The Removal of Hydrogen Sulfide in the Biodesulfurization System Using Granulated Phosphogypsum

L.D. Plyatsuk and Ye.Yu. Chernysh

Sumy State University, 2, Rymskogo-Korsakova st., 40007 Sumy, Ukraine

Article info

Received:
10 September 2015

Received and revised form:
18 October 2015

Accepted:
20 November 2015

Keywords:
phosphogypsum, gas purification, biogas, sulfide removal, elemental sulfur

Nomenclature
PG – phosphogypsum
pH – negative log of hydrogen ion activity
CFU – colony forming units
ATP – adenosine triphosphate
EBRT – empty bed residence time
SRB – sulfate-reducing bacteria

Abstract

This paper focuses on the study the possibility of phosphogypsum utilization in the biotechnological processes for hydrogen sulfide removal from biogas. The optimal parameters of the process of granulation dihydrate phosphogypsum were determined. The biochemical characteristics of granular carrier based on phosphogypsum was studied. The efficiency of the gas cleaning under immobilization of the thiobacillus on the surface support medium was analyzed. The main parameters of the gas cleaning process were determined. The degree of H_2S removal from a gas stream was 98.22% at pH = 5.0 and optimum empty bed residence time of 10 h. The possibility of the phosphogypsum using as a new type of mineral support medium for the associations of sulfur-oxidizing microorganisms developing was determined in the process of biological gases purification from sulfur compounds. It was the first time both theoretically and experimentally proved that the mineral support on the basis of phosphogypsum has a sufficient micro and macro elements that necessary for the thiobacteria development. Thus it is eliminating the need to supply additional sources of feeding organisms to biofilter. The gas purification biotechnology with support medium using on the basis of phosphogypsum was developed. This technological solution which allows providing the high quality of gas stream purification with a high content of sulfur compounds, particularly hydrogen sulfide (more than 10% of the gas total volume).

1.Introduction

Raw biogas has produced many applications. However, its hydrogen sulfide content must be eliminated to avoid possible damages to equipment and hazards to users and the environment. Microorganisms have been used for the removal of H_2S from biogas. Ideal microorganisms would have the ability to transform H_2S to elemental sulfur, could use CO_2 as their carbon source, could produce elemental sulfur that is to separate from the biomass, would avoid biomass accumulation to prevent clogging problems, and would be able to withstand a variety of conditions (fluctuation in temperature, moisture, pH, O_2/H_2S ratio, for example). Chemotrophic bacterial species, particularly from the *Thiobacillus* genus, are commonly used. *Chemotrophic thiobacteria* can be used both aerobically and anaerobically. They can utilize CO_2 as a carbon source and use chemical energy from the oxidation of reduced inorganic compounds, such as H_2S [1]. Literature have reported the use of different *Thiobacillus species* (*thioparus, thiobacilli, denitricans, thiooxidans, ferroxidans*) and other microorganisms (*chlorobiaciae, xanthomonas*) converting the H_2S into reduced sulfur compounds [2].

In biological sulfide oxidation end product will be produced based on the oxygen concentration. Under oxygen limiting conditions, that is at O_2 concentration below 0.1 mg/l, sulfur is the end product of the sulfide oxidation, while sulfate is formed under circumstances of sulfide limitation [3].

The THIOPAQ [4] process has been developed to remove H_2S from low pressure biogas streams. In this process, a gas stream containing H_2S contacts an aqueous soda solution containing thiobacillus bacteria in an absorber. System pH ranges from 8.2 to 9. But this diapason of pH in the biofilter is inhibited the growth of many bacterial species of thiobacillus. The sulfur oxidizing bacterium *Thiobacillus sp.* are the gram negative, aerobic, motile cells, grow well at 20–30 °C and a pH of around 2 to 6 [5].

* Corresponding author. E-mail: e.ur.chernish@gmail.com

The soda absorbs the H_2S and is transferred to an aerated atmospheric tank where the bacteria biologically converts the H_2S to elemental sulfur. Treated outlet gas can readily meet a less than 100 ppm H_2S specification (typical requirement for biogas) or as low as 5 ppm. The application range is from approximately 45 kg per day to 20 tons of sulfur per day [4, 6].

Biological oxidation of sulfur granules is a critical component in elemental sulfur fertilizers since it converts sulfur to plant available sulfate [7]. Sulfur is a major nutrient which ranks fifth or sixth in quantity of macronutrients taken up by plants, comparable to the demand of phosphorus. However, most of the sulfur is taken up by plant roots as water soluble sulfate.

