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INTENSIFICATION OF THE ANAEROBIC MICROBIOLOGICAL DEGRADATION OF SEWAGE SLUDGE UNDER BIO-SULFIDOGENIC CONDITIONS

Possibility of using phosphogypsum as a mineral substrate for the sewage sludge treatment in bio-sulfidogenic conditions has been investigated. Anaerobic microbiological degradation (AMD) on the way of sulfidogenesis was used to produce biogenic hydrogen sulfide. The ozonation of the sludge prior to the AMD under bio-sulfidogenic conditions led to higher efficiencies of hydrogen sulfide production and organic matter removal. After 20 days of digestion, the elimination of CODt and CODs varied between 78–84% and 83–87%, respectively, within advanced operation of anaerobic bioreactor. Technological applications of such system has been discussed.

SOME ABBREVIATIONS USED IN THE TEXT

TS – total solids, mg/dm^3
VS – volatile solids, mg/dm^3
TSS – total suspended solids, mg/dm^3
VSS – volatile suspended solids, mg/dm^3
VFA – volatile fatty acids, mg/dm^3
COD_t – total chemical oxygen demand, mg/dm^3
COD_s – soluble chemical oxygen demand, mg/dm^3
SRT – sludge retention time, day
SRB – sulfate-reducing bacteria
AMD – anaerobic microbiological degradation
WWTP – wastewater treatment plants

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1. INTRODUCTION

Recycling of organic sewage sludge in the whole territory of Ukraine is an urgent problem, which requires a solution. In particular, in the municipal wastewater treatment plants (WWTP), unfortunately, removal, treatment and recycling of sewage sludge are not properly resolved. At present, a general part of sewage sludge is not carried out. It contains toxic chemicals, mainly heavy metals, originating from the industrial wastewater which come into the city sewer system after insufficient treatment or without treatment. The other problem is sludge production with high phosphate content. Ferrous or ferric phosphates have low solubility and are therefore of limited agricultural use [1, 2].

As a result, sewage sludge is sent to sludge pit and storage sites, which occupy large land areas. Therefore, the process of heavy metals removing and biological release of phosphates from sewage sludge is paramount importance to allow using sewage sludge as an organic fertilizer in agriculture. The management of solids and concentrated contaminants present in the sludge is still one of the most difficult and expensive problems in the field of wastewater engineering. The most common treatment options include aerobic digestion, anaerobic digestion and composting. Among these biological treatments, anaerobic digestion is frequently the most cost-effective, due to the high-energy recovery linked to the process and its limited environmental impact. Technologies and processes for heavy metals removing and recovery of phosphorus are introduced to industrial scale. The system of anaerobic microbiological degradation (AMD) with deposition of heavy metals (HM) by biogenic hydrogen sulfide is a promising procedure of recoverable resources treatment [1–4].

Microbiological investigations [3] have shown that the mineral substrate for the cultivation of SRB can not only be soluble sulfates, but also insoluble sulfates of barium, lead, gypsum, etc. However, today there is no developed technology based on the use of insoluble solid mineral substrates for sulfate reduction process. The promise of such systems in the industrial scale is a matter of waste disposal and reducing the anthropogenic impact on the environment as a whole. In most cases, sludge contains lots of large organic polymers. In order to access the energy potential of the materials, the long chains must firstly be broken down to their smaller components such as sugars being readily available for other bacteria. It is well known that the hydrolysis is the limited stage in AMD. Yet, until now, none of the pretreatment technologies has found a real breakthrough.

Sulfidogenic microorganisms association cultivated in a bioreactor can be effective in introducing the system on an industrial scale. Inoculation of SRB culture has a positive effect for a certain period. Its effective stable growth and metabolism depend on the change of concentration of chemical components in the sewage sludge of wastewater treatment plants. Own microbiocenosis is formed in bioreactors. It can inhibit the growth of stock culture. In addition, the SRB biomass is removed along with the fermented waste mixture. The recycling of waste fermented mixture (return of the fermented substrate into the reactor – pretreatment) is one of techniques of effective sludge treatment.

Suschka et al. [2] demonstrated efficient recovery of phosphates from Fe-P precipitate in a biological anaerobic process. SRB could effectively be used for phosphate release from Fe-P sludge. Under strict anaerobic conditions, sulfides produced are immediately associated with iron, forming ferrous sulfide, releasing at the same time phosphates.

Many novel treatment technologies, usually representing a pre-treatment prior to the biological degradation process, have been developed in order to improve the recycling and reuse of sewage sludge. Thermal, chemical, biological and mechanical processes, as well as combinations of these, have been studied [5–10] as possible pre-treatments causing the lysis or disintegration of sludge cells permitting the release of intracellular matter that becomes more accessible to anaerobic microorganisms. Among all the methods available, chemical ones (ozonation) have been considered in this study.

Several authors [5–9] investigated the impact of ozone dose on sludge biodegradation to improve biogas production and methanogenesis with differing efficiencies. Saktaywin et al. [8] found that around 60% of soluble COD generated due to ozonation was biodegradable at its early stage, while the remaining soluble organic matter was refractory. Yeom et al. [9] showed that when the ozone dose was 0.1 g O_3/g TSS, the biodegradation was about 2–3 times greater compared with raw sludge in both aerobic and anaerobic conditions for 5 days. According to Weemaes et al. [10], the biogas production increased by 80% at ozone treatment with 0.1 g O_3/g COD, the effect was not pronounced at higher ozone concentrations.

