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Original Research

Effect of Reduced Pressure on Anaerobic Blood Biodegradation

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Abstract

The aim of this study was to determine the effect of reduced pressure on methane fermentation of blood, which is the main slaughter waste in the meat processing industry. The experiment was conducted in laboratory anaerobic reactors. A reactor used under atmospheric pressure was used as a control system in relation to a reactor connected to a vacuum pump, producing sub-atmospheric pressure in the reaction chamber of the bioreactor. A stable and high pH value (7.92 on average) was observed throughout the experiment at a reduced pressure, regardless of the waste load on the anaerobic sludge. In a control system, the accumulation of the waste load caused a systematic decrease in pH value, which in the final stage of the experiment dropped to 5.53. The fermentation gas, produced in the reactor with sub-atmospheric pressure, contained higher concentrations of methane – by 5-15%, ammonia – by 3-4% and hydrogen sulphide – by about 1%.

Keywords: methane fermentation, slaughter waste, blood, biogas, sub-atmospheric pressure

Introduction

Among recently proposed solutions in the technology of waste treatment, methods involving anaerobic decomposition of contaminants have been attracting more and more attention. For many years, these methods have only been used for stabilizing sludge. Nowadays, thanks to improved knowledge of the physical, chemical and biological principles of the methane fermentation process, these technologies can be applied to the treatment of highly loaded industrial waste, particularly in such branches as the distillery industry, sugar production, dairy, fruit and vegetable processing or the meat processing industries [1, 2, 3].

Blood is a major source of waste organic compounds in the meat processing industry. Considering the fact that the value of the BOD index for blood is 140-200 g/dm^3 [4, 5, 6], purification of blood-containing waste or biodegradation of the pure product seems impossible in conventional systems of waste treatment, particularly in aerobic ones. The processes of anaerobic degradation are currently a very popular method of meat industry waste treatment. The solutions applied here are based on systems with contact reactors, UASB reactors or anaerobic filters. Johns [5] found a contact system to be one of the most popular and most effective technologies of meat industry treatment, as it eliminates the problem of waste frothing, which frequently occurs in other anaerobic systems. The data collected by the author indicate that operating a contact reactor in a US-based slaughterhouse enabled a reduction of the contaminant levels in waste – in BOD by 93% and in COD by 84%, whereas the effectiveness of eliminating the total suspensions was as high as 75%. Applying a 3,000 dm^3 contact reactor at a site in France for the treatment of this sort of waste produced a satisfactory result even at BOD loads as high as 5 g/dm^3·d. Rajeshwari et al. [3] compared the technological effects

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obtained in the treatment of slaughterhouse waste in three types of reactors: UASB, an anaerobic filter and anaerobic contact system. The contact system was found to be the best method because the presence of large amounts of suspensions and fats did not adversely affect the system operation. When the reactor was loaded with a load of 3 g COD/$dm^3 \cdot d$, the effectiveness of reducing the COD concentration in the waste reached 92.6%. The effectiveness in the other systems was only 85%.

Among the numerous values of anaerobic biodegradation of organic compounds the following factors should be considered important from a technological point of view: the low index value of the anaerobic sludge biomass growth, the relatively small area which is required to operate the treatment station systems, the limited propagation of aerosols and odors in the environment, the possibility of purifying a highly concentrated substrate and operating the anaerobic reactors in a campaign, together with producing the fermentation gas [7, 8, 9]. Using biogas obtained in anaerobic processes for heating or in producing electricity generates considerable economic profits for the plant, which operates an anaerobic waste treatment station. It has been calculated that the output of methane production in the process of fermentation of industrial waste in the food processing industry, loaded mainly with easily degradable organic compounds, amounts to as much as 30 and even 500 dm^3/kg of waste [7]. The average biogas output from methane fermentation of waste ranges from 300 – 500 dm^3/kg of the removed COD [3, 10, 11]. Hobson and Whealthey [12] give the calorific value of low-energetic biogas with 65% of methane as equal to 6.2 kWh/m^3 (at standard temperature and pressure). According to other literature data, the calorific value of biogas of the same methane content ranges from 6.5 to 7.1 kWh/m^3.