The use of support medium for the immobilization of microorganisms is widely known to provide a surface for microbial growth and a shelter that protects the microorganisms from inhibitory compounds. Granular activated carbon often used for immobilization of microorganisms. For removal of hydrogen sulphide the packing material media used in conventional biofilter beds consist mostly of peat and compost, but a wide variety of other materials has been used, ca-alginate, polypropylene pall rings, porous lava, wood, and polyurethane foam [8]. In this way the search for new support mediums is very actually. On the other hand inventing new ways to recycle and reuse waste is a major task today.

Millions of tons of phosphogypsum (PG) is stacked worldwide every year and is progressively considered as an asset more than an environmental burden. PG consists mainly of calcium sulfate dehydrate ($CaSO_4 \cdot 2H_2O$) and contains impurities of not decomposed phosphate, phosphates and silicates. PG is a waste by-product from the processing of phosphate rock by the "wet acid method" of fertilizer production, which currently accounts for over 90% of phosphoric acid production. World PG production is variously estimated to be around 100–280 Mtons per year [9] and the main producers of phosphate rock and phosphate fertilizers are in the USA, China, Russia, Africa and the Middle East. In Ukraine the most common of all kinds of gypsum wastes is PG. PG is formed in an amount of about 100 tons annually in Sumy region (Ukraine). Currently, over 14 million tons of PG was accumulated.

The methods described in the literature to minimize the negative effects of this waste are classified by treatment type, i.e. physical, chemical, thermal, etc., and different suggested applications for PG are detailed [10–13]. PG has been used in the cement industry as a setting regulator in place of natural gypsum [9] and in the gypsum industry to make gypsum plaster. PG also has been used as agricultural fertilizers or soil stabilisation amendments [14, 15]. But only 10–15% of PG is used today.

This paper focuses on the study the possibility of phosphogypsum utilization in the biotechnological processes for hydrogen sulfide removal from biogas. Thus, objectives of this work next:

- research of the PG granulation process;
- analysis of biochemical reactions during anaerobic digestion under bio-sulfidogenic condition;
- immobilization of *Thiobacillus sp.* on granular PG;
- determination of hydrogen sulfide removal efficiencies and the influence on the efficiency of empty bed residence time (EBRT) and culture pH.

Thus the reduction of anthropogenic impact on the environment through the biotechnology development of phosphogypsum utilization in the process of biogas purification from sulfur compounds.

1.1. Experimental setup of granulation phosphogypsum

PG was transferred into a rotating plate granulator. Plate with diameter 250 mm and height 75 mm was made of stainless steel. The rotational speed was varied from plates 50 and 80 rev/min. The time of granulation ranged from 1 to 10–30 min. Dehydrated PG powder was wetted with addition of tap water with hydrated lime in an amount of 3–6% by weight of PG (dry basis). The most significant content of the fractions were spherical granules of size 3–6 mm. Then granules obtained by rounding carefully transferred to a desiccator for a set of strength (d_o8–9 kg/cm^2) and drying to a moisture content of 10%.

1.2. Experimental model of aerobic microbiological hydrogen sulfide removal from biogas

Figure 1 shows the anaerobic bioreactor, a continuous stirred tank reactor with an internal settling zone, that had working volume of 5 dm^3. The biofilter looked like a plexiglass column with volume 2 dm^3, which consist granulated support medium of phosphogypsum. PG granules were inoculated with enrichment culture of sulfideoxidizing bacteria. The biogas from anaerobic bioreactor fed to the bottom of the column through the sleeve. Water was used