The authors of the present paper studied the system of AMD with deposition of heavy metals by biogenic hydrogen sulfide. In that case, mainly biogas consists of hydrogen sulfide and carbon dioxide. The paper focuses on the study of using phosphogypsum as a mineral substrate for the growth of sulfate-reducing bacteria (SRB). The other objective is investigation of the ozonation pretreatment method to improve sewage sludge anaerobic degradability and to achieve increasing hydrogen sulfide production in AMD system. In this way, the impact of ozonation was determined. The sludge biodegradation is affected by ozone dose. Efficient technological application of sewage sludge treatment was developed.

2. MATERIALS AND METHODS

2.1. THE POSSIBILITY OF CHEMICAL REACTION IN THE ANAEROBIC MICROBIOLOGICAL DEGRADATION SYSTEM UNDER BIO-SULFIDOGENIC CONDITIONS

AMD has two basic alternative ways of development of the fermentation. The first several stages are hydrolysis, acidogenesis, acetatogenesis. The final stage of AMD is determined by the possibility of microorganisms to use various terminal electron acceptors being determined by thermodynamic factors. Reactions which have the greatest energetic effect begin first [11]. Decomposition reaction of volatile fatty acids (VFA) to acetate and hydrogen is thermodynamically unfavourable and can provide microbial growth only with the very low concentration of the reaction products [12]. Thus, rapid and complete removal of H_2 must be in the microbial association. SRB obtain hydrogen and acetate more easily than methane-forming bacteria. It is due to a higher SRB affinity for the substrate [11, 12].

In the microbial association, SRB has the ability to remove acetates and H_2 forming lower threshold than methanogenic microorganisms [11], i.e. methane-forming bacteria – 100–10 p.p.m. H_2 (gas), SRB – 10–1 p.p.m. H_2 (gas). Thus sulfidogenesis is more energetically favourable for the microbial association than methanogenesis.

Consequently, AMD can develop not only on the way of methane production but also as sulfidogenesis, which was confirmed in [3, 4, 11–13] and in studies of the authors. A schematic model of AMD is shown in Fig. 1. Basic chemical reactions of sedimentation of metals in the form of sulfides are also shown.

Thus, the biochemical model of AMD contains two basic alternative ways of fermentation. Each of them can dominate under specific conditions. If sulfates are present, SRB multiply. Hydrogen is used with sulfates by SRB and hydrogen sulfide (H_2S) is produced. Methanogenesis is usually the last stage of AMD in the biogas technology for production of biomethane. This study of sewage sludge fermentation was aimed to sedimentation of HM by biogenic hydrogen sulfide.

Biogenic hydrogen sulfide is the product of SRB belonging to generic taxon Desulfovibrio, Desulfobacter, Desulfomaculum (Desulfosporosinus), Desulfococcus, etc. [3].

The reactions responsible for sulfate reduction when H_2, CH_3COOH and CO are used as energy sources are the following:

$$5H_2 + SO_4^{2-} \rightarrow H_2S + 4H_2O \tag{1}$$

$$CH_3COOH + 2H^+ + SO_4^{2-} \rightarrow H_2S + 2CO_2 + 2H_2O \tag{2}$$

$$4CO + SO_4^{2-} \rightarrow S^{2-} + 4CO_2 \tag{3}$$

Hydrogen sulfide in water dissociates into hydrogen ions and sulfide ions in two stages:

$$H_2S \leftrightarrow H^+ + HS^- \leftrightarrow 2H^+ + S^{2-} \tag{4}$$

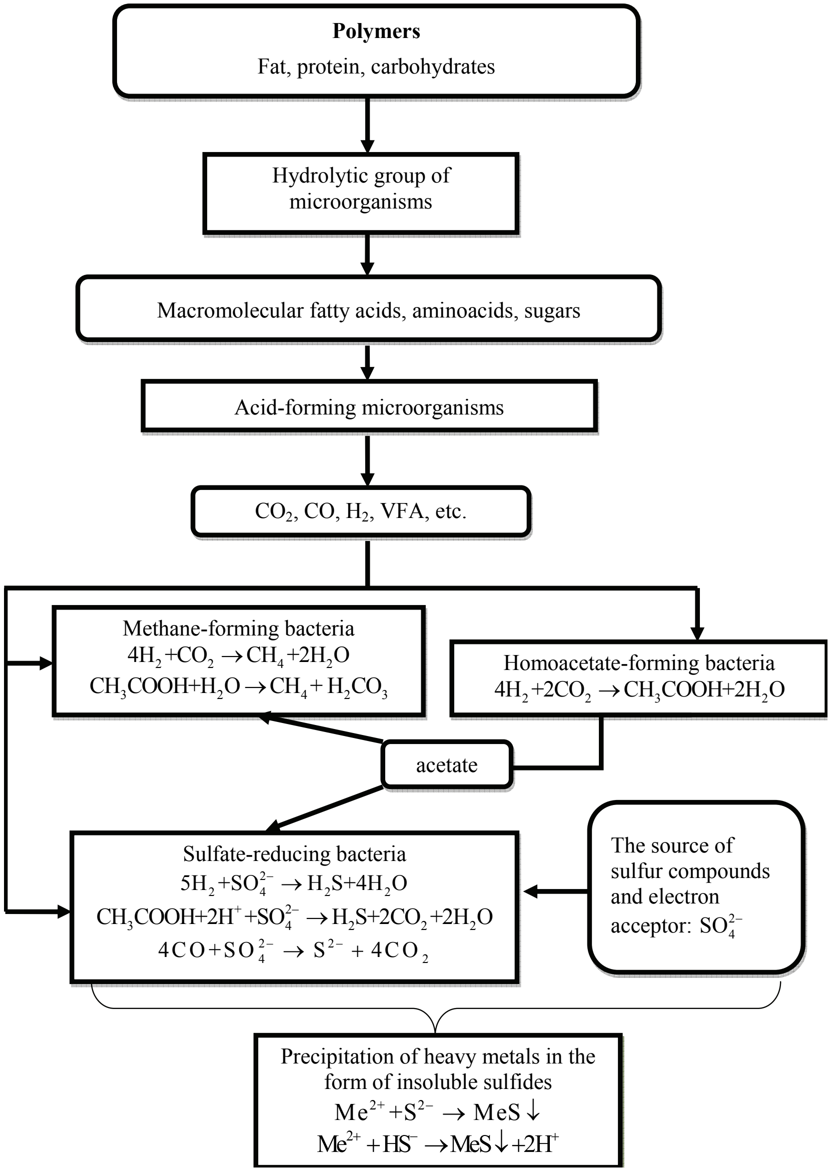

Fig. 1. Biochemical model of sewage sludge fermentation:
two ways are methanogenesis and sulfidogenesis

Its chemical reactions with metal ions occurring in the sediments can be schematically written as:

$$Me^{2+} + S^{2-} \rightarrow MeS\downarrow \tag{5}$$

$$\mathrm{Me}^{2+} + \mathrm{HS}^{-} \rightarrow \mathrm{MeS}^{-} + 2\mathrm{H}^{+} \tag{6}$$

$$n\mathrm{MeS} + m\mathrm{S}^{2-} \rightarrow (\mathrm{Me}_n\mathrm{S}_{n+m})^{2m-}\downarrow \tag{7}$$

where: Me is a heavy metal ion, MeS is a metallic sulfide.

General reaction of the microbial system state in AMD process is as follows:

$$\begin{aligned} &a1_L\,\mathrm{C_{13}H_{31}O_4N} + a1_{SO}\,\mathrm{SO_4^{2-}} + a1_H\,\mathrm{H_2O} \\ &\rightarrow\; a1_Y\,C_5\mathrm{H_7O_2N} + a1_N\,\mathrm{NH_3} + a1_S\,\mathrm{HS^-} + a1_{CO}\,\mathrm{CO_2} + a1_C\,\mathrm{CH_4} \end{aligned} \tag{8}$$

where: $a1_L$, $a1_{SO}$, $a1_H$, $a1_Y$, $a1_N$, $a1_S$, $a1_C$, are stoichiometric coefficients; $C_{13}H_{31}O_4N$ is chemical formula of sewage sludge; $C_5H_7O_2N$ is chemical formula representing the average proportion of main elements in the cellular material of bacteria.

Production dynamics of biogenic hydrogen sulfide is a very important object for studies. This gas is the product of SRB and it is has major importance for the sewage sludge detoxification. However, methane could be present in the gaseous phase as well.

The chemical reactions of sewage sludge detoxification in the last stage of AMD under sulfate reduction conditions are:

$$\begin{aligned} &\mathrm{CH_3COO^-} + a2_{SO}\,\mathrm{SO_4^{2-}} + a2_N\,\mathrm{NH_3} \\ &\rightarrow\; a2_Y\,\mathrm{C_5H_7O_2N} + a2_S\,\mathrm{HS^-} + a2_S\,\mathrm{H_2S} + a2_{CO}\,\mathrm{CO_2} + a2_H\,\mathrm{H_2O} \end{aligned} \tag{9}$$

where: $a2_{SO}$, $a2_N$, $a2_Y$, $a2_S$, $a2_{CO}$, $a2_H$, are stoichiometric coefficients

$$8\mathrm{HS^-} + 9\mathrm{Me}^{n+} + \mathrm{SO_4^{2-}} \rightarrow 9\mathrm{MeS}\downarrow + 4\mathrm{H_2O} \tag{10}$$

Consequently, the process of methanogenesis does not dominate in anaerobic conditions with constant source of sulfates. The sulfidogenic association of microorganisms is reproduced in the bioreactor.

The other way of using sulfate reduction is biological release of phosphates. Phosphorus is the essential nutrient with the lowest reserves and it cannot be substituted by other elements. Assuming a simplified summary reaction which incorporates biological reduction activity of microorganisms, it can be approximated with the following reaction [2]:

$$\mathrm{Fe_3(PO_4)_2} + 3\mathrm{H_2SO_4} + 6\mathrm{C_{org.}} \xrightarrow{\text{microorganisms}} 3\mathrm{FeS} + 2\mathrm{H_3PO_4} + 6\mathrm{CO_2} \tag{11}$$

where: C_{org} represents an organic substrate.

Hence, oxidation of the metal compounds to sulfates is limited in conditions of AMD treatment. Heavy metals are fixed in the form of insoluble sulfides in the separated solid fraction of fermented sludge mixture. This process is interesting in possibility to use sewage sludge as organic-mineral fertilizer.