In order to make the process of anaerobic degradation of organic compounds even more attractive compared to other innovative technological solutions in waste treatment, many measures have been taken in order to improve the effectiveness of the biochemical processes which occur with the limited concentration of molecular oxygen. It seems that pressure is the physical factor which, by modifying the conditions in the reaction chamber, can indirectly increase the rate of metabolism of anaerobic sludge.

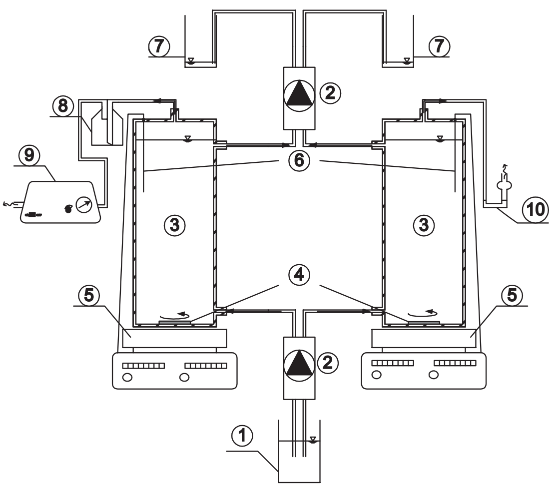
There have been numerous reports on the biochemical and mechanical effect of pressure on living organisms [13, 14, 15]. The effect that a sub-atmospheric pressure has on a living organism is called hypobaria. The biochemical effect of the particular gases which make up the mix depend on their partial pressures, but it does not depend on their percentage in the mix. This interaction is directly related to the chemical affinity of a given gas to the components of living organisms; consequently, the type of gas present in the organism's habitat is the most important biochemically. The mechanical interaction of pressure and gases which are present in living organisms is described by Boyle's law [13]. The solubility of gases in liquids in a lowered pressure decreases as per Henry's law [14, 15]. It seems, therefore, that this fact also significantly affects the operation of anaerobic systems, where the components of biogas, produced at a decreased pressure, are less soluble in waste and therefore have a less toxic effect on the anaerobic sludge.

This study aimed at determining the effect of a reduced pressure on the process of anaerobic decomposition of the organic compounds found in blood.

Experimental Procedures

The effect of a reduced pressure on the course of methane fermentation of blood was examined in a dynamic system of anaerobic reactors in a laboratory.

The research stand consisted of two anaerobic reactors, with one of them being used as a control in relation to the system working at sub-atmospheric pressure (Fig. 1).



1 – raw waste container
2 – peristaltic pump
3 – laboratory anaerobic reactor
4 – magnetic stirring system
5 – thermal stand
6 – pH measuring electrodes
7 – purified waste container
8 – liquid condenser
9 – vacuum pump
10 – siphon

Fig. 1. Scheme of the research stand.

Before beginning the study, the anaerobic sludge was adapted for 30 days to the composition of blood used in the experiment. Subsequently, the reaction chambers of the reactors were filled with 1.95 dm^3 of previously prepared anaerobic sludge. The anaerobic sludge originated from laboratory UASB reactor which was used to treat dairy wastes.