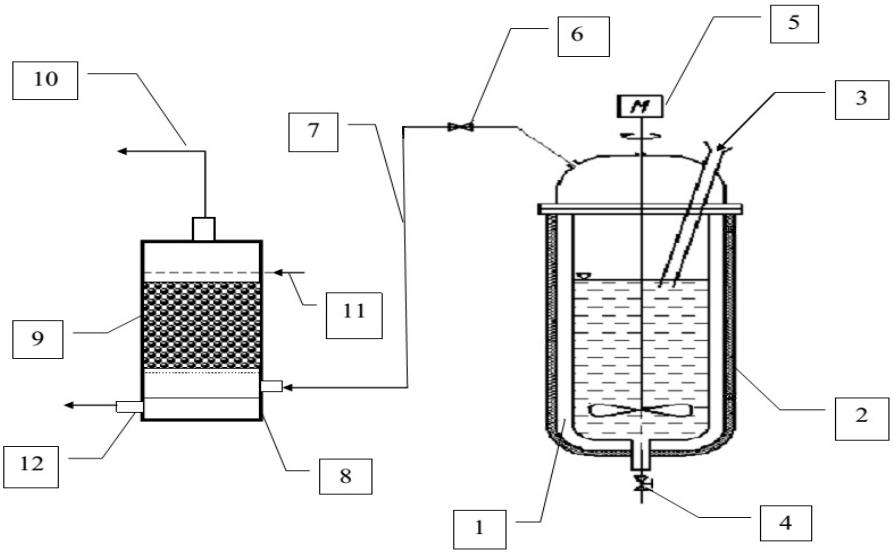

Fig. 1. Experimental model: 1 – anaerobic bioreactor; 2 – insulation; 3 – opening for sewage sludge loading; 4 – port for sludge removal; 5 – mix device; 6 – control valve; 7 – biogas inflow; 8 – biofilter; 9 – granulated phosphogypsum; 10 – purified biogas outflow; 11 – water supply for irrigation; 12 – removal of biosulfur suspension.

in the water feed to the biofilter. Air was bubbled into the water that comes from irrigation system to biofilter. At the top of the column was positioned to drain fitting gas that was clean, and sampling for analysis.

Concentration of H_2S in biogas and bacterial population at a regular interval (5, 10, 15 h) of EBRT consisted of 30 tests for each interval. Concentration of H_2S gas and bacterial populations were measured at optimum EBRT for various pH values (4.0, 4.5, 5.0, 5.5, 6.0 and 6.5) of culture. Each pH value was maintained by 15 h for 1 test. This experiment consisted of 30 tests for each pH value.

1.3. Analytical methods

Study of the gaseous phase was carried out on a laboratory gas chromatograph SelmiChrom-1 (Ukraine). The thermal conductivity detector (katharometer) was used. Argon was chosen as the mobile phase. Compounds are separated by traveling through a coated column and separate based on size and intermolecular interactions. The sample is 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 follow out of the column-1 as a single chromatographic strip, then methane, carbon monoxide (IV) and hydrogen sulfide are separated. The ballast column-2 is filled with an inert carrier "Chromaton N-AW-DMCS". The H_2 and N_2 were separated in the HP-Plot Mole Sieve column-3. Registration and processing of chromatograms was performed by the software Multichrom version 1.52x. Operating temperature range of the thermostat columns from 50 to 400 °C, speed range of linear programming from 5 to 25 °C/min; injection volume (auto sampler) was $0.5 \cdot 10^{-3}$cm^3. Carrier gas of 10 cm^3/min, linear velocity 43 cm/s.

The quality analyses of materials were performed on the scanning electron microscope-microanalyzer REMMA-102 (Ukraine). Measurements were performed at five points on each sample. Processing spectrometric data, perform the necessary calibration measurements, the decryption of X-ray spectra, qualitative and quantitative electron probe analysis were performed using software microanalysis system. The sensitivity of the measurement was at 1%. Additionally surveyed elemental composition of waste by X-ray fluorescence analysis, which made it possible to determine the concentration of elements in the ppm level. Photomicrographs received and processed using the digital output of image «SEO Scan ICX 285 AK-F IEE-1394» and morphometric program «SEO Image Lab 2.0» and on the scanning electron microscope REMMA-102.

pH was analyzed by pX-meter pX-150 (ionometer) (Belarus).

Sieve analysis included the calculation of the number of granules with different size and dimension of the mass fraction of the granules different fractions.

1.4. Microbiological investigation

Isolation of sulfide oxidizing bacteria was done from aerobic sewage sludge collected from municipal wastewater treatment plant. The aerobic sludge samples were collected and screened for the removal of big particles.

Then the sludge is kept in aerobic conditions by continuous aeration in order to prevent growth of any anaerobic bacteria for a period of 10 days at temperature of 35 °C.

The enrichment medium for *Thiobacillus sp.* cultivation has the following composition: NH_4Cl, 1.0 g; K_2HPO_4, 0.6 g; $CaCl_2 \cdot 2H_2O$, 0.2 g; $FeCl_3 \cdot H_2O$, 0.02 g; we tacked such sulfates: $ZnSO_4 \cdot 7H_2O$, 40 mg; $CaSO_4 \cdot 2H_2O$, 70 mg; $MnSO_4$, 15 mg; $Na_2B_4O_7$, 10 mg; distilled water, 1000 cm^3; pH, 5.0.

The enrichment medium for nitrifying bacteria (*Nitrosomonas sp.*) cultivation has the following composition: $(NH_4)_2SO_4$, 2.0 g; K_2HPO_4, 1.0 g; NaCl, 2.0 g; $MgSO_4 \cdot 7H_2O$, 0.5 g; $FeSO_4 \cdot 7H_2O$, 0.01 g; $CaCO_3$, 0.001 g; distilled water, 1000 cm^3; pH, 5.0.

