**Audra SKAISGIRIENĖ**

# INVESTIGATION OF BIOLOGICAL WASTEWATER TREATMENT USING AN ENZYMES PREPARATIONS

**Summary of Doctoral Dissertation**

**Technological Sciences,**
**Environmental Engineering and Landscape Management (04T)**

**1294**

VGTU
LEIDYKLA TECHNIKA
Vilnius 2006

VILNIUS GEDIMINAS TECHNICAL UNIVERSITY

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The summary of the doctoral dissertation was distributed on 21 August 2006.
A copy of the doctoral dissertation is available for review at the Library of Vilnius Gediminas Technical University (Saulėtekio al. 14, LT-10223 Vilnius, Lithuania).

VILNIAUS GEDIMINO TECHNIKOS UNIVERSITETAS

**Audra SKAISGIRIENĖ**

# BIOLOGINIO NUOTEKŲ VALYMO TYRIMAI NAUDOJANT FERMENTINIUS PREPARATUS

Daktaro disertacijos santrauka

Technologijos mokslai, aplinkos inžinerija ir kraštotvarka (04T)

VGTU LEIDYKLA TECHNIKA
Vilnius 2006

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Disertacija bus ginama viešame Aplinkos inžinerijos ir kraštotvarkos mokslo krypties tarybos posėdyje 2006 m. rugsėjo 22 d. 13 val. Vilniaus Gedimino technikos universiteto senato posėdžių salėje.
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Disertacijos santrauka išsiuntinėta 2006 m. rugpjūčio 21 d.
Disertaciją galima peržiūrėti Vilniaus Gedimino technikos universiteto bibliotekoje (Saulėtekio al. 14, LT-10223 Vilnius, Lietuva).

VGTU leidyklos „Technika“ 1294 mokslo literatūros knyga

**General Characteristic of the Dissertation**

***Topicality of the problem***

Along with a rapid development of the industrial and energy sectors the quantities of water used in these sectors including production processes are increasing. With each discharge of sewage both the equilibrium of the water ecosystem and the self–purification processes are disrupted.

A great number of complex live organism systems exist in open water bodies; these systems change in time, while changes in the natural systems may be irreversible. The water bodies‘ protection issues, sewage treatment and investigations into treatment methods are closely interrelated.

Use of active sludge is highly widespread in sewage treatment. The polluting organic substances present in the water are used by active sludge for nutrition. The efficiency of treatment depends on microorganisms' capability to assimilate substrate. This is related to the ability of ferments (or enzymes) to decompose primary macromolecular nutrients to the level where they become accessible, both by size and chemical composition, to microorganism cells. Many vital processes including nutrition, breathing and reproduction are impossible without ferments. They activate both the biochemical synthesis of new compounds in organisms and the decomposition of complex components of the nutrient medium.

Substances often contained in sewage include chlorides, sulfates and oxidising substances. All of them are difficult or impossible to remove from sewage by the biological treatment method, while their effect upon the active sludge microorganisms has not been well studied.

Nitrogen is an important biogenic substance that influences eutrophication of water. Nitrogen in the inorganic form is much more mobile than phosphorus in inorganic form; its sorption into small particles is not so strong, therefore, nitrogen is not removed from the water medium. In order to avoid eutrophication processes in natural water sources, concentrations of nitric compounds in sewage must be reduced to allowable levels.

Energy is a key property of an ecosystem. Part of energy is transformed into action, part is transformed into heat, and part turns into biomass or accumulates in the ecosystem. Analysis of energy flows is necessary to investigate ecosystems and to describe them including their stability more exactly. Trophic links, energy transformations and energy flows in ecosystems are often complex and highly interrelated.

***Aim and tasks of the work*** – to assess the efficiency of and to investigate biological sewage treatment facilities operating under aerobic conditions and using

of ferment preparations and to determine patterns of energy changes in the trophic link of the active sludge system.

- Investigate, both theoretically and experimentally, the concentrations of inorganic pollutants, chlorides, sulfates, inorganic nitric compounds and oxidising substances influencing the efficiency of the treatment process and the growth of active sludge microorganisms; assess changes in the active sludge system.
- Study the effect of a selected ferment preparation upon the quality of active sludge and energy consumption; assess the opportunities for using it in the biological sewage treatment process.
- Make computations of changes in the kinetics of biochemical reactions, such changes taking place in the process of adsorption of polluting substances in the active sludge system.
- Study the efficiency of the ferment preparation in biological treatment facilities.
- Conduct numerical modelling of biological treatment of sewage using a ferment preparation.

***Scientific novelty***

Comprehensive investigation of the effect of polluting substances upon active sludge and of the opportunities for immobilisation of ferment preparations in the process of biological aerobic treatment of sewage.

Refined mathematical models describing the population dynamics and identification of degradation rations based on the results of experimental investigations. The models obtained describe both the active sludge dynamics and the substrate biodegradation reasonably accurately, therefore, they can be applied to computations of sewage treatment in practice.

***Practical value***

The experimental investigations conducted and the results obtained have shown that ferment preparations produced of unprocessed microbial stock can be successfully used for the intensification of the operation of biological sewage treatment facilities and for the active sludge renewal when pollutants of very high instantaneous concentrations having an inhibiting effect upon active sludge get into the treatment facilities.

***The scope of the scientific work.***

The scientific work consists of the general characteristic of the dissertation, 5 chapters, conclusions, list of literature and list of publications. The total scope of the dissertation – 134 pages, 73 pictures and 12 tables.