2.2. RAW SEWAGE SLUDGE CHARACTERISTICS

Raw sewage sludge used in this work was collected from WWTP corresponding to a population of approximately 290 000 inhabitants located in city Sumy (Ukraine). A mixture (50:50, v/v) of primary and excess activated sludge collected from the thickener and flotator, respectively, was used as feeding of the anaerobic microbiological degradation pilot plant.

Primary sludge (PS). Primary is also called raw sludge which comes from the bottom of the primary clarifier. Primary sludge is easily biodegradable since it consists of more easily digestible carbohydrates and fats, compared to activated sludge which consists of complex carbohydrates, proteins and long chain hydrocarbons. Thus biogas is more easily produced from primary sludge.

Activated sludge (AS). Activated sludge is also called excess sludge or waste activated sludge which comes from the secondary treatment. It iss a result of over production of microorganisms in the activated sludge process. The content of activated sludge was just mentioned above. Activated sludge is more difficult to digest than primary sludge. Hence, it is very important to use efficient pretreatment method. The main characteristics of this feeding are given in Table 1.

Table 1

Main components of the raw sludge [mg/dm^3]

Sludge	TS	VS	TSS	VSS	COD_t	COD_s
PS	2130	1831	2240	1960	2500	690
AS	2038	1800	2100	1867	2370	500
Mix.	2088	1815.5	2170	1913.5	2435	595

The increase of the solubility of COD is the one of the main aims of any pretreatment method.

The solid sewage sludge with a minimum shelf life (not longer than six months) or liquid fraction from the primary clarifiers and excess activated sludge are processed. This will allow efficient use of the organic matter contained in sewage sludge. Its amount must provide sewage sludge moisture at least 95%. It is necessary for the smooth exchange of substances on surface boundary of the phases at digestion. The viscosity of the organic sewage sludge allow free movement of bacteria and gas bubbles between the liquid and solid substances contained in it. Accordingly, a high degree of degustation and intensity of the hydrogen sulfide reaction is achieved.

2.3. CHARACTERISTICS OF PHOSPHOGYPSUM

It is advisable to use gypsum wastes as a source of sulfate in biosulfide treatment of sewage sludge. Of all kinds of gypsum wastes of Sumy region (Ukraine), the most appropriate is phosphogypsum. Phosphogypsum is formed in an amount of about 100 tons annually. Currently, over 14 million tons of phosphogypsum are accumulated [14]. It is a multi tonnage waste of extraction phosphoric acid production. Solid waste generated in the process of decomposition of sulfuric acid of natural phosphate raw material and the solid phase (calcium sulfate) separation from phosphoric acid solutions. The reaction proceeds as follows [11]:

$$Ca_5(PO_4)_3F + 5H_2SO_4 \rightarrow 5CaSO_4 + 3H_3PO_4 + HF \tag{12}$$

The precipitate consists mainly of calcium sulfate dehydrate ($CaSO_4 \cdot 2H_2O$) and contains impurities of phosphate, which is not decomposed, phosphates and silicates. The quantitative content of impurities depends on the mineral composition of the feedstock, smooth flow of production, serviceability of equipment and process discipline, etc. (Table 2).

Table 2

Composition of phosphogypsum in terms of oxides [%] [11]

CaO	SO_3	$A1_2O_3$	Fe_2O_3
30–42	44–52	0.3–5.0	0.2–2.0
SiO_2	F	P_2O_5	H_2O
0.3–1.0	0.1–1.0	1–4	25–40

The solubility of phosphogypsum in water is 2100 mg/dm^3 at 35 °C. Using phosphogypsum as a mineral substrate for SRB growth has the following advantages: low-cost raw material base, enrichment biogenic elements (fluorine, phosphorus, etc.) of sewage sludge; sulfur compounds contained in the waste can be freely used by SRB as a mineral substrate for their growth which is due to high sulfate/sulfite ions affinity of microbial cells, reducing of chemical waste development pressure on the environment.

2.4. ANAEROBIC MICROBIOLOGICAL DEGRADATION PILOT PLANT

Experimental setup consists of anaerobic bioreactor, where the process proceeds directly to sulfate reduction. A covered cylindrically shaped anaerobic bioreactor with a zinc acetate trap for sulfide was used to generate sulfate-reducing biomass for the study. The digested sludge outlet was on the bottom of the reactor and it was used for sampling purposes. The bioreactor had a total volume of 6 dm^3. Initial pH level of the system was 7.5. The anaerobic bioreactor was equipped with a temperature regulation device. Constant temperature of 35 °C was kept in the incubator.

In order to maintain anaerobic conditions in the flask, aluminum foil was used to exclude light and to prevent the growth of photosynthetic bacteria. Syringes were used for sampling purposes and a sealed nitrogen injection port was created. The feeding for reactor was stored at 4 °C, from where it is pumped to reactor. A stable mixture was placed in a bioreactor. Retention time (SRT) required for sludge digestion was 10–30 d. Digested mixture was fed into a centrifuge to separate solid and liquid fractions. The concentrations of sulfide ions and sulfate ions were controlled in the liquid fraction.

The components of the gaseous phase were determined. Sampling was carried out in a rubber camera connected to the pipe conducting gas from the bioreactor. The chamber was purged with the analyzed gas two times. The rubber bladder volume was 0.22 dm^3. The samples were analyzed on the day of the selection. Certified calibration gas mixtures containing graded amount of component of the gaseous phase were used for calibration.