2. Enumeration and Characterization

Five days old cultures of the two isolated strains are studied under scanning electron microscope for morphological characteristics like size and shape and also gram stain. Standard plate count method is used for the colony count at different serial dilution ranging from 10^{-1} to 10^{-10}.

Photomicrographs microbial preparations are also processed by a digital image output system «SEO Scan ICX 285 AK-F IEE-1394» and morphometric program «SEO Image Lab 2.0» (Sumy, Ukraine). Identification of bacterial culture produced by Burge on the basis of morphology, physiology and biochemical properties of microbial cells.

Samples of phosphogypsum granules were taken from the biofilter and made crop on enrichment mediums in Petri dishes in accordance with the procedure described above.

2. Results and Discussion

2.1. Research of the granulation process of phosphogypsum

Figure 2 shows granules (diameter 4–5 mm, humidity 10%) that made on the basis PG. The formation of spherical granules was observed and their average size increase with increasing time of granulation in the range of 0–25 min.

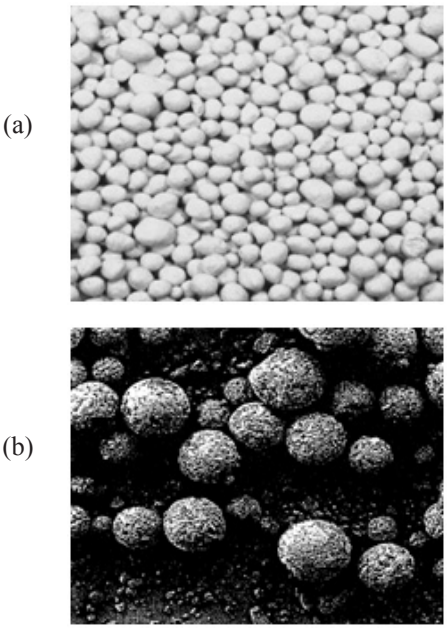

Fig. 2. The granulated phosphogypsum: a – general view; b – granules, magnification: x100.

The optimum granules size with diameter of 4–5 mm was observed under such parameters of PG granulation: PG moisture content of 38% and the addition of hydrated lime in an amount of 5% by weight of PG (Fig. 3). Phosphogypsum was neutralized by lime to pH 5 and phosphogypsum granules became more hydrophobic.

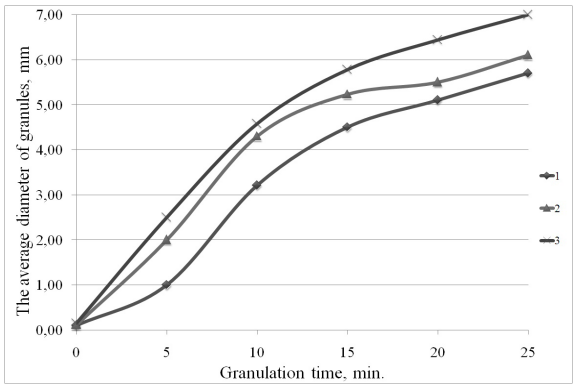

Fig. 3. The average diameter of granules according to granulation time and humidity of dihydrate phosphogypsum 1 – w = 38%; 2 – w = 32%; 3 – w = 26%.

The changing of the average radius of granules during contact time depended on the process of drusen buildup on proto-granule PG by adhesive forces. The interaction of lime with PG was for seal of assemblies of PG under the influence of the intra-pressure crystallization of the resulting gypsum hemihydrate and monohydrate, which contained in PG. Pores overgrown crystals of gypsum

dihydrate and partially cemented existing units of gypsum dihydrate in PG. The particle size distribution was depended on the degree of hydration of PG and residence time in the granulator (Fig. 3). The results obtained are also shown in the Table 1, the physical properties of the granules are described in the Table 2.

Table 1
Mass part of granules fractions depending on humidity

Fraction d_{av}, mm	Mass part of fractions under different moisture of phosphogypsum		
	w = 38 %	*w* = 32 %	*w* = 26 %
<1.0	-	-	-
1-2	<0.01	0.09	<0.01
2-3	0.2	0.2	0.1
3-4	0.3	0.5	0.2
5-6	0.4	0.3	0.4
6-7	0.1	-	0.3

Table 2
The physical properties of granulated phosphogypsum

Parameters	Size
Specific surface area (m^2/g)	215-325
Specific pore volume (cm^3/g)	0.20-0.35
Density (kg/m^3)	635-789
pH (5% aqueous solution)	4.0-5.0
Humidity,%	10-15%
Average particle size (mm)	4-5

The biochemical reactions during anaerobic digestion under bio-sulfidogenic condition

The system of anaerobic digestion with the heavy metals (HM) sedimentation by biogenic hydrogen sulfide is the promising way of sewage sludge detoxification. The biosulfidogenic technology was developed for sewage sludge detoxification together with phosphogypsum in Sumy State University [16]. Thus, we studied biogas that has the quite high of hydrogen sulfide concentration. However, methane present in gaseous phase too (Table 3).