The active volume of one reactor was 2 dm^3. Blood from a poultry processing plant was used as the substrate for fermentation. As the blood tended to clog, it was homogenized and collected in the container with raw waste. 0.05 dm^3 of this blood was subsequently dosed once in 24 hours with a peristaltic pump to the lower part of each of the reactors. A single load of 10 g COD/dm^3 of organic waste was fed to a single system. In the upper part of reactors there were pipes to drain off the fermented blood, which was removed every 24 hours in amounts of 0.05 dm^3, with a peristaltic pump, to two containers with purified waste. In order to obtain the proper concentration of anaerobic sludge in reactors, fermented blood was filtrated and the separated anaerobic sludge was recirculated to the reactors. The ascending flow of waste, together with the magnetic stirring system, brought about the mixing of the content of both bioreactor chambers. In order to prevent any adverse effects caused by too intensive stirring, the speed of the magnetic stirrer was set to 100 rpm, the device itself being integrated with an electronic timer, which switched it on every hour for 10 minutes. At the top of the anaerobic reactors, degasifier outlets were situated for letting off the fermentation gas. The outlet of gas in the first reactor was connected to a vacuum pump, which pumped out the gas and produced sub-atmospheric pressure in the bioreactor's reaction chamber. The pressure was set at -1hPa. Maintenance sub-atmospheric pressure in the bioreactor's reaction chamber relied on switching off a vacuum pump, when sub-atmospheric pressure attained the level -1hPa and switching on it, when the pressure increased to -0.6hPa. The biogas outlet from the control reactor was fitted with a siphon which enabled the outflow of the generated gas and maintained the anaerobic conditions in the reaction chamber. The control reactor ran under atmospheric conditions. During the experiment, the temperature inside the reaction chambers of the laboratory reactors was maintained with a thermal stand (integrated with the electronic stirring system) in the mesophilic range at 32ºC.

Samples were taken every 48 hours directly from the fermented blood containers. The measurements included COD value, which was the basis for calculation of the organic load in wastes. The pH value in the reaction chambers of anaerobic systems, which is an indicator of the correct course of a fermentation process, was monitored throughout the experiment. The qualitative composition of the fermentation gas, generated during the biochemical processes, was also determined.

The COD index was determined (by the dichromate method) according to the standard methodology [16]. The waste pH value was measured with a digital pH-meter, with ± 0.01 accuracy. The qualitative analyses of biogas were performed with an electronic analyzer with an accuracy of ±0.1%.

Results

In the course of this experiment, the waste of homogenized blood from a poultry processing plant was anaerobically biodegraded in laboratory anaerobic reactors. The COD index and pH values measured in raw blood waste fell within the typical ranges reported in literature and equaled 200 g O_2/dm^3 and 7.5 pH, respectively [4, 5, 6],

Dosing the blood to the reactors caused the accumulation of the organic load in the reaction chambers and, consequently, the operation of the system with a steadily growing load of the anaerobic sludge with an unremoved load of organic compounds. During the first 24 hours of the experiment, the amount of COD per 1 g of the anaerobic sludge dry substance was 0.076 g, whereas in the last, 28^{th} day, the amount was 1.614 g COD (Fig.2).

The pH values of the waste environment in this experiment were different in the reduced pressure reactor and in the control. Considerable differences were also found in terms of the composition of the fermentation gas produced in each system (Figs. 3, 4, 5).

During the experiment, a tendency in the system of reduced pressure was observed for the pH value to maintain a stable level of 7.92 on average. The accumulation of the load of organic compounds in the reactor's reaction chamber did not bring about any rapid changes in the pH value. At the lowest organic load at the beginning of the experiment (0.076 g COD/g of anaerobic sludge dry substance), the pH value of the waste was 8.76, while at the final stage, at the highest organic load (1.614 g COD/g of anaerobic sludge dry substance), the pH value dropped to 7.93 (Fig. 3.).

The pH values in the control reactor were lower throughout the experiment. It was also noted that they decreased with the increase in the level of organic compounds in the reactor. The pH values gradually decreased from 7.15 at the beginning to 5.53 (Fig. 3.).

The fermentation gas produced in the reactor connected to the vacuum pump contained more methane as compared to that produced in the control system. During the first eight days of the experiment, the highest concentration of methane in the biogas was measured, ranging from 80 to 85% v/v. The concentrations of the other components of the biogas were as follows: carbon dioxide 13-19% v/v, ammonia 0.5-1% v/v, hydrogen sulphide 0.1-0.3% v/v. The gradual increase in the load of organic compounds to 1.116 g COD/g of the anaerobic sludge dry substance on day 18 of the experiment produced a decrease in the methane content to 60%, while the concentration of the other components increased. Carbon dioxide accounted for 20-40% v/v, ammonia 2-3% v/v and the concentration of hydrogen sulphide grew to 0.5-1% v/v. At the highest organic compounds accumulation (1.614 g COD/g of an-

