletal deposit was carried out.

The basis for the assessment of hydrodynamics of gas flow through the adhesive bed is the flow characteristic that results from the pressure forcing this flow. In each case, the determination of this characteristic consists in determining the impact of the biogas stream on the value of this overpressure, equivalent to a pressure drop – this is tantamount to determining the total resistance of biogas flow through the adhesive bed.

RESULTS AND DISCUSSION

The raw biogas production system consists of the following unit elements. The digestion tank is designed for a system in a vertical position. The bottom of the tank is

truncated cone shaped with a centrally located trigger. The fermentation tank is sealed by a lid that closes the fermenter with a sealing element.

Circulation system – description of the system operation. The fermentation tank is filled with biomass from the top to ensure directional migration of the fraction through the entire system. The biomass vertical circulation system and the freshly extracted biogas circulation system are used for mixing the fermentation tank content. The biomass mixing system ensures the homogenization of the composition and temperature of the ferment as well as delivery of certain ingredients supporting the fermentation process. Mixing the content of the fermenter for averaging its composition is performed by means of a barbotage. This is done in such a way that part of the biogas is extracted from the fermenter gas compartment by means of a blower and fed through a non-return valve into the lower part of the bioreactor via a bubbler system. Gas flows out of the bubblers in the form of bubbles and mixes the suspension upwards.

Immobilization system – description of the system operation. Inside the fermentation tank there is a fill, i.e.

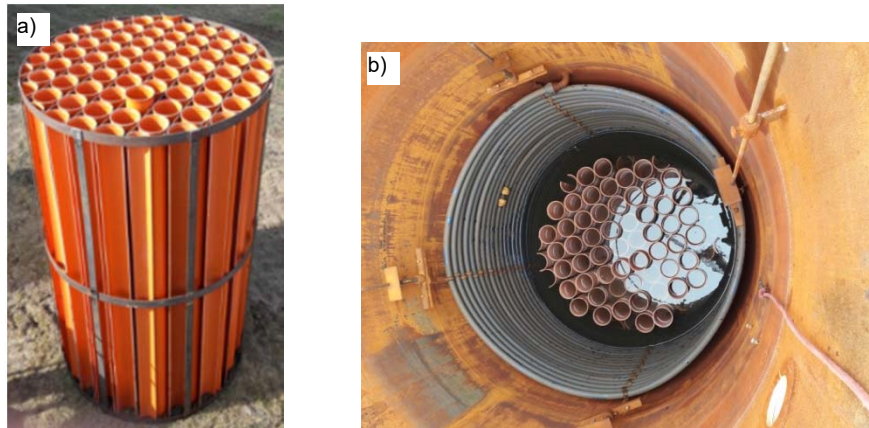


Photo 2. Adhesive bed: a) the filling is a “basket” with pipes type A PVC-U S4 UD – roughness after sanding is 80 μm , b) during the pressure test flooded with an inert agent (phot. G. Walowski)

a skeletal deposit made of vertical pipes PVC constituting the so-called “basket”, whose purpose is to increase the active surface area for the flora of fermenting bacteria. The filling is located at a height of 1.22 m from the bottom of the tank, the so-called “basket” is based on supports that simultaneously center it relative to the system axis (Photo 2).

Heating system – description of the system operation. The inner wall of the fermentation tank is equipped with a heating spiral in the form of a plastic pipe DN32. The heating medium is hot water taken from the main heat exchanger located near the co-generator. For optimal biogas conditions, the walls, conical bottom and fermenter cover are insulated to limit heat emission to the outside. The optimal working conditions of the fermenter are as follows temperature 35–40°C, gas hypertension 1020 kPa.

Currently, intensive work is underway to test the software for controlling the installation. After the acceptance of the installation, the start-up phase on pig slurry occurred. After the acceptance of the installation, the start-up phase on pig slurry occurred.

The start-up was carried out for 10 days on a digestate liquid inoculum from a biogas plant from the Wielkopolskie voivodeship with analytical parameters of the process: temperature 27.2°C, pH 8, dry mass 4.37% DM, dry organic mass 62.25% DM, OWN 17.641 $\text{mg}\cdot\text{dm}^{-3}$, LKT 3.117 $\text{mg CH}_3\text{COOH}\cdot\text{dm}^{-3}$, APB 0.177 for a starting volume of 10 m^3 .

The liquid substrate was slurry of fattening pigs with analytical parameters of the process: temperature (26.5–30.5) °C, pH (7.8–8.0), dry mass 3.92%, dry organic mass 66.70%, OWN 19.678 $\text{mg}\cdot\text{dm}^{-3}$, LKT 8.958 $\text{mg CH}_3\text{COOH}\cdot\text{dm}^{-3}$, APB 0.450 for the feeding volume 250–500 dm^3 [ITP 2018].