Table 3
The analysis of biogas in the anaerobic fermentation of excess active sludge

The components of biogas	Volume fraction, %
methane	45.1 ± 2.05
carbon dioxide	26.9 ± 6.37
hydrogen sulfide	19.3 ± 5.21
ammonia	4.7 ± 3.07

The final stage of anaerobic digestion is determined by the possibility of microorganisms to use the various terminal electron acceptors. Preference electron acceptors using determined by thermodynamic factors. The firstly reactions begin which have the greatest energetic effect. Reduction of sulfur to H_2S begins the first than reduction of carbon dioxide to CH_4. Decomposition reaction of volatile fatty acids to acetate and hydrogen is thermodynamically unfavorable and can provide microbial growth only with the very low concentration of reaction products. Thus, rapid and complete removal of H_2 must be in the microbial association.

If sulfates are present, sulfate-reducing bacteria (SRB) such as *Desulfovibrio desulfuricans* multiply. Their multiplicationor reproduction often requires the use of hydrogen and acetate – the same substrates used by methane-forming bacteria.

SRB obtain hydrogen and acetate more easily than methane-forming bacteria. It is due to a higher SRB affinity for the substrate. In the microbial association SRB has the ability to the acetate and H_2 remove to a lower threshold than methane-forming bacteria:

methanogenic – 100-10 p.p.m. H_2 (gas)
SRB – 10-1 p.p.m. H_2 (gas)

Thus, sulfidogenesis is more energetically favorable for the microbial association than methanogenesis.

Consequently, the process of methanogenesis is not dominant in anaerobic conditions with constant source of sulphate. The sulfidogenic community of microorganisms is will be developed in the bioreactor.

Note again, the electrons acceptor for SRB were constantly introduced to the medium of bioreactor during investigates. After that the methanogenesis is not dominated and sulfidogenesis was becoming the fourth step of anaerobic digested. For this method of sludge treatment is not basic purpose of biomethane production. But also methane could be present in gaseous phase in that process too.

The process of anaerobic digested was studied for aim to sedimentation of HM by biogenic hydrogen sulfide. Thus, we studied the anaerobic digested under sulfidogenesis. It is not similar to methane fermentation.

Thus, according to substratum-product transformation during anaerobic digestion under bio-sulfidogenic conditions we formed biochemical equations of wastes detoxification on the final digested stage:

$$\begin{cases} CH_3COOH + Y_{X_1/S}Y_{X_1/SO}SO_4^{2-} + Y_{X_1/S}Y_{X_1/NH_3}NH_3 \rightarrow Y_{X_1/s}Y_{X_2/s}C_5H_7O_2N + Y_{H_2S/S}Y_{H_2S/X_1}H_2S \\ \qquad + Y_{X_1/S}Y_{CO_2/(X_1+X_2)}CO_2 + Y_{X_2/S}Y_{CH_4/X_2}CH_4 + \alpha 1_H H_2O, \quad (1) \\ C_5H_7O_2N + Y_{X_1/SO}SO_4^{2-} + \alpha 2_H H_2O \rightarrow Y_{H_2S/X_1}H_2S + Y_{CO_2/X_1}CO_2 + Y_{NH_3/X_1}NH_3, \quad (2) \\ CH_3COO^- + \frac{3}{2}H^+ + SO_4^{2-} \rightarrow \frac{1}{2}H_2S + \frac{1}{2}HS^- + 2CO_2 + HCO_3^- + H_2O, \quad (3) \\ nMeS + mS^{2-} \rightarrow (Me_nS_{n+m})^{2m-} \downarrow, \quad (4) \\ 8HS^- + 9Me^{n+} + SO_4^{2-} \rightarrow 9MeS \downarrow + 4H_2O. \quad (5) \end{cases}$$