In order to determine the maximum solubilization of the sewage sludge under sulfidogenic system, the flask was analyzed until the COD concentration reached minimum values. Samples for analysis were taken every two days.

The following parameters were monitored: moisture content of the substrate, total and soluble COD, pH, concentration of hydrogen sulfide in the gas fraction; concentrations of dissolved hydrogen sulfide and sulfate ions in the liquid fraction.

2.5. OZONATION PRE-TREATMENT

The sludge was ozonated in a tubular 2 dm^3 reactor fitted with a sintered glass dispenser that released the gas from the bottom of the reactor. Ozone was generated by passing pure gaseous oxygen through an ozone generator (OZAT® CFS) at the flow rate of 0.35 $dm^3\cdot min^{-1}$ and introduced into the reactor continuously. The amount of ozone produced was determined spectrophotometrically at 258 nm (ε = 3000 $dm^3\cdot mol^{-1}\cdot cm^{-1}$) in the gas phase by passing the mixture of oxygen and ozone through a flow cell. The ozone concentration of 25 mg/dm^3 was applied for 20 min in a batch reactor. Samples of sludge mixture were submitted to the ozonation process at room temperature. Samples of the ozonated sludge mixture were periodically collected. The ozone dose transferred to the sludge was calculated from the difference between the amounts of ozone at the inlet and outlet of the ozonation reactor per amount of initial TSS. It was 0.03 g O_3/g TSS.

2.6. ANALYTICAL METHODS

A Merck® spectroquant test kit was used to determine the total and soluble COD of the reactor. Prior to COD determination, all samples were acidified with concentrated HCl to pH = 2 in order to remove any dissolved sulfide. The sulphate concentration was determined as barium sulphate by the turbidimetric method [15]. pH was analyzed with a pX-150 ionometer (Belarus). Sulfide (gas phase), samples were collected in tubes containing 100 μdm^3 of 0.1 M zinc acetate solution to prevent sulfide from escaping, and then analysed according to the method of Dubois et al. [16]. A standard curve was created using analytical grade sodium sulfide. Statistica Version 10 was used for statistical analysis of the recorded data. Sulfide (liqua fraction) was determined by measuring the blue colour formation by N,N′-diethyl-D-phenylenediamine in the presence of iron(III) ions. The intensity of the colour is proportional to the concentration of sulfide [17].

Study of the gaseous phase was carried out using a laboratory gas chromatograph SELMICHROM-1 (Ukraine). The thermal conductivity detector (katharometer) was used. Argon was chosen as the mobile phase. Compounds were separated by passingpassed through a coated column and were separated based on size and intermolecular interactions. The sample was injected into the pre-PLOT (porous layer open tubular) column-1 with a porous layer of sorbent PoraPlot Q (styrene-divinylbenzene coating). The gas mixture passing through the column-1 goes to one of the two katharometer cells. The hydrogen, oxygen, nitrogen followed out of the column-1 as a single chromatographic strips, then methane, carbon monoxide(IV) and hydrogen sulfide were separated. The ballast column-2 is filled with an inert carrier Chromaton N-AW-DMCS. The HP-PLOT mole sieve column-3 separated H_2, O_2 and N_2. Recording and processing of chromatograms was performed by the software Multichrom version 1.52x.

3. RESULTS AND DISCUSSION

3.1. ANAEROBIC MICROBIOLOGICAL DEGRADATION UNDER BIO-SULFIDOGENIC CONDITIONS

AMD under condition of sulfidogenesis was studied to examine how adding of phosphogypsum to sludge influences bacterial growth and sulfide hydrogen production. Phosphogypsum is the source of sulfates. Thus, electron acceptors for SRB were constantly introduced to the medium of bioreactor. When sulfate is used to degrade an organic compound, it is reduced to hydrogen sulfide. Phosphogypsum also provides some of the major nutrients. This led to the creation of conditions in which the SRB dominate and inhibit methane forming bacteria growth.

Consequently, phosphogypsum was added to sewage sludge for stimulation growth of SRB. The sulfidogenic community of microorganisms was developed in the bioreactor. It is interesting to note that sedimentation of metals by biogenic hydrogen sulfide is also a natural protection mechanism of the bacterial cells from the toxic effect of HM and protects microbial associations from the toxic action of hydrogen sulfide. The activity of SRB was assessed by decrease in sulfates concentration and simultaneous increase in sulfide concentration (Table 3).

Table 3

Sulfate removal and hydrogen sulfide production under steady state conditions at each SRT (advanced operation of bioreactor)

Time of digestion [day]	Concentration of sulfate [mg/dm^3]	Concentration of hydrogen sulfide [mg/dm^3]
0	2100	0
2	1905	30
4	1745	196
6	1524	565
8	1400	588
10	975	658
12	676	519
14	489	587
16	390	727
18	380	760
20	276	778
22	245	521
24	179	312
26	156	187
28	138	134
30	110	97

The AMD process depends on the time required to reproduce microorganisms, gas formation, and also to remove of COD. A decrease in the sludge retention time (SRT) decreases the extent of the reactions and vice versa. Each time sludge was withdrawn, a fraction of the bacterial population was removed thus implying that the cell growth must at least compensate the cell removal to ensure steady state and avoid process failure. Thus, the purpose of performing studies for 30 days was the analysis of the influence of SRT on microbiological stability of the system.