**Short content of dissertation**

## 1. Investigations of ferment preparation

The mechanism of metabolism and the change of energy in a living cell have been studied in detail in modern biochemistry. Oxygen demand plays central role in these processes. The chemical energy is released in 3 phases – hydrolysis, glycolysis and substances' oxidation to tricarbonic acids. Part of the energy is converted into heat, while the remaining part is consumed in cell formation and accumulation in adenosinetriphosphatases (ATF). This is critical for the breathing process conducted by certain ferments – proteases and oxygenases. The rate of ferment reactions is determined by concentration of oxygen, therefore, the rate of reactions involving ferment under laboratory conditions was calculated. The concentration of substrate varied during the test: 0.8 g/l, 1.2 g/l and 2.0 g/l. The quantity of *Roebic K-47* ferment preparation added – 0.1 ml/l. Based on the results obtained, the dependence of the specific pollutant oxidation rate upon the concentration of dissolved oxygen with varied substrate concentration was determined. The dependence was analysed using a correlation/regression method. As is shown by the charts, the specific pollutant oxidation rate is closely linked (correlation ratio $R^2 = 0.94$) with the concentration of dissolved oxygen, however, the greatest effect is observed when the substrate concentration does not act as a limiting factor. The effect diminishes with the decrease in the substrate concentration.

Microbiological investigations of *Roebic K-47* bacterial preparation have been conducted in the microbiological laboratory of Biocentras UAB. The concentration of viable cells of microorganisms has been established by the Koch method by sowing the preparation out on Oxoid rigid nutrient medium. Pure cultures have been separated out of individual colonies that had grown on the rigid nutrient medium and the capacity of such pure cultures to produce ekzoferment (hydrolase) has been estimated. Concentration of microorganisms has been established upon sowing *Roebic K-47* out on the rigid nutrient medium - 49000 cells/ml, pH = 5.3 and 6 bacterial stems (including two stems of bacteria forming spores) have been identified. The capability of the stems to produce certain ekzoferments (protease, amylase, cellulose, lipase and collagenase) has been studied on rigid media using the following substrates, respectively: casein, starch, cellulose, twin-80 and hide powder. It has been established that the stem IR was most active; it hydrolysed starch, casein and hide powder. Twin-80 has been hydrolysed only by the stem 3R, while cellulose was not decomposed by any of the stems investigated.

## 2. The Influence of Pollutants upon Biological Treatment of Wastewater

Water contaminated with organic pollutants and having stable composition has been used for the experiments. Pollutants selected for the experiments (KCl, $K_2SO_4$, $H_2O_2$, $KMnO_4$) had the same cations and variable cations or differing in their properties. It is known that $Cl^-$ crosses the membrane and destroys the active sludge system, sulfates increase acidity of the medium and inhibit self-purification processes, while $H_2O_2$ and $KMnO_4$ have been selected as oxidising substances that may stimulate or decompose the sludge system. The effect of the pollutants upon the biological oxidation processes and the decomposition of organic pollutants has been established during the investigations. It has also been established how the pollutants influence the energy circulation in the active sludge system and the amount of energy in sewage both before and after treatment. Changes in the load with organic substances have been computed. The active sludge load influences the oxygen demand, active sludge settlement characteristics, and the amount and age of excess sludge. Concentrations of pollutants that produce an irreversible impact upon active sludge have been established.

The demand of biogenic substances in sewage treatment is unstable as the microorganism increment depends upon the composition and concentration of pollutants being removed. However, the amount of biogenic substances in the water after treatment is a very important factor. Experiments have been conducted involving the addition of $(NH_4)_2SO_4$ and $KNO_3$ nitric salts of different concentrations.

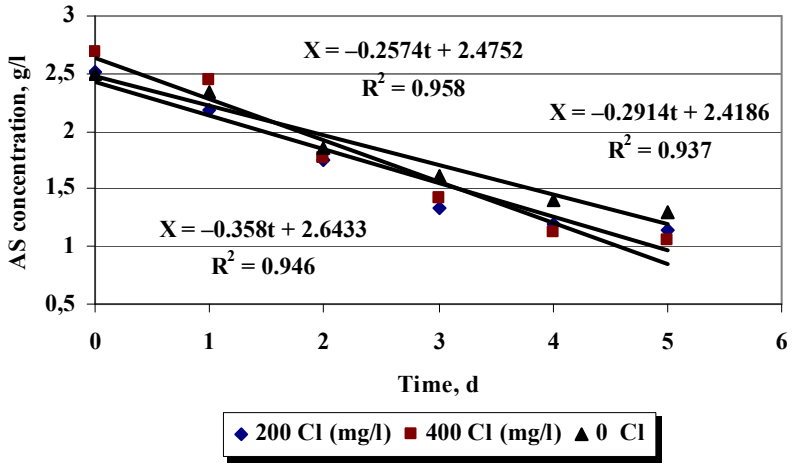


**Fig 1.** Dependence of activated sludge (AS) concentration dynamics upon chloride concentration, without enzymes

The following concentrations have been selected for the experiment with chlorides ($Cl^-$): 200 mg/l, 400 mg/l and 700 mg $Cl^-$/l. The results of this experiment are shown in Figures 1 and 2.