Here $Y_{X_1/S}$ and $Y_{X_2/S}$ are yield coefficient, biomass of SRB and methane-forming bacteria, respectively, produced per mass of substrate utilized ($g \cdot g^{-1}$); $Y_{X_1/SO}$ is yield coefficient, biomass of SRB produced per mass of sulfates utilized ($g \cdot g^{-1}$); Y_{X_1/NH_3} is yield coefficient, biomass of SRB produced per mass of ammonia utilized ($g \cdot g^{-1}$); Y_{H_2S/X_1} is yield coefficient, mass of hydrogen sulfide produced by biomass of SRB ($g \cdot g^{-1}$); $Y_{CO_2/(X_1 + X_2)}$ is yield coefficient, mass of carbon dioxide produced by biomass of SRB and methane-forming bacteria ($g \cdot g^{-1}$); Y_{CH_4/X_2} is yield coefficient, mass of methane produced by biomass of methane-forming bacteria ($g \cdot g^{-1}$); Y_{CO_2/X_1} is yield coefficient, mass of carbon dioxide produced by biomass of SRB ($g \cdot g^{-1}$); Y_{NH_3/X_1} is yield coefficient, mass of ammonia decomposition of biomass SRB ($g \cdot g^{-1}$); *Me* is a generic notation of heavy metal ions, *MeS* is a generic notation of metallic sulfides, $a1_H$ and $a2_H$ are stoichiometric coefficients.

Acetates decomposition process to hydrogen sulfide, methane and carbon dioxide for the synthesis of microbial biomass ($C_2H_7NO_2$), and decay of the biomass are presented in the Eqs. (1) and (2). Since the anaerobic digestion was conducted under biosulfidogenic conditions in the Eq. (3) is shown the process sulfate reduction dissimilation of acetate decomposition without methanogenesis. In the Eqs. (4) and (5) are presented the chemical reactions of HM ions precipitation by hydrogen sulfide.

2.2. Immobilization of Thiobacillus sp. on granular phosphogypsum

PG granules were suggested to implement the method of adsorptive immobilization of microorganisms. The humidity of granulated support medium of biofilter was increased, which at the beginning of the experiment was in the process of desulfurization unit. The humidity was 10% at the beginning of the experiment, at 30 days was 15% and at 50 day was 17%. This indicates the saturation of its moisture, not only from irrigation systems, but also moisture contained in the sludge formed during sludge digestion biogas. The water film was formed on the surface granulated support medium of PG. There absorbed hydrogen sulfide and ammonia with subsequent transformation of the association of aerobic microorganisms that due process autoselection changed its species composition (Table 4).

It should be noted that the bacterial matrix penetrated through the fine pores (compared with the size of cells) deep into granules, the cells were subjected to enzyme transformation of mineral components. Thus sulfur was detected on the surface of the granules and subjected to removal. Thus, biosulfur was obtained. This elemental sulfur some different in composition and properties from gaseous sulfur produced using the Claus reaction. Biosulfur contains contaminants in the form of dead biomass of microorganisms and phosphogypsum particles. This sulfur can be used in agriculture.

Table 4
Microorganisms immobilized on granulated support medium of the phosphogypsum in the biofilter of desulfurization unit

The duration of experiments	CFU/g (granules)	
	Sulfideoxidizing bacteria	Nitrifying bacteria
10	10^6	10^5
20	10^7	10^4
30	10^8	10^3
40	10^9	10^2
50	10^{10}	0

In the Fig. 4 can see the clusters bacteria of *Thiobacillus sp.* (including by *T. thiooxidans* and *T. ferrooxidans*) immobilized on the surface of the support medium (phosphogypsum) and biosulfur deposits. The total immobilized biomass at the end of batches was of $1.9–3.7 \cdot 10^{10}$ CFU/g of PG. During period operated desulfurization system this material didn't require regeneration.

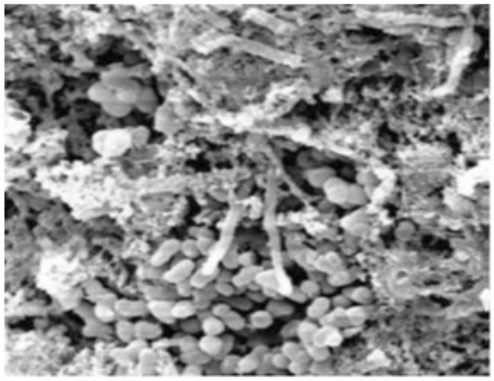

Fig. 4. Scanning electron micrographs of the structure of phosphogypsum granules with *Thiobacillus* biomass during immobilization process. Magnification: 10 μm.

Figure 5 schematically shows the structure of the granules on the basis of PG with formed bacterial matrix. The metabolic reactions of the cell comes through the layer of support material based on phosphogypsum, where was occurred the diffusion-controlled transport of nutrients and discharge metabolites.