As a result, the gas pressure in the installation was obtained 1.5–2.5 kPa and the concentration of biogas components: CH_4 57.3%, CO_2 28.5%, O_2 0.3%, H_2S 0.000232%.

In order to understand the hydrodynamic substrate mixing system conditions in the adhesive bed (Photo 3), experimental investigations have been carried out to assess the biogas flow by the adhesive bed in the context of biogas production.

The research material was made of skeletal bed (72 pipes – Photo 2a) with parameters: height $h_z = 2030$ mm;



Photo 3. The mixing system in the reactor – pressing of the inert agent during the tightness test (phot. G. Walowski)

diameter $d_z = 1620$ mm; bed volume $V_z = 0.4564$ m^3 ; the bed porosity $\epsilon = 10.91\%$, $\epsilon \approx 0.11$; cross-sectional area $A_z = 0.2266$ m^2 . The elementary skeleton bed unit was a pipe (1 item is an apparent elementary bed unit): height $h_r = 2030$ mm; diameter $d_r = 160$ mm; the volume of the pipe (ring) $V_r = 0.00634$ m^3 .

The experimental research concerned a measurement system for the assessment of the amount of biogas in the adhesive bed the conditions of the biogas production process (Tab. 2). The research was carried out in the field of biogas flow rate measurement resulting from the reference pressure in the fermenter. An independent assessment of the amount of biogas and pressure drop on the adhesive substrate was carried out.

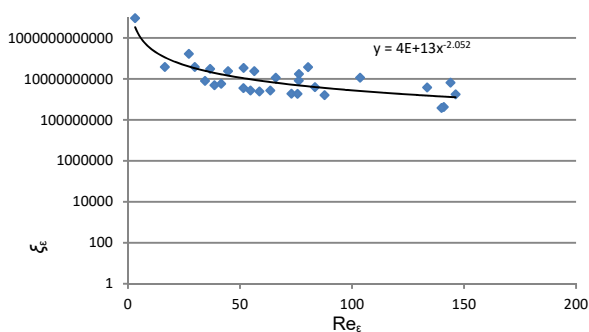
The basis for the assessment of hydrodynamics of gas flow through the adhesive bed is the flow characteristics (Fig. 2), which results from the pressure forcing this flow. In each case, the determination of this characteristic consists in determining the impact of the biogas stream on the value of this overpressure, equivalent to a pressure drop – this is tantamount to determining the total resistance of biogas flow through the adhesive bed.

When assessing the flow resistance, the analogy to the flow through closed ducts is most often used in accordance with the Darcy and Weisbach equations [WAŁOWSKI 2017]. However, the flow resistance coefficient (Eq. 1) is described as a function of the Reynolds number (Eq. 2):

Table 2. Initial results of the biogas production process

Velocity equivalent w_ε , m·s ⁻¹	Pressure drop ΔP , Pa	Flow resistance coefficient ξ_ε	Reynolds number Re
0.000174071	118	0.0001552744	34.4
0.000649894	91	0.0001740708	38.6
0.000232953	1 681	0.0006498937	144.0
0.000187764	1 127	0.0002329528	51.6
0.000362644	123	0.0001877641	41.6
0.000636227	3 017	0.0003626439	80.3
0.000344336	104	0.0006362274	141.0
0.000232449	1 240	0.0003443364	76.3
0.000396154	117	0.0002324491	51.5
0.000297674	155	0.0003961538	87.8
0.000344176	621	0.0002976745	66.0
0.000329170	601	0.0003441756	76.3
0.000341487	124	0.0003291695	72.9
0.000254322	133	0.0003414870	75.7
0.000122433	949	0.0002543217	56.3
0.0000142357	1 518	0.0001224328	27.1
0.000246768	1 149	0.0000142357	3.2
0.00028684	100	0.0002467683	54.7
0.000376683	136	0.0002868405	63.6
0.000201475	340	0.0003766826	83.5
0.0000742641	592	0.0002014750	44.6
0.000264775	128	0.0000742641	16.5
0.000165373	103	0.0002647754	58.7
0.000134335	515	0.0001653731	36.6
0.000467543	420	0.0001343348	29.8
0.00060271	1 526	0.0004675428	103.6
0.0006317	847	0.0006027099	133.5
0.000659961	92	0.0006316997	140.0
0.000155274	463	0.0006599607	146.2

Source: own elaboration.