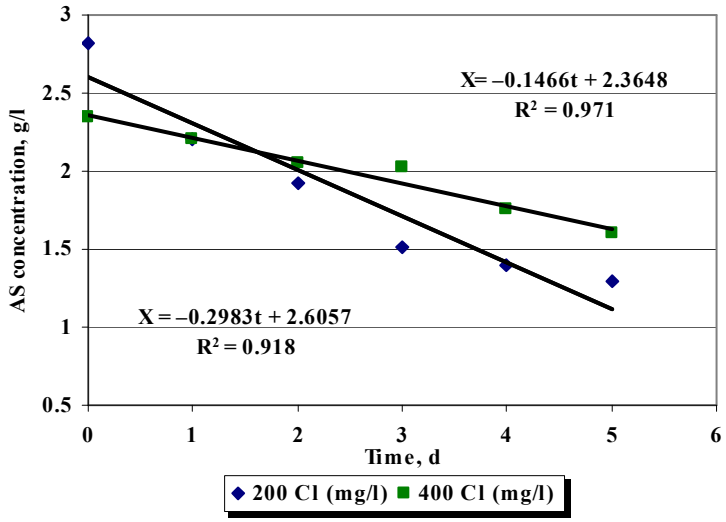


**Fig 2.** Dependence of activated sludge concentration dynamics upon chloride concentration, with enzymes

Upon computation of the active sludge reduction ratio (degradation ratio $k_d$) based on straight-line dependencies it is seen that the reduction of the active sludge concentration is slower with the chloride concentrations studies. For the experiment with potassium sulfate $K_2SO_4$, two sulphate concentrations (300 mg $SO_4^{-2}$/l and 700 mg $SO_4^{-2}$/l) have been chosen. This experiment was aimed to determine the effect of sulfate ions upon the viability and efficiency of the active sludge microorganisms.

To determine the effect of the ferment preparation, the active sludge concentrations obtained during the two experiments (with and without ferments) with the sulphate concentration in sewage equal to 300 mg/l have been compared. The straight-line dependencies shown in Figure 3 are parallel, therefore, one may assert that the ferment preparation had no effect upon the active sludge concentration.

Aerobic organisms have a specific requirement of the free oxygen quantity, therefore, excessive concentrations of oxidising substances do not stimulate the growth of microorganisms; on the contrary – suppress it, sometimes even totally.

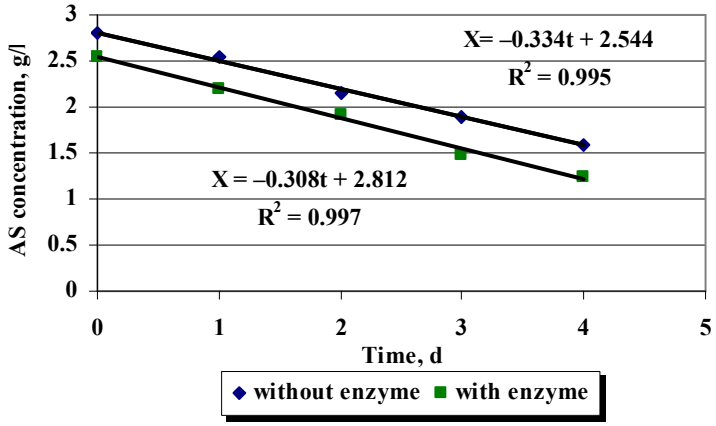


**Fig 3.** Dynamics of activated sludge concentration (sulphate concentration 300 mg/l)

To investigate the effect of hydrogen peroxide ($H_2O_2$) upon active sludge, three concentrations have been selected: 1.46 g/l, 2.92 g/l and 5.84 g/l. The latter two concentrations have a negative effect upon active sludge: the results of the experiment have shown a decrease in the concentration. Upon introduction of the ferment preparation the active sludge concentration increased with any hydrogen peroxide concentration (Fig 4).

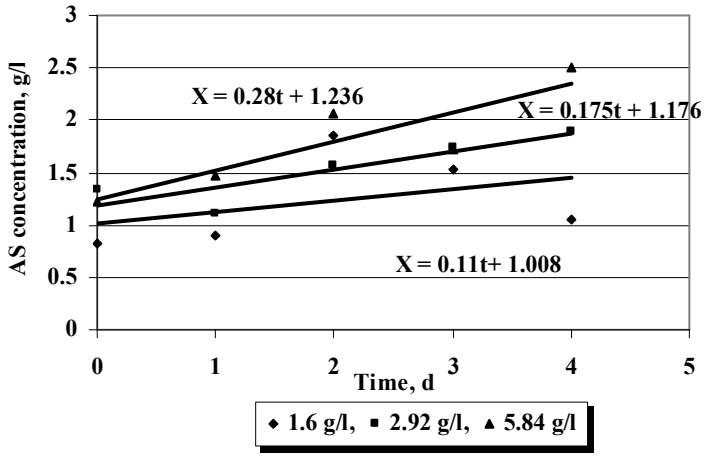


**Fig 4.** Dependence of activated sludge concentration dynamics upon hidrogen peroxide concentration, with enzymes

In this case a positive influence of the ferment preparation is observed. Catalase is a ferment contained in aerobic cells and decomposing hydrogen peroxide into molecular oxygen and water. The main function of the ferment is to protect cells against toxic impact of hydrogen peroxide. This could be explained by the fact that the active sludge system reacts instantaneously to the strong impact on the part of the environment by changing the ferment concentration in cells and starts the synthesis of new ferments corresponding to the conditions of the environment.

Potassium permanganate ($KMnO_4$), just as hydrogen peroxide ($H_2O_2$), is characterised by bactericide and acidifying properties. During the experiment with potassium permanganate its effect upon both active sludge and biological treatment processes has been investigated. Three concentrations were used in the investigations: 0.01; 0.02 and 0.03 g/l. A gradual deterioration of the quality of treatment according to $BOD_5$ was observed, with no positive effect of the ferment preparation.

The following concentrations of biogenic substances have been chosen: $(NH_4)_2SO_4$ – 0.033; 0.067 and 0.10 g/l; $KNO_3$ – 0.05; 0.075 and 0.01 g/l. The results obtained show the effect of the ferment upon the decomposition of nitric substances. The efficiency of treatment has been computed for total nitrogen $N_B$.

The energy of organic substances used is the source of energy for the active sludge microorganisms. Upon completion of the experiment and analysis of its results one can see that any change in a system component causes significant changes in the entire system.

Active sludge microorganisms are sensitive to different changes in technological conditions. They react by a “qualitative” and “quantitative” strategy and may change ferment microsurroundings or even the metabolic process as a whole. It was possible to determine these changes by recording the biochemical oxygen demand and by computing energy transformations in the system. Based on the active sludge load computation the dependencies among the energy transformation, the active sludge load and the treatment quality have been estimated.