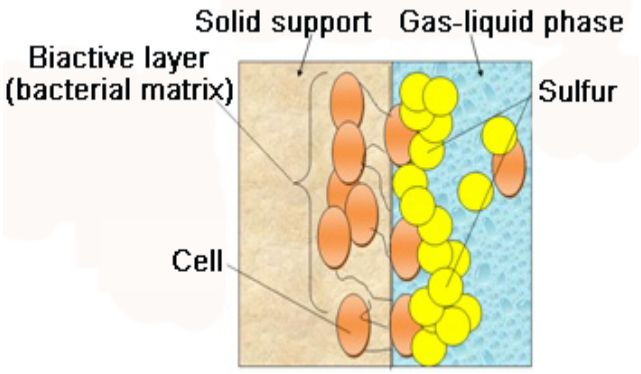

Fig. 5. The structure of the mineral carrier based on phosphogypsum with internal bacterial matrix formed in the process of gas purification.

Bacterial matrix closely associated with the support matrix, there by minimizing removal of the active mass of the sulfide-oxidizing bacteria of the biological filter during washing with loading and removing the surface bio-sulfur. Thus the bioactive inner layer in a matrix of PG during its interaction with the bacterial cells was formed.

2.3. Effect of empty bed residence time and culture pH on H_2S removal from biogas

Removal efficiency is the fraction of the pollutant (H_2S) removed in the biofilter expressed as a percentage. It defined as

$$RE = \frac{\left(C_{g-in} - C_{g-out}\right)}{C_{g-in}} \cdot 100[\%], \qquad (6)$$

where: H_2S concentration at inlet (C_{g-in}) and outlet (C_{g-out}).

Figure 6 shows the depending degree of biogas purification from hydrogen sulfide on pH and empty bed residence time (EBRT) that the gas stream contacted with biomass of bacteria that are immobilized on mineral support of dehydrate phosphogypsum.

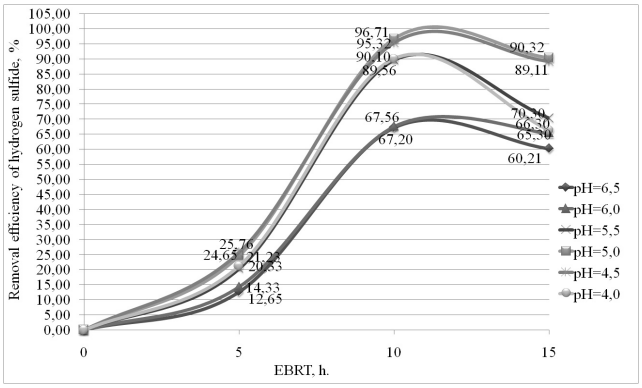

Fig. 6. Dependence of hydrogen sulfide removal efficiency from EBRT: 1 – pH = 6.5; 2 – pH = 6.0; 3 – pH = 5.5; 4– pH = 5.0; 5 – pH = 4.5; 6 – pH = 4.0.

Variations of pH (4.0–6.5) led to changes in the metabolism of *Thiobacillus sp*. Respectively, it led to change of the dynamics of increasing biomass by bacteria and the degree of removal of H_2S. The degree of H_2S removal was increased in the pH range of 4.0 to 5.0. The minimum H_2S removal was only 67.20% for 10 days at initial culture pH increase to 6.5 (Fig. 6). The maximum level of H_2S removal was 98.22% at pH=5.0 and optimum EBRT of 10 h, when bacterial growth reached maximum (3.7·1010 CFU/g).

The results indicate that the displacement of the pH in the acidic side acidophilus group microorganisms developing rapidly with the building of biomass. The degree of removal of hydrogen sulfide increased and reached maximum at pH=5. Removals of H_2S from gas were 24.65%, 96.71% and 90.32% at 5, 10 and 15 h of EBRT respectively. Bacterial desulphurization of H_2S increases with increase of EBRT (10 h) and then declines at 15 h (Fig. 6). This was due to the accumulation of metabolic products of bacteria. The period of lag phase of growth *Thiobacillus sp.* initially depended on the concentration of H_2S and oxygen in the system. Further lowering the pH to 4.0 did not lead to an increase in the degree of hydrogen sulfide removal.

The granulated PG as carrier for microorganisms and as nutrient supplier has the following advantages: it has low cost; it stimulates the development of needful ecological trophic groups of microorganisms; it creates favorable conditions for the formation of biofilm on their surface; the contact surface extends with a gas stream; it is resistant to higher acidity; it has the protection function blocking toxic components; it increases the yield of biosulfur. The natural sorption mechanism is characteristic of bacteria living cells. This mechanism provides important minerals (microelements and macronutrients) for microorganisms in appropriate concentrations, which come from mineral substrate that was PG granules. Cells with specialized transport systems that use energy of ATP hydrolysis provide transport of ions into the cell or selection in the extracellular space. Inside cells metals released in ions or in the form associated with various components of the cytoplasm.