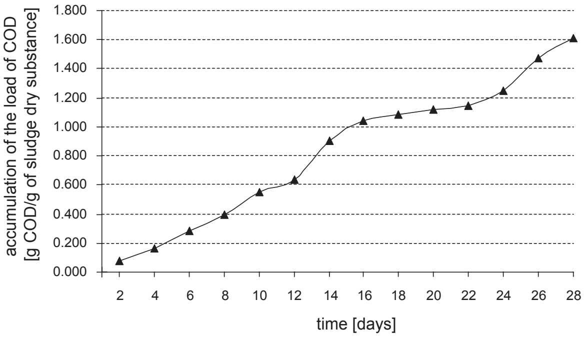


Fig. 2. Accumulation of organic load in the reaction chambers.

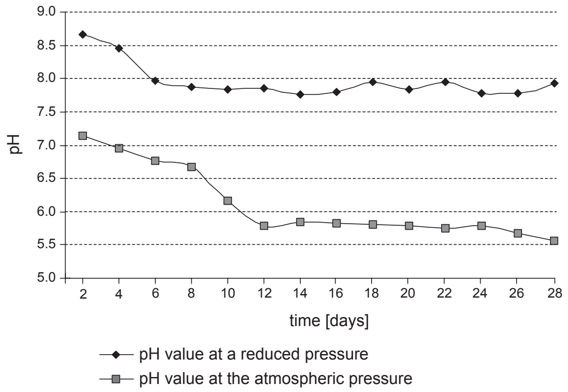


Fig. 3. The relationship between pH value and COD load.

aerobic sludge dry substance), the composition of the fermentation gas produced in the reduced pressure system was as follows: methane 20-30% v/v, carbon dioxide 30-40% v/v, ammonia 2-4% v/v, hydrogen sulphide 0.5-1% v/v (Figs. 4, 5).

The increase in the level of organic compounds in the system run under atmospheric conditions produced a gradual decrease in methane content in the biogas and an increase in carbon dioxide concentration. However, no significant changes in the concentrations of ammonia or hydrogen sulphide in the fermentation gas were observed. During the first 8 days of the experiment, the biogas produced in the system contained: methane 75-80% v/v, carbon dioxide 12-20% v/v, ammonia 80-150 ppm, hydrogen sulphide 0.0-5.0 ppm. Accumulating the organic compounds to 1.116 g COD/g of the sludge dry substance produced a reduction in methane content in the biogas to 41%. At the end of the experiment, with 1.614 g COD per 1 g of the anaerobic sludge dry substance, the percentage of various components of the fermentation gas was as follows: methane 13-15% v/v, carbon dioxide 40-60% v/v, ammonia 500-1000 ppm, hydrogen sulphide 100-350 ppm (Figs. 4, 5).

Discussion of Results

The experiment conducted as part of the study aimed at determining the effect of reduced pressure on the conditions of anaerobic degradation of blood.

It seems that an appropriate modification of the pressure in the reaction chambers of the anaerobic reactors can reduce the adverse phenomena associated with the production of fermentation gas which contains a lot of components toxic to microorganisms, such as hydrogen sulphide or ammonia.

Hydrogen sulphide, produced in anaerobic systems, both undissociated (as gas) and dissociated, not only creates operational problems associated with emitting smelly substances, corrosion of the treatment plant equipment or increasing the COD concentration in the