The most significant negative impact upon BOD quality has been observed in case of contamination with potassium permanganate, sulfates and hydrogen peroxide.

No effect of the ferment preparation upon treatment quality has been observed in case of potassium permanganate. A marked positive effect of the ferment has been observed while using hydrogen peroxide. The sewage treatment quality is improved up to 24 % as compared to the results of the experiment without the ferment, while energy consumption increases up to 60 %. The proportion of energy used for breathing varies from 19 to 21 %. One may state that the microorganisms

were active as the level of energy consumption was quite high (particularly with the ferments used).

Microorganisms are particularly sensitive to changes in osmotic pressure. Amounts of mineral salts or other substances larger than those inside microorganism cells cause plasmolysis. Water contained in the cell diffuses to the medium due to different substance concentration in the environment and in the cell, while the cytoplasm shrinks, wrinkles, comes off the membrane and the wall; nutrients do not get inside the cell and the microorganisms perish. This process has been observed during the experiment with chlorides. Certain microorganisms have mutated trying to adapt to the new conditions (the entire metabolic process underwent changes).

The presence of sulfates in sewage deteriorates the quality of treatment by as much as 33 % as compared with the experiment involving contamination with chlorides and as much as 50 % when the active sludge nutrition consisted only in artificial sludge.

Potassium permanganate slows down the active sludge's live processes significantly, treatment quality deteriorates, and energy consumption decreases. The reduction in the use of energy for breathing is particularly marked (only 15–17 %) with the potassium permanganate concentration 0.03 mg/l. Both total energy consumption and decomposition levels are also very low.

## 3. Microbiological Analysis of Activated Sludge

It is also possible to determine the quality of sewage treatment by certain indicator microorganisms. For this purpose a microbiological analysis of active sludge has been carried out during the experiments. The protozoa species have been described based on the methodology established for the technological operation control of urban sewage treatment facilities.

Active sludge microorganisms are sensitive to different violations of the set technological conditions and the related deterioration of the sewage treatment quality. An experiment has been conducted by taking samples from reservoirs operating with and without the ferment preparation respectively; chlorides and sulfate salts were also added to the nutrient substrate.

Large flakes, abundant filamentous bacteria and a great variety of microorganisms (*Vorticella convalaria*, *Notommata ansata, Cathlypna luna, Opercularia glomerata)* have been observed in the imported active sludge. The indicator microorganisms were mobile but extremely small. Filamentous bacteria assist good quality of sewage treatment; however, they slow down active sludge settlement considerably and make its separation from the treated water difficult.

During the experiment with chlorides, the quantity of filamentous bacteria was markedly reduced, which can be related to changes in the nutrient medium as

microorganisms are very sensitive to changes in the environment. *Rotatoria* have been found in the samples; *Ciliata* were less frequent. The *Rotatoria* perish in a shrunken state as a result of a sudden change in the chemical composition of sewage. Protozoa, which are indicators of the cleanliness of water, are found in sewage treatment facilities. Active sludge of good quality (1g) containing 1 million bacteria must have 10–15 protozoa. In reservoirs with the $Cl^-$ concentration level of 400 mg/l the number of both the protozoa species and the protozoa was smaller. The ciliary zone of infusoria was closed, especially in the reservoir operating without the ferment preparation.

Microorganisms are particularly sensitive to changes in osmotic pressure in the medium. Amounts of mineral salts or other substances that are larger than those inside microorganisms cause plasmolysis. KCl and NaCl salts increase the osmotic pressure in the environment, making development of microorganisms difficult. Furthermore, upon dissociation of molecules of these salts, the microorganisms are negatively affected by $Cl^-$ ions. Chloride salts reduce the oxygen solubility in water and increase the microbes' sensitivity to carbon dioxide.

Upon introduction of $K_2SO_4$ the number of filamentous bacteria has decreased insignificantly. Infusoria dominated the species composition of microorganisms. An analysis of imported active sludge has shown that the ciliary zone of the infusoria was slightly open and the cilia were moving actively. With the sulfate concentration of 300 mg/l – the microorganisms were small in size but quite active. Upon increase of the concentration up to 700 mg/l, the amount of oxygen in the active sludge decreased. When there is a lack of oxygen in water the stems of vorticella become more transparent and the head breaks loose. If the amount of oxygen in water falls 2 mg/l the vorticella's head inflates and explodes.

The quantity of microorganisms in active sludge decreases considerably along with the increase of the quantity of salt in water. One may assume that, due to their capability of making the medium more acid, the sulfates damage the active centres of ferments given off by the microorganisms thus disrupting the nutrition process. For this reason the larger part of the active sludge microorganisms have perished, while the remaining ones have been inactive and the ciliary zone closed.

## 4. Modelling of the Biological Sewage Treatment Process

A logistical differential equation proposed by P. F. Ferchiulst has been applied to describe the increase in the active sludge concentration with the substrate amount being unlimited.

$$X(t) = \frac{KX_0}{X_0 + (K - X_0)e^{-\lambda K(t-t_0)}}, \tag{1}$$

where $X_0$ is initial concentration of activated sludge.

The analytical solution of the equation shows that the activated sludge concentration reaches its maximum after 9–10 days.

J. Monod's degradation ratio has been introduced in the model of growth of bacterial cultures:

$$\frac{dX}{dt} = \mu X - k_d X, \quad (2)$$

where $X$ is microbial concentration, $\mu$ is the specific growth rate coefficient, $k_d$ is the decay coefficient.