The acidophil association of microorganisms was formed during the desulfurization. This association is able to oxidize hydrogen sulfide to form elemental sulfur (biosulfur) in acid medium.

The advantage of immobilizing bacteria adsorption method on a support of PG is that it allows the binding of bacteria in the mass of the support medium. This creates a stable biomineral structure with effective sulfide conversion to elemental sulfur. The regeneration of granular loading was during its washing with running water and dosed adding of new granules.

Conclusions

The biomass of *Thiobacillus sp.* was immobilized on material medium, which consisted of dehydrate PG. The total immobilized biomass was $1.9–3.7 \cdot 10^{10}$ CFU/g. During period operated desulfurization system this material didn't require regeneration. Bacterial growth reached maximum (3.7·1010 CFU/g) with maximum H_2S removal (98.22%) at initial culture pH 5.0 at optimum EBRT of 10 h. Variations (4.0–6.5) in pH of culture media resulted in changes in activity of *Thiobacillus* and hence bacterial populations as well as H_2S removal.

Desulfurization resulted in forming of acidophilic microorganisms association that is able to oxidize hydrogen sulfide to form elemental sulfur in an acidic environment. Optimum conditions of biological system of hydrogen sulfide removal from biogas were determined. The basic patterns and mechanisms of adsorption immobilization of sulfide-oxidizing bacteria in the mass of mineral support medium made of phosphogypsum were determined for gas purification system. The advantage of the adsorption method on the granulated support medium of PG is that it allows to bind the bacteria in the granules with the bioactive layer formation. Granules based on PG are characterized by permeability to *Thiobacillus sp.* and contain useful minerals for bacteria growth such as calcium, magnesium, phosphorus, etc. Bacterial matrix is closely associated with the support medium matrix.

References

[1]. A.M. Siefers, A novel and cost-effective hydrogen sulfide removal technology using tire derived rubber particles. Graduate Theses and Dissertations. Paper 11281. (2010), 93 p.
[2]. M. Ramirez, J.M. Gómez, D. Cantero. Bioresour. Technol. 100 (21) (2009) 4989–4995.
[3]. P. Ravichandra, G. Mugeraya, A. Gangagni Rao, M. Ramakrishna, and A. Jetty. J. Environ. Biol. 28 (4) (2007) 819–823.
[4]. THIOPAQ Bio-Desulfurization Process. Cameron. Printed in USA, 07/10 TC9814-047 (2010), 2 p.
[5]. A.J. Janssen, S.C. Ma, P. Lens, G. Lettinga. Biotechnol. Bioeng. 53 (1) (1997) 32–40.
[6]. Environmental Technology Verification report. Katec, Inc. Aerosolv®. California Environmental Protection Agency Department of Toxic Substances Control Office of Pollution Prevention and Technology Development Sacramento, California (1999), 54 pp.
[7]. P.H. Nielsen. Water Sci. Technol. 17 (2–3) (1984) 167–181.
[8]. Carrier Byoung-Gi Park, Won Sik Shin, Jong Shik Chung. Environ. Eng. Res. 13 (1) (2008) 19–27.
[9]. A.B. Parreira, Jr, A.R.K. Kobayashi, O.B. Silvestre. J. Environ. Eng. 129 (2003) 956–960.
[10]. D.A. Mays, J.J. Mortvedt. J. Environ. Qual. 15 (1) (1986) 78–81.
[11]. I.S. Alcordo, and J.E. Rechcigl. "Phosphogypsum and other by-products gypsums". In Soil and Amendments and Environmental Quality, J.E. Rechcigl (ed.). pp. 365–425. Boca Raton, FL: CRC/Lewis Publishers. 1995.
[12]. J. Murphy, J.P. Riley. Anal. Chim. Acta 27 (1962) 31–36.
[13]. A.I. Akın, S. Yesim. Cem. Concr. Res. 34 (4) (2004) 677–680.
[14]. N. Degirmenci, A. Okucu, A. Turabi. Building and Environment 42 (9) (2007) 3393–3398.
[15]. C. Papastefanou, S. Stoulos, A. Ioannidou, M. Manolopoulou. J. Environ. Radioact. 89 (2) (2006) 188–198.
[16]. L. Plyatsuk, E. Chernish. J. of Solid Waste Technology and Management 40 (1) (2014) 10–23. http://dx.doi.org/10.5276/JSWTM.2014.10