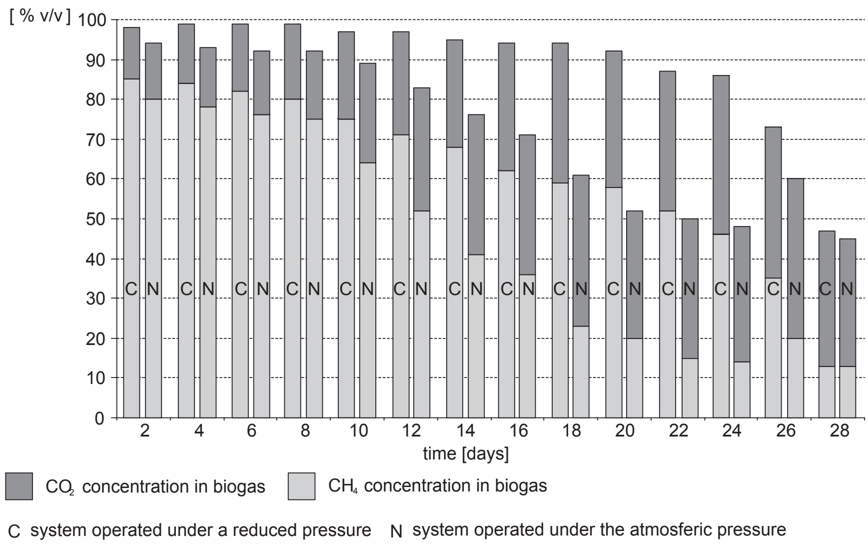


Fig. 4. A comparison of the concentration of methane and carbon dioxide (v/v) in the fermentation gas produced in both research systems.

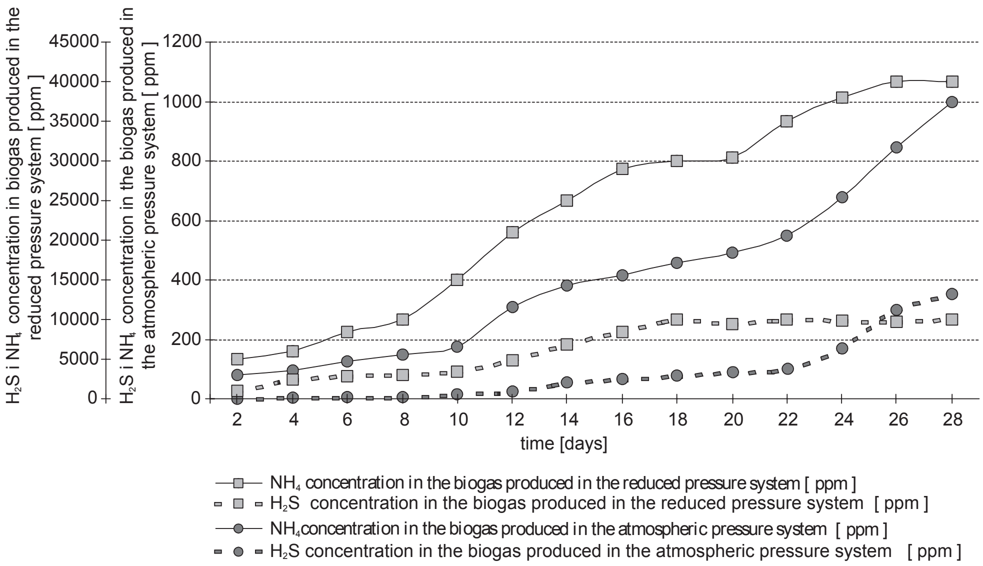


Fig. 5. A comparison of the concentration of hydrogen sulphide and ammonia (ppm) in the fermentation gas produced in both research systems.

purified waste, but it can also create microbiological problems. The data available in the literature confirm the toxic effect that undissociated hydrogen sulphide has on methanogenic archaea [17, 18, 19, 20]. This effect can be predicted by determining the amount of undissociated hydrogen sulphide, produced in anaerobic systems, which, unlike its dissociated form, can infiltrate through membranes into bacterial cells [17, 21]. Furthermore, a concentration of undissociated H_2S is closely linked to the pH of the reaction environment. The presence of small amounts of sulphur compounds in an anaerobic waste environment does not cause a toxic effect. Sulphides are basic biogenic elements which are indispensable for methanogenic archaea to develop normally [21]. A literature review indicates that the negative effect of hydrogen sulphide produced during the fermentation of industrial waste can be reduced by applying beds filled with iron oxides, which are washed over by waste [17, 22]. An experiment was also carried out which proved that applying steel elements as an additional filling in a reaction chamber of an anaerobic bioreactor reduces the aggressive effect of hydrogen sulphide on the microflora of the anaerobic sludge [23].