Upon solving the differential equation and adding the $k_d$ dependence upon polluting substances the following analytical solution of the equation has been obtained:

$$X = X_0 \cdot e^{[0.071-(0.4075c^2+0.085c+0.2574)]t}, \quad (3)$$

where $X_0$ is initial concentration of activated sludge; $c$ is pollutance concentration (Fig 5).

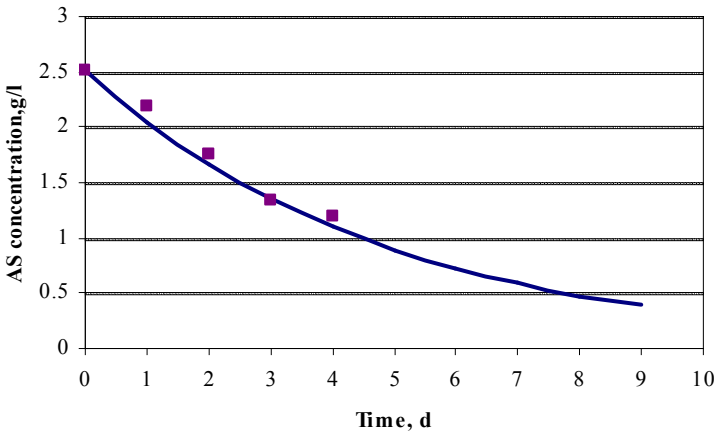


**Fig 5.** The model of activated sludge concentration dynamics upon chlorides concentration

Figure 5 shows a model of the decrease in the active sludge concentration in

time marked by a line, while the concentrations obtained on certain days of the experiment are marked by dots. The points of the modelled curve nearly coincide with the results of the experiment, therefore, one may state that this model is suitable for modelling the dynamics of the active sludge population at different chloride concentrations.

An analogous model has been formed and compared with the experimental results by adding sulfates of different concentrations to the substrate.

The kinetics of the microorganism growth is closely related to the decrease in the substrate concentration (biodegradation). A system of equations has been used for the modelling of the substrate's biodegradation:

$$\begin{cases} \dfrac{dX}{dt} = \mu X - k_d X, \\ -\dfrac{dS}{dt} = \dfrac{\mu_{max} XS}{Y(K_S + S)}, \end{cases} \tag{4}$$

where the rate of the substrate's biodegradation depends upon the mass balance of the active sludge microorganisms' growth. A solution for this system of equations has been found upon solving it by the Runge-Kutt numerical method (Fig 6).

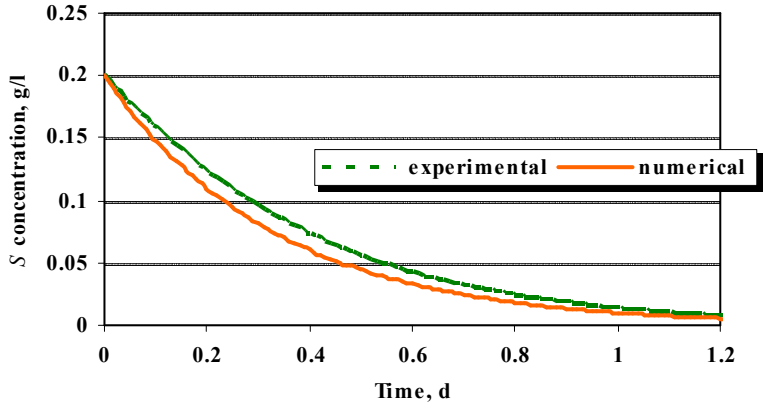


**Fig 6.** The dynamics model of substrate (*S*) biodegradation (4 equations)

Two curves representing the substrate dynamics have been compared in Figure 6. One has been derived from the results of the experiment and it is assumed that it adheres to the law of exponent (5):

$$S = S_0 \cdot e^{-\alpha t}. \tag{5}$$

Another curve has been obtained upon solving the system of equations by the Runge-Kutt method. As it is seen the curves are similar in character and one may assume that the description of the substrate biodegradation provided by both methods is sufficiently accurate.

## 5. The Use of the Bacterial Ferment Preparation at Salantai Wastewater Treatment Plant

To test the production efficiency of *Roebic K-47* ferment preparation, Salantai town biological wastewater treatment plant, which are small in size and the capacity of which is around 150 $m^3$ per day, have been selected.

The technological scheme of the facilities is as follows. Sewage from the sandtrap is delivered to a two-level treatment facility. The first level consists of an aeration channel (inoperative at present), while the second one of an aeration tank. The aeration tank runs in the periodic mode of operation, performing also the function of a settlement tank. To improve both oxygen solubility and mixing of active sludge, two mechanical aeration units installed horizontally on two buoys are used. The time of settlement of active sludge is 2 hours. Upon separation from active sludge, the water after treatment is discharged to the Salantas stream.

Upon estimating the efficiency of sewage treatment at Salantai treatment facilities it has been established that organic pollutants account for the largest part of polluting substances discharged to the Salantas stream. The lowest evaluation results from total phosphorus values. Quantities of phosphorus are increasing both in the inflow and in the water after treatment discharged to the stream.

An analysis of the operation of the facilities has shown that the natural ecosystem's capability to absorb both water and nutrient flows had been destabilised. After adding ferments the process of decomposition of complex organic substances into simpler ones is speeded up and the latter are more effectively absorbed by microorganisms. 10 litres of the ferment preparation were added to the aeration tank with the working volume of 552 $m^3$, i. e. concentration equal to 0.05 mg/l. Sewage samples ($BOD_7$, COD, detergents, total phosphorus and total nitrogen) were taken weekly during two months and the results of the experiment were compared with the data collected over recent years.

The results of the investigation have shown that upon adding the ferment preparation to the facilities $BOD_7$ values have reduced nearly by half. The efficiency of sewage treatment according to $BOD_7$ has increased from 82.6 % to 91.7 %; the use of *Roebic–47* has enabled to improve the efficiency according to COD by 2.5 times; the efficiency has increased from 84.1 % to 93.4 %. The best results have been shown by analyses of total phosphorus and phosphates, with the treatment efficiency increased by 47 % and 29 % respectively.