The influence of SRT on the efficiency of reducing of COD indicates that the retention time of 6 days is insufficient for a stable fermentation: removal of COD is slowed down due to acclimatization of microorganisms and washout of SRB (i), COD values are still relatively high for SRT of 6–12 days: there is an incomplete breakdown of organic compounds (ii), stable fermentation is obtained after 12–20 days: low COD values, active breakdown of organic compounds started (iii), and the breakdown curve stabilizes at SRT 30 days (iv); all sludge organic compounds are significantly reduced (efficient removing of COD_t and COD_s) (Fig. 2). The SRT is a fundamental design and operating parameter for all anaerobic processes. Thus, fermentation was taken for a long period under laboratory conditions. In the industrial technological implementation, the SRT will be reduced in accordance with determined parameters.

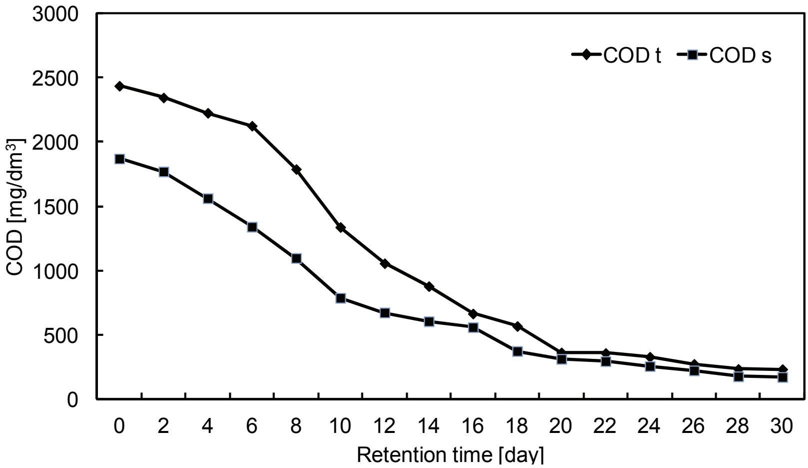

Fig. 2. COD profiles for process AMD within advanced operation of the anaerobic bioreactor

Note that two-step hydrolysis may be a reflection of two classes of organic matter within the sludge, i.e. readily and slowly degradable material. Furthermore, the results suggest that while the readily degradable fraction is removed under both electron acceptor conditions, a larger proportion of the less readily available material is only available under pretreatment conditions.

Solid–liquid separation in the system was good yielding average particulate. Total COD removal of 85% (decreased from 2435 mg COD /dm^3 to 365 mg COD/dm^3) was recorded for the sulfidogenic system over the period of 20 days. The concentration of sulfide (as hydrogen sulfide) increased in the sulfidogenic system to reach the maximum concentration of 778 mg/dm^3 on day 20, indicating that the SRB population was active (Table 3). After day 20, it decreased from 778 mg/dm^3 to 97 mg/dm^3 (day 30). These data is confirmed by inhibition activity of SRB (Table 3).

Sulfate reduction should cause the pH of the medium to increase as the reduction of sulfate results in the formation of sulfide ions. A portion of these ions dissolve in the liquid phase and is involved in chemical reactions with the ions of heavy metals. The remainder escapes as hydrogen sulfide gas.

Each mole of hydrogen sulfide that leaves the liquid phase translates into two moles of hydrogen ions that are removed from the system, corresponding to an increase in pH. This is precisely what was seen in the bioreactor by the day 6, when the pH was beginning to increase. The removal of hydrogen ions from the solution and the corresponding pH increased slowly occurred to 8.0. After 10 day complete oxidation of organic nutrients was carried out by SRB and other sludge bacteria, causing the production of carbon dioxide. Some of this CO_2 dissolved to form carbonic acid and pH had decreased to 6.9 after 20 day. Then the pH decrease became progressively slower. It reached 6.7 on day 30.

The system adapted relatively rapidly to these SRT decrease, indicated by the rapid decrease in influent sulfate. In the anaerobic bioreactor, the elimination of solids increased when they were run at higher SRT. These results demonstrate that the system readily accommodated the decreases in SRT (increase in loading), but required time to develop the acid-forming microorganisms and SRB biomasses. These data, during the 20-days experiment indicate that both the degree and rate of solubilisation of complix organic substrate was improved in the presence of biosulfidogenic activity. Hence, the most efficient SRT required for sludge digestion is 20 days.

Significant presence of SRB is indicated by low effluent sulfate concentrations and good sulfate removal achieved (ca. 94.5% on day 20, Table 3). The most important is that sulfate contained in the phosphogypsum can be freely used by SRB as a mineral substrate for their growth. Phosphogypsum is a class IV of hazard (low hazard). Waste of this class of hazard does not contain toxic substances that can damage normal functioning of the groups of microorganisms in the anaerobic bioreactor. In addition, after fermentation, the concentration of sulfide ions in the liquid fraction of digested waste mixture was 0.10 mg/dm^3 under effluent standard 1 mg/dm^3. Thus, we get pure environmental product.