Biochemical transformations of proteins in anaerobic reactors lead to the accumulation of ammonia in the waste environment. Ammonia, like hydrogen sulphide, has a toxic effect on methanogenic archaea, reducing its metabolic activity and, in consequence, inhibiting the multistage process of anaerobic degradation of contaminants [24, 25, 26, 27]. It has been shown that the inhibiting effect of ammonia on methanogenic archaea depends on the form in which it is found in waste; it is particularly high with large amounts of accumulated, undissociated ammonia [27]. A concentration of the undissociated form of NH_3 depends directly on the pH of the environment and, to a lesser extent, on the temperature. According to Kayhanian [26] at pH 7.0, the fraction in question accounts for merely 1% and increases to 10.2% (the remaining part is present as NH_4^+ammonium ions). As a literature review shows, an undissociated ammonia concentration in the waste environment of anaerobic reactors, ranging from 0.08 to 0.1 g/dm^3 has a toxic effect on methanogenic archaea at pH = 7.5 [27, 28]. It should be emphasized that adapting microorganisms to higher concentrations of ammonia enables the faultless operation of bioreactors at increased concentrations of ammonia.

A negative effect of undissociated hydrogen sulphide and ammonia on the microorganisms which take part in biochemical transformations can be reduced by applying sub-atmospheric pressure.

At a constant temperature, solubility of gases in liquids increases up to a certain critical level, while it decreases under a reduced pressure. The relationship is described by Henry's law [14, 15]. If a liquid is in contact with a gaseous solution, each of its components conforms to the law, dissolving to a larger or lesser extent in the liquid at a given temperature and pressure (Henry's-Dalton's law) [14]. Consequently, reducing the pressure in the air space of the reaction chamber of anaerobic bioreactors reduces the content of the toxic components of the biogas in the waste environment because of the lower partial pressures of the components over the liquid solution.

An analysis of the qualitative composition of the fermentation gas produced in the two reactors revealed a higher percentage of hydrogen sulphide and ammonia in the biogas from the reduced pressure system. Operating the system under ambient pressure resulted in generating the fermentation gas with a 30-times lower concentration of hydrogen sulphide and 40-times lower concentration of ammonia. A higher concentration of toxic components of biogas in the fermentation gas produced in the reduced pressure system may indicate its lower concentration in the waste environment. Reducing the toxic effect of ammonia and hydrogen sulphide on methanogenic microflora is reflected in a stable and high pH value in the reaction chamber, usually averaging 7.92. Despite the accumulation of organic compounds in the reactor, a decrease in the pH value, which is a characteristic feature in the operation of anaerobic bioreactor at increasing load, was not observed [29, 30, 31]. The solubility of ammonia and hydrogen sulphide in the ambient temperature and pressure in the control system was higher, and, consequently, the metabolic paths of methanogenic archaea were partly inhibited. Limiting the activity of the bacteria in the waste environment of a reactor brought about an accumulation of acidic products of fermentation and a steady decrease of the pH value, down to the critical value of 5.53.

In the system operated under a reduced pressure, a higher concentration of methane in biogas was observed. Initially, the observed difference in the methane concentration in the fermentation between the control reactor and the sub-atmospheric pressure system was found to be 5%. Further operation of both systems with an increasing amount of organic compounds increased the difference in the methane concentration in biogas, although its concentration in both the bioreactors was constantly decreasing. In the final stage of the experiment, the concentration of methane in the fermentation gas produced in the reduced pressure system was 15% higher than in the control system. This is economically significant, as methane is a highly calorific component of biogas which is increasingly used in the production of so-called “ecological” energy.

Conclusions

- The concentration of ammonia and hydrogen sulphide in the biogas produced in the sub-atmospheric pressure reactor was 40 and 30 times lower, respectively, than in the fermentation gas produced in the control system.
- During the experiment, the pH value in the control system was found to decrease steadily down to 5.53, while in the reduced pressure system, it was stable and relatively high – 7.92.
- Despite the decreasing concentration of methane in the fermentation gas in both systems, the concentration of this gas was found to be higher in the system operated under a reduced pressure.

Units

COD – chemical oxygen demand [g O_2/dm^3]
BOD – biochemical oxygen demand [g O_2/dm^3]

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