An analysis of the results of the experiments shows that *Roebic–47* ferment preparation is beneficial as it increases the degree of removal of pollutants that have a negative effect upon processes taking place in water bodies.

**General conclusions**

1. During the experiments the pollutant concentrations influencing the biological treatment process have been established: chloride concentrations of 400 mg/l and higher disrupt the activity of microorganisms; sulfates are characterised by acidifying properties, with the concentrations exceeding 700 mg/l violate the active centres of ferments; concentrations of hydrogen peroxide up to 1.46 g/l improve the quality of the biological sewage treatment on average by 25 % according to $BOD_5$, when the concentration is 8.76 g/l, microorganisms perish instantaneously, 0.04 g/l of permanganate has slowed down both microorganisms activity and vital processes significantly.
2. Treatment of active sludge microorganisms with oxidising substances has causes qualitative changes in active sludge: the sludge flakes have become smaller, the number of protozoa has decreased and they have become smaller in size, the sludge settlement speed has increase, and the number of filamentous bacteria has been reduced.
3. The effect of ferment preparations has been examined: upon adding of sulfate and permanganate salts (from minimum to maximum concentrations), the ferment produced no effect upon the dynamics of the active sludge concentration, biodegradation of the substrate, and accumulation of energy in the system. The effect of the ferment has been observed at higher concentrations of hydrogen peroxide. With $H_2O_2$ concentration of 2.92 g/l consumption of energy increases up to 60 %. As the number of nutritive links increases the biomass growth is reduced and the energy consumption increases.
4. The dependence between the energy and the biomass increment in the active sludge ecosystem has been established: the ferment preparation lends more energy to the system (up to 20 kcal). During the experiment involving sulfates the scope of energy accumulation is smaller from 5 to 40 kcal compared to the experiment with chlorides. Permanganate reduces energy consumption in

active sludge.

5. A modelling of the dynamics of the active sludge biocenosis in the aerobic sewage treatment process and of the substrate biodegradation has been carried out: the dynamics of the active sludge concentration has been modelled based on chloride and sulfate concentrations and a solution of the differential equation identifying biodegradation ratios has been obtained. A substrate biodegradation model including a description of dynamics of the active sludge concentration and of the system of equations for the substrate biodegradation has been formulated.
6. *Roebic–47* ferment preparation has been tested at Salantai sewage treatment facilities: $BOD_7$ values after treatment were reduced almost by half, sewage treatment was improved 2.5 times according to COD, and the efficiency increased by 9.3 %. The detergent concentration has been reduced by half compared to baseline data, while the efficiency of sewage treatment increased by 23.3 % based on detergent values.

**Recommendations**

1. The experimental investigations and their results have shown that ferment preparations produced of microbial stock can be successfully used to intensify the operation of biological sewage treatment facilities.
2. It is recommended, based on the results of investigations into the dynamics of the active sludge biocenosis, that ferment preparations are used for the sludge renewal when polluting substances of high instantaneous concentrations, producing an inhibitory effect upon active sludge, get into treatment facilities.
3. Ferment preparations should be used in aerobic treatment facilities using active sludge and running in the periodic mode.
4. The recommended concentration of ferment preparations is 1.2 ml/l of sewage being treated; the dissolved oxygen concentration in a reservoir with active sludge should be maintained at 3–4 mg/l as the highest specific speed of pollutant oxidation is achieved at this concentration. Both ph and temperature of the medium should be optimal for the activity of microorganisms.
5. It is recommended that the active sludge dynamics model and the substrate biodegradation models, which were refined using the results of the experiments, are used for sewage treatment computations in practice.

***Published works on the topic of the dissertation***

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Wastewater. In: Proceedings of 6th International Conference "Environmental Engineering". Vilnius: Technika, Vol 1, 2005, p. 267–271. ISBN 9986-05-850-3.

***In journals included into Lithuanian list or International data bases***

1. Skaisgirienė, A.; Lapinskienė, A. Investigation of Ferment Preparation Influence upon the Quality of Biological Wastewater Treatment. *Journal of Environmental Engineering and Landscape Management*, Vol XI, No 3, Vilnius: Technika, 2003, p 126–132.
2. Skaisgirienė, A.; Vaitiekūnas, P.; Zabukas, V. Influence of Chlorides and Sulphates on Quality of Biological Wastewater Treatment Using Enzyme Preparations. *Journal of Environmental engineering and Landscape Management*, Vol XII, No 3, Vilnius: Technika, 2004, p. 91–95.

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jaunųjų mokslininkų konferencijos „Lietuva be mokslo – Lietuva be ateities“, Aplinkos apsaugos inžinerijos sekcijos pranešimų medžiaga). Vilnius: Technika, 2004, p. 63–70.
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**About the author**

Audra Skaisgirienė was born in Kaišiadorys, on 28 of december 1959.

In 1978, she entered Kaunas University of Technology (contemporary Kaunas A. Sniečkus Politechnical institute), Faculty of Civil Engineering and Architecture. In 1983 graduated the University and got qualification of engineer and technologist of water and wastewater treatment. 1983–1986 m. worked at the engineering centre “VIKTA”, she took the place of engineer–chemist, 1986–1995 m. – manageress of chemical laboratory. In 1996–2001 she worked at the department of Technological Process of Klaipėda University as an assistant; in 2001–2006 – as a lecturer. In 2003–2006 is a PhD student at the department of Environmental Protection of Vilnius Gediminas Technical University.