3.2. DYNAMICS OF HYDROGEN SULFIDE IN THE GASEOUS PHASE

The process of production of hydrogen sulfide was substantiated by the gas chromatographic analysis. The conditions of gas chromatographic analysis were as follows:

- preliminary PLOT column-1: the internal diameter 0.32 mm, length 10 m, sorbent PoraPLOT Q,
- the column-2, the inner diameter of 3 mm, 5 m; sorbent Chromaton N-AW-DMCS,
- the HP-PLOT Mole Sieve column-3: 0.32 mm, length 10 m, fixed solids: molecular sieve,
- the column temperature – 60 °C,
- the carrier gas velocity – 25×10^{-3} dm^3/min,
- the bridge current – 220 mA.

Results of the analysis are given in Table 4.

Table 4

Results of the analysis of the gaseous phase formed during biosulfidogenesis

Compound	Volume [%]
Hydrogen sulfide	48.8±1.56
Carbon dioxide	19.3±1.12
Methane	25.4±1.05
Hydrogen	2.8±0.02
Oxygen	0.36±0.06
Nitrogen	5.34±0.03

For this method, sewage sludge treatment is not basic purpose of biomethane production although methane was present in the gaseous phase. Methane-forming bacteria can develop in parallel with the growth of SRB but they do not dominate under condition when significant amount of sulfate was present in the substratum. Note that investigation of the hydrogen sulfide production is very important for developed technology of sewage sludge detoxification. Therefore, chromatograms with peaks corresponding to hydrogen sulfide were studied for analysis of the dynamics of this gas production changing in time (Fig. 3). In the chromatograms, extension of the peak and corresponding increase in the amount of hydrogen sulfide is clearly visible in the gaseous phase during the AMD.

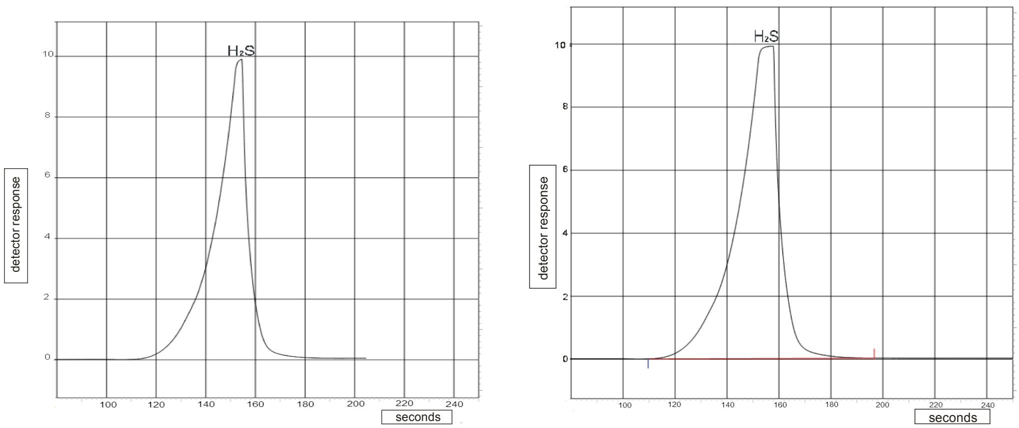

Fig. 3. Chromatograms of the biosulfidogenesis showing the dynamics of product accumulation (sulfide in the gaseous phase) after 16 days (left) and 20 days (right)

SRB is used on organic and mineral substrate (mixture of sludge and phosphogypsum) for biomass growth, that is accompanied by the formation of hydrogen sulfide. Thus the active SRB multiplication in the space of the bioreactor was shown by results of the investigation.

The second stage of the study was estimated the efficient of ozonation. The effectiveness of pre-treatment was analyzed in terms of organic matter (COD) solubilization (increase of soluble COD) and mineralization (removal of total COD). Ozonation treatment of raw sludge caused a transfer of organic substances from the sludge solids into the aqueous phase as demonstrated by an increase of COD. In ozonation, the highest increase of soluble COD (2.5-fold) was achieved.

In the anaerobic bioreactor, after 20 days, the elimination of COD_t and COD_s varied between 78–84% and 83–87%, respectively (Fig. 2), within advanced operation of anaerobic bioreactor. The elimination of COD_t increased with the advanced operation of reactor, regardless of SRT. Table 5 shows the results obtained.

Table 5

Performance of conventional and advanced operation of the anaerobic bioreactor (35 °C)

Parameter	Conventional		Advanced: ozonation pretreatment	
	20 d	12 d	20 d	12 d
Amount of H_2S, mg/dm^3	435	394	778	658
Removal efficiency, %:				
COD_t	62±8	43±9	81±3	57±5
COD_s	64±12	58±13	85±2	65±5

The hydrogen sulfide production after ozonation was 1.67 times higher and organic matter removal more efficient under mesophilic conditions with respect to that in conventional operation of anaerobic bioreactor.

Ozone is a strong cell-lytic agent which can kill the microorganisms in activated sludge and further oxidize the organic substances released from the cells. Of the techniques to disintegrate sludge, ozonation is one of the most effective ways and yields the highest degree of disintegration. After ozonation, the characteristics of the sludge are greatly changed. The sludge flocs are broken into fine, dispersed particles. Flocs integration and solubilization generates a large number of microparticles dispersed in the supernatant in addition to soluble organic substances [7].

