



## WORKSHOP PROPOSAL

**M.Auezov South Kazakhstan State University (SKSU)**

**Workshop Title:** *Biotechnological approach in solution of problems of land degradation*

**Data of the workshop:** 15<sup>th</sup> of December 2017

**Location:** Conference hall of SKSU (Shymkent) 14.00

### **Aim**

Environmental Biotechnology is one of today's fastest growing and most practically useful scientific fields. Research into the genetics, biochemistry and physiology of exploitable microorganisms is rapidly being translated into commercially available technologies for reversing and preventing further deterioration of the earth's environment. In this way environmental biotechnology can make a significant contribution to sustainable development.

The following basic methods are used successfully in practice of ecobiotechnology:

- restoration of the surface layer and properties of soils;
- recycling and utilization of organic waste.
- biological purification of sewage;
- biological purification and deodorization gases;

At the moment the bank of ecologically safe beneficial types of microorganisms is created. Preparations based on them are characterized by a complex action. They have a stimulating effect on plant growth, suppress of a number of diseases, improve the mineral nutrition of plants, enhance soil fertility, and significantly reduce the pesticide load. Application of a new generation of biological products not only increases the productivity of the plants but also allows getting an earlier production, improving its safety.

### **Specific Objectives are:**

1. To adopt production processes that make optimal use of natural resources, by recycling biomass, recovering energy and minimizing waste generation.
2. To promote the use of biotechnological techniques with emphasis on bioremediation of land and water, waste treatment, soil conservation, reforestation, afforestation and land rehabilitation.
3. To apply biotechnological processes and their products to protect environmental integrity with a view to long-term ecological security.

Use of biotechnology to treat pollution problems is not a new idea. Communities have depended on complex populations of naturally occurring microbes for sewage treatment for over a century. Every living organism—animals, plants, bacteria and so forth—ingests nutrients to live and produces a waste as a by-product. Different organisms need different types of nutrients.

Certain bacteria thrive on the chemical components of waste products. Some organisms feed on materials toxic to others. Research related environmental biotechnology is vital in developing effective solutions for mitigating, preventing and reversing environmental damage with the help of these living forms.

Thus, the main goal of the workshop - is to create a forum for discussion on the theme of lesson.

### **Some additional aims are:**

- To give a general overview on the condition of land resources of South Kazakhstan region.



- To acquaint with biotechnological approach in solution of problems of land degradation.
- To present our approach to mitigation of “Land Degradation” and how it might be of use to all project partners working in Europe, Central Asia and China.

### **Participants**

Participation is open to all project partners’ researchers and professors, via a Webex platform. Researchers and professors from other universities and research organisations are welcome.

### **Program of the Workshop**

- 14-00 *Introduction of the participants* (prof. Saparbekova Almira, SKSU)
- *General overview about condition of land resources of Kazakhstan. Application of biotechnology for solving ecological problems.* (prof. Saparbekova Almira Amangeldievna, SKSU)
- 14-30 *The Influence antropogenic efforts to high plants and biotechnological methods of protection them.* (Professor Eshibaev A.A.)
- 15-00 *Lecture on theme «Research of medicinal plant raw materials for production of biological preparations»* (PhD Aimenova Zhanar, SKSU)
- 15-20 *Influence of soil microorganisms on soil structure* (prof. Mamitova Aigul, SKSU)
- 15-40 *Anaerobic digestion of agricultural waste for biogas and fertilizer production. Production of bio fertilizers on the base of soil microorganisms* (prof. Mutaliev Botagoz, SKSU)
- 16-00 *Question and debate* (prof. Saparbekova Almira, SKSU)
- 17.00 *Evaluation of the workshop*



## Abstract of the lectures

### General overview about condition of land resources of Kazakhstan. Application of biotechnology for solving ecological problems.

**Prof. Saparbekova Almira Amangeldievna, SKSU**

Scientists and experts from different countries, governments and the public understand that environmental problems cannot solve in only one country. The ecological problem is priority in the National plan of action in protection of environment in Republic of Kazakhstan and the first problem is land protection.

On the vast territory of the Republic, there are a number of regions where the combination of various forms of environmentally damaged soils has resulted in a crisis situation. Disastrous environmental conditions can be observed in the Aral Sea area: zones of intensives oil desertification, salinisation and deflation. The regions of South and East Kazakhstan, which are the major industrial centers, are the centres of technogenic disturbances and the industrial pollution of soils with toxic chemicals (lead, mercury, chromium, etc.).