**BIOLOGINIO NUOTEKŲ VALYMO TYRIMAI NAUDOJANT FERMENTINIUS PREPARATUS**

***Mokslo problemos aktualumas***

Sparčiai vystantis pramonei ir energetikai dideli kiekiai vandens, naudojami ne tik buities reikmėms, bet ir pramonei, energetikai, gamybos technologiniuose

procesuose. Kiekvienas nuotekų išleidimas sutrikdo vandens ekosistemos pusiausvyrą ir savaiminius apsivalymo procesus.

Atviruose vandens telkiniuose egzistuoja daug sudėtingų gyvųjų organizmų sistemų, kurios kintant aplinkai keičiasi, o natūralių gamtinių sistemų pokyčiai gali būti negrįžtami. Vandens telkinių apsaugos problemos, nuotekų valymas, valymo metodų tyrimai yra tarpusavyje glaudžiai susiję.

Nuotekų valymui labai paplitęs veikliojo dumblo naudojimas. Veiklusis dumblas vandenyje esančias teršiančias organines medžiagas naudoja maistui. Valymo efektyvumas priklauso nuo mikroorganizmų gebos pasisavinti substratą. Tai susiję su fermentų (enzimų) savybe skaidyti pradines stambiamolekulines maisto medžiagas iki pagal dydį ir cheminę sudėtį prieinamų mikroorganizmų ląstelėms. Daugelis gyvybinių procesų (mityba, kvėpavimas, dauginimasis) nevyksta be fermentų pagalbos. Jie aktyvina organizmuose vykstančią biocheminę naujų junginių sintezę bei sudėtingų mitybinės terpės komponentų ardymą.

Chloridai, sulfatai, oksogenai – tai dažnai į nuotekas patenkančios medžiagos. Šios medžiagos biologiniu valymo būdu iš nuotekų pašalinamos sunkiai arba nepašalinamos, o jų poveikis veikliojo dumblo mikroorganizmams labai mažai ištirtas.

Azotas – tai svarbi biogeninė medžiaga, kuri daro įtaką vandenų eutrofikacijai. Neorganinės azoto formos yra daug mobilesnės nei neorganinio fosforo, jos taip stipriai nesisorbuoja į mažas daleles, todėl iš vandens terpės nepasišalina. Kad išvengtume eutrofikacinių procesų gamtiniuose vandens šaltiniuose, būtina pašalinti azoto junginius nuotekose iki leistinų koncentracijų.

Energija – viena iš svarbiausių ekosistemos savybių. Dalis energijos paverčiama darbu, kita – šiluma, trečia – pereina į biomasę ar tiesiog kaupiasi ekosistemoje. Norint ištirti ir tiksliau aprašyti ekosistemas, jų stabilumą būtina išanalizuoti energijos srautus. Trofinės grandys, energijos virsmai ir energetiniai srautai ekosistemose dažnai būna sudėtingi ir persipynę.

***<u>Darbo tikslas ir uždaviniai</u>***

Tikslas – biologinių nuotekų valymo įrenginių, dirbančių aerobinėmis sąlygomis ir naudojant fermentinius preparatus, efektyvumo įvertinimas, tyrimai ir energijos pokyčių dėsningumų nustatymas veikliojo dumblo sistemos trofinėje grandyje. Darbo uždaviniai:

- Teoriškai ir eksperimentiškai ištirti neorganinių teršalų, chloridų, sulfatų, azoto neorganinių junginių bei oksiduojančių medžiagų koncentracijas, įtakojančias biologinio valymo proceso efektyvumą ir veikliojo dumblo mikroorganizmų augimą bei įvertinti veikliojo dumblo sistemos pokyčius.
- Atlikti pasirinkto fermentinio preparato įtakos veikliojo dumblo kokybei ir energijos sunaudojimui tyrimus ir įvertinti jo panaudojimo galimybes biologinio

nuotekų valymo procese.

- Atlikti biocheminių reakcijų kinetikos pokyčių skaičiavimus, kurie vyksta veikliojo dumblo sistemoje adsorbuojant teršiančias medžiagas.
- Atlikti fermentinio preparato efektyvumo tyrimus biologinio valymo įrenginiuose.
- Atlikti biologinio nuotekų valymo efektyvumo panaudojant fermentinį preparatą skaitinį modeliavimą.

***Mokslinis naujumas***

Kompleksiškai ištirtas teršiančių medžiagų poveikis vekliąjam dumblui bei fermentinių preparatų įmobilizavimo galimybės biologiniame aerobiniame nuotekų valymo procese.

Patikslinti matematiniai modeliai aprašantys populiacijos dinamiką, remiantis eksperimentinių tyrimų rezultatais identifikuoti degradacijos koeficientai. Gauti modeliai pakankamai tiksliai aprašo tiek veikliojo dumblo dinamiką, tiek substrato biodegradaciją, todėl gali būti pritaikyti praktiniam nuotekų valymo skaičiavimui.

***Praktinė vertė***

Atlikti eksperimentiniai tyrimai bei gauti rezultatai parodė, kad fermentinis preparatas, pagamintas iš neapdorotos mikrobinės žaliavos, gali būti sėkmingai naudojamas biologinių nuotekų valyklų darbo intensyvinimui, bei veikliojo dumblo atsinaujinimui, kai į valyklas patenka labai didelės momentinės koncentracijos teršiančių medžiagų, turinčių inhibitorinį poveikį veikliajam dumblui.

***Darbo aprobavimas***

Darbo rezultatai paskelbti 3 straipsniuose, recenzuojamuose periodiniuose leidiniuose, įtrauktuose į LMT patvirtintą bazių sąrašą. 2 straipsniai Tarptautinių mokslinių konferencijų leidiniuose ir 7 straipsniuose respublikinių mokslinių konferencijų ir kituose leidiniuose.