Ozonation treatment has two effects: degradation of molecules and cell structures that are undegradable for SRB may increase hydrogen sulfide production; oxidation of organic molecules that are degradable for SRB may increase hydrogen sulfide content. Hence, the use of the sludge pre-treatments prior to the anaerobic digestion process led to higher hydrogen sulfide production and efficiency of organic matter removal in mesophilic conditions. Thus, ozonation is possible to be used in the future as a sludge pretreatment method.

Prerequisites for developing of sludge detoxification method are:

- increasing the land areas for sludge disposal;
- increasing operational costs at the wastewater treatment;
- increasing negative influence of the sludge accumulation on the environment, due to the presence of toxic elements in the sludge.

The solution of sludge deposition problem can be found by the way of the AMD intensification under biosulfidogenic conditions. Thus, development and implementation of co-processing technology of organic and gypsum wastes treatment on the WWTP will provide an opportunity to reduce anthropogenic negative impacts on the environment in the region.

Ozonation can be introduced to the sludge digestion line as a pretreatment stage. As a result, the amount of sludge that can serve as substrate for biogas production is decreased dramatically. Particularly, for AMD, the hydrogen sulfide can be increased. Ozonation adopted as pretreatment before anaerobic digestion is used to enhance the solubility of sludge solids and increase the degree of degradation. The final amount of sludge for disposal and the digestion time can thus be reduced. The schematic process is shown in Fig. 4. The conceptual unit process train would consist of a number of unit processes where heavy metals can be precipitated under the biochemical sulfate reduction. Raw sewage sludge is mixed with phosphogypsum using a mixing mechanism on the pretreatment stage prior to the digestion.

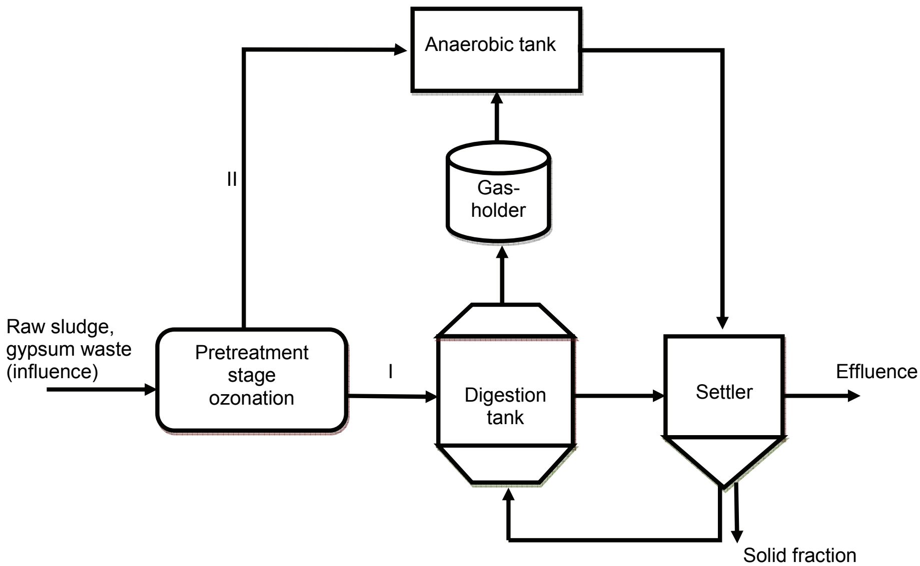

Fig. 4. Aplication of sewage sludge detoxication. The SRB is inoculated in the digester tank

Ozonation is introduced to the sludge digestion line (route I) and to the anaerobic sludge treatment line (route II) (Fig. 4). For route I, ozonation adopted as pretreatment before anaerobic digestion is used to destroy organic polymers, to achieve increase of COD_s and to obtain a homogeneous and stable mixture of waste. It stimulates the initial growth of facultative anaerobic microorganisms and quick SRB biomass growth in the digester tank. For route II, the ozonation basic aim is to enhance the solubility of forms of heavy metals and increase the degree of degradation sludge. In this way, ions of heavy metals more actively react with hydrogen sulfide.

Hydrogen sulfide from a digester tank is passed to a gas-holder for later use in removing heavy metals from the new batch of stable mixture of wastes in the anaerobic tank. Digested mixture goes to the settler for separated to liquid and solid fraction. The part of digested mixture is recycled to digestion tank (Fig. 4).

4. CONCLUSION

The system of anaerobic microbiological degradation (AMD) with the deposition of heavy metals by biogenic hydrogen sulfide is a promising orientation of recoverable resources treatment. The most important is that sulfate contained in the phoshpogypsum can be freely used by SRB as a mineral substrate for their growth.

Following ozonation pre-treatment prior to AMD, the characteristics of the sludge are greatly changes. In ozonation, the highest increase of soluble COD (2.5-fold) was achieved. In the ozonation the sludge, flocs are broken into fine, dispersed particles. In the anaerobic bioreactor, after 20 days of digesting, the elimination of COD_t and COD_s varied between 78–84% and 83–87%, respectively, within advanced operation of anaerobic bioreactor. The sulfate reduction equal to 94.5% in the system was maintained. The hydrogen sulfide production after ozonation was 1.67 times higher and organic matter removal more efficient under mesophilic conditions than that after conventional operation of anaerobic bioreactor.

Efficiency of technological applications of such systems is analyzed. Development and implementation of co-processing technology of organic and gypsum wastes treatment on the WWTP will provide an opportunity to reduce anthropogenic negative impacts on the environment in the region. There is a promising field of environmental management.

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