Analysis of the current status of the soil landscape has shown that intensive land degradation and desertification processes can be observed, with deterioration of the soil and environmental conditions of the country taking place. According to the data of the Agency on Land Resources Management, more than 75 % of the territory of Kazakhstan is subjected to degradation and desertification; over 14% of pastures have reached an extreme degree of degradation or are completely degraded, and it is impossible to use them. Therefore, 15.2 % of lands are classified as “waste” fallow lands.

About 21 billion tons of solid waste of all types has accumulated in the country. Its incremental growth amounts to 1 billion tons a year (Annex Most solid waste is stored in Karaganda (29.4%), Eastern Kazakhstan (25.7%), Kostanai (17.0%) and Pavlodar (14.6%) oblasts. The development of the mining industry in the Republic determines the quality and condition of the land, causing pollution by such toxic substances as radioactive nuclides, heavy metals, etc

Almost one third of industrial waste in the country is accumulated in Karaganda region - more than 8.5 billion tons by the end of 2012. The main volume of emissions in Karaganda region accounts for metallurgical sphere and equals to 70%.

The major ecological problem include: increasing accumulation of municipal solid waste. According to the Ministry of Environment, the country has accumulated 23 billion tons of municipal solid waste (MSW). Annual increase of accumulated solid waste equals to 700 million tons. Only **3-5% of garbage** is being recycled. About **97% is stored** in outdoor dumps. The greatest amount of MSW is concentrated in [Almaty](#), the most populous city in the country. For the last year city dumps accumulated 470 thousand tons of solid waste.

Persistent organic pollutants are a group of chemicals - industrial substances harmful waste products of dioxin type - the compounds and mixtures of which have toxic properties, are resistant to decomposition. As a result of transboundary transfer by air and water, they settle long distances from their emission points, accumulating in water and land ecosystems.

In the oil regions of Western Kazakhstan more than 500 thousand ha, there are large sections of soil contaminated with oil and radioactive materials, high levels of salinity with industrial waste water and technological transformation of the soil landscape, leading to the accumulation of toxic heavy metals (lead, cobalt, nickel, vanadium etc.) and radionuclides (thorium, barium, radium). Lands contaminated with heavy metals and radioactive substances



occupy about 21.5 M.ha. These pollutants are spread on 59% of the area in the Atyrau region, 19% in Aktobe, 13% in West Kazakhstan and 9% in Mangystau.

In Southern Kazakhstan, in recent years, the processes of erosion on irrigated fields and pastures have developed rapidly: every year 19 million tones of soil are washed off with 400 K.tonnes of humus. Every year about 2.5–2.6 million tones of manure would be needed to cover these losses. Progressive salinisation of irrigated land is taking place in the Arys-Turkestan, Tashutkul, Bakanas and other irrigated areas.

Intensive use of agricultural lands of the country without consideration of the agro-ecological potential of the territory and scientifically justified land cultivation systems has led to a significant decrease in soil fertility. The loss of humus after the cultivation of virgin and fallow lands was one-third of the original content, or 45–48% including the most valuable humic acids and hydrolyzed nitrogen, and up to 60% in irrigated conditions. The annual losses of humus in agriculture in Kazakhstan are 1.2–1.6 t/ha.

Use of biotechnology for solving ecological problems is not a new idea. Mix bacterial populations have used for wastewater treatment for over a hundred years yet.

Specialists in the sphere of environmental protection used two methods of bioremediation (bioremediation techniques) contaminated lands by organic waste; inoculate in contaminated soil specialized strains of bacteria or nutrients that stimulate the activity of microorganisms which have already presented there. Bacteria absorb toxins and decompose into harmless products of vital activity. After the entire stock of toxic compounds are processed numbers of bacteria-cleaners comes back to normal levels, or they die.

In the field of bioremediation the most actual studies are purification soil from oil products, pesticides, heavy metals, chlororganic compounds by methods in situ. The new technology of production ready forms biological preparations for cleaning petropolluted water reservoirs and water areas, improve the efficiency of their use (immobilized cells, biofilters, fillers, etc.) have developed.

The following basic methods are used successfully in practice of ecobiotechnology:

- biological purification of sewage;
- biological purification and deodorization gases;
- restoration of the surface layer and properties of soils;
- recycling and utilization of organic waste.

At the moment the bank of ecologically safe beneficial types of microorganisms is created. Preparations based on them are characterized by a complex action. They have a stimulating effect on plant growth, suppress of a number