**Fig. 2.** The influence of the Reynolds number on the downward biogas resistance factor for the adhesive deposit; own study

$$\xi_\varepsilon = \frac{2}{\rho_\varepsilon w_\varepsilon^2} \Delta P_{zm} \quad (1)$$

Where: ΔP = pressure drop (Pa); ρ = density (kg·m⁻³); w = velocity (m·s⁻¹); index: ε = equivalent; zm – measured.

$$Re_\varepsilon = \frac{w_\varepsilon d_\varepsilon \rho_\varepsilon}{\eta_\varepsilon} \quad (2)$$

Where: w = velocity (m·s⁻¹); d = diameter (m); ρ = density (kg·m⁻³); η = viscosity Pa·s; others as in Eq. (1).

Interpreting Fig. 2, it should be noted that there is a non-linear tendency characteristic of the dominance of turbulent flow – this is related to the derogation from Darcy's law [WAŁOWSKI 2017].

CONCLUSIONS

The assessment of biogas flow through an adhesive bed in analogy to gas-permeability for a structural model of a porous material allowed to recognize the problem of biogas production for skeletal deposits. In the context of the criterion number for hydrodynamic conditions, the issue of biogas flow through an adhesive bed is described. An experimental investigation into the hydrodynamics of substrate mixing in a mono-substrate fermenter was made and the hydrodynamic phenomenon resulting from the drop in gas flow pressure was evaluated. The paper presents preliminary results of experimental studies, which indicate a clear influence of flow resistance in relation to Reynolds' numbers.

It was found that the production of biogas using the adhesive bed is determined by the characteristic parameters: the degree of porosity $\varepsilon = 10.91\%$, $\varepsilon \approx 0.11$; for the gas flow $w_\varepsilon = (0.0000142357 \div 0.000659961)$ m·s⁻¹ and the Reynolds number $Re = (3.2 \div 146.2)$, which together with the increase causes a decrease in the descending rate of biogas flow resistance. Taking into account the indicated parameters, a model and methodology concerning the quality and quantity of biogas production can be developed in this way.

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Ocena przepływu wielofazowego w procesie fermentacji w reaktorze monosubstratowym ze złożem szkieletowym

STRESZCZENIE

Dokonano przeglądu wybranych technik oraz scharakteryzowano ich technologiczne aspekty w kontekście przepływu wielofazowego w produkcji biogazu. Przeanalizowano warunki fermentacji beztlenowej gnojowicy świńskiej w reaktorze monosubstratowym ze złożem szkieletowym. Wskazano na wymagane kryteria techniczno-technologiczne wytwarzania surowego biogazu.

Projekt oraz budowę modelu monosubstratowego, przepływowego reaktora biogazowego zrealizowano z przeznaczeniem do współpracy z budynkami inwentarskimi o różnej wielkości oraz mocy od 2,5 do 40 kW. Instalacja ma formę szczelnego zbiornika fermentacyjnego wypełnionego złożem szkieletowym stanowiącym swoisty przestrzenny układ o regularnych kształtach i chropowatej powierzchni.

Wkomponowanie w taki cykl produkcyjny instalacji umożliwiającej skierowanie całego strumienia gnojowicy z kanałów podrusztowych obory lub chlewni do pracującego w trybie przepływowym reaktora, w którym będzie zachodziła fermentacja beztlenowa, umożliwia uzyskanie biogazu. W pracy przedstawiono wstępne wyniki badań eksperymentalnych z zakresu hydrodynamicznego układu mieszania substratu na potrzeby oceny przepływu biogazu przez złożo adhezyjne w kontekście produkcji biogazu. Celem badań była ocena i wykazanie wpływu liczby Reynoldsa na współczynnik oporów przepływu biogazu w procesie fermentacji w reaktorze monosubstratowym ze złożem adhezyjnym.

Badaniom poddano natężenie przepływu biogazu wynikające z ciśnienia odniesienia w fermentorze. Przeprowadzono niezależną ocenę ilości biogazu i spadku ciśnienia na złożu szkieletowym. Podstawą oceny hydrodynamiki przepływu gazu przez złożo adhezyjne jest charakterystyka przepływu, wynikająca z ciśnienia wymuszającego ten przepływ. W każdym przypadku wyznaczenie tej charakterystyki polega na określeniu wpływu strumienia biogazu na wartość tego nadciśnienia, równoważnego spadkowi ciśnienia – jest to równoznaczne z wyznaczeniem całkowitych oporów przepływu biogazu przez złożo adhezyjne.

Wyniki pomiarów wskazują na wyraźny wpływ średnicy ekwiwalentnej $d_e = 2.01$ m zawartej w liczbie Reynoldsa na zastępczy współczynnik oporów przepływu ζ_e dla złoża adhezyjnego.

Słowa kluczowe: proces fermentacji, przepływ wielofazowy, reaktor monosubstratowy, złożo szkieletowe