***Apimtis ir struktūra***

Disertaciją sudaro įvadas, 5 skyriai, darbo rezultatų apibendrinimas ir rekomendacijos, bendros literatūros, autorės publikacijų bei skaitytų pranešimų darbo tematika sąrašai. Darbo apimtis – 134 teksto puslapiai, 73 paveikslai, 12 lentelių.

**Darbo rezultatų apibendrinimas**

1. Eksperimentų metu nustatytos tiriamųjų teršalų koncentracijos, įtakojusios biologinį valymo procesą: chloridų koncentracija 400 mg /l ir didesnė sutrikdo

mikroorganizmų veiklą; sulfatai pasižymi terpę rūgštinančiomis savybėmis, todėl jų koncentracijos daugiau kaip 700 mg/l pažeidžia aktyviuosius fermentų centru; vandenilio peroksido koncentracijos nuotekose iki 1,46 g/l pagerina biologinio vandens valymo kokybę vidutiniškai iki 25 % pagal $BDS_5$, kai koncentracija 8,76 g/l mikroorganizmai žūva; 0,04 g/l permanganato labai sulėtina mikroorganizmų veiklą ir gyvybinius procesus.

2. Veikiant oksidatoriais veikliojo dumblo mikroorganizmus vyksta šie dumblo kokybiniai pokyčiai – dumblo dribsnių susmulkėjimas, pirmuonių skaitlingumo sumažėjimas bei smulkėjimas, padidėjęs dumblo sėdimo greitis, siūlinių bakterijų sumažėjimas.
3. Kompleksiškai ištirtas fermentinio preparato ir druskų poveikis: įvedus sulfato ir permanganato druskas (nuo minimalios iki maksimalios tirtos koncentracijos) fermentinis preparatas veikliojo dumblo koncentracijos dinamikai ir substrato biodegradacijai bei energijos akumuliacijai sistemoje įtakos neturėjo. Fermento įtaka pastebima teršiant didesnėmis vandenilio peroksido koncentracijomis. Kai $H_2O_2$ koncentracija 2,92 g/l – energijos sunaudojimas padidėja iki 60 %. Didėjant mitybinių grandžių skaičiui, mažiau priauga biomasės ir daugiau sunaudojama energijos.
4. Nustatyta priklausomybė tarp energijos ir biomasės prieaugio veikliojo dumblo ekosistemoje: fermentinis preparatas suteikia sistemai papildomos energijos (iki 20 kcal). Eksperimento su sulfatais metu energijos akumuliuojama nuo 5 iki 40 kcal mažiau nei eksperimento su chloridais metu. Permanganatas veikliajame dumble mažina energijos sunaudojimą.
5. Atliktas veikliojo dumblo biocenozės dinamikos aerobiniame nuotekų valyme bei substrato biodegradacijos modeliavimas: veikliojo dumblo koncentracijos dinamika modeliuota priklausomai nuo chloridų ir sulfatų koncentracijos ir gautas diferencialinės lygties sprendinys, kurioje identifikuoti biodegradacijos koeficientai. Sudarytas substrato biodegradacijos modelis, kuris buvo aprašytas veikliojo dumblo koncentracijos dinamikos ir substrato biodegradacijos lygčių sistema ir išspręstas Rungės – Kutto metodu.
6. Fermentinis preparatas *Roebic–47* išbandytas Salantų nuotekų valykloje: $BDS_7$ rodikliai po valymo sumažėjo beveik 2 kartus, nuotekų išvalymas pagal ChDS rodiklį pagerėjo 2,5 karto, efektyvumas padidėjo 9,3 %. Detergentų koncentracija palyginus su baziniais duomenimis sumažėjo 2 kartus, nuotekų išvalymo efektyvumas pagal detergentų rodiklius padidėjo 23,3 %.

**Rekomendacijos**

1. Atlikti eksperimentiniai tyrimai bei gauti rezultatai parodė, kad fermentiniai preparatai, pagaminti iš mikrobinės žaliavos, gali būti sėkmingai naudojami biologinių nuotekų valyklų darbo intensyvinimui.

2. Rekomenduojama, remiantis veikliojo dumblo biocenozės dinamikos tyrimų rezultatais, naudoti fermentinius preparatus dumblo atsinaujinimui, kai į valyklas patenka didelės momentinės koncentracijos teršiančių medžiagų, turinčių inhibitorinį poveikį veikliajam dumblui.
3. Fermentinius preparatus rekomenduojama taikyti aerobinio biologinio valymo įrenginiuose, dibančiuose su veikliuoju dumblu, periodinio veikimo režimu.
4. Rekomenduojama fermentinio preparato koncentracija 1,2 ml/l valomų nuotekų; ištirpusio deguonies koncentracija rezervuare su veikliuoju dumblu rekomenduojama palaikyti 3–4 mg/l, nes prie tokios koncentracijos pasiekiama didžiausia teršalų oksidacijos savitoji sparta. Terpės pH ir temperatūra turėtų būti palaikoma optimali mikroorganizmų veiklai.
5. Praktiniam nuotekų valymo skaičiavimui rekomenduojama naudoti veikliojo dumblo dinamikos bei substrato biodegradacijos modelius, kurie buvo patikslinti eksperimentinių tyrimų rezultatais.

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**INVESTIGATION OF BIOLOGICAL WASTEWATER TREATMENT USING AN ENZYMES PREPARATIONS**

**Summary of Doctoral Dissertation**
**Technological Sciences,**
**Environmental Engineering and Landscape Management (04T)**

**Audra Skaisgirienė**

**BIOLOGINIO NUOTEKŲ VALYMO TYRIMAI NAUDOJANT FERMENTINIUS PREPARATUS**

**Daktaro disertacijos santrauka**
**Technologijos mokslai, aplinkos inžinerija ir kraštotvarka (04T)**

2006 07 03. 1,5 sp. l. Tiražas 100 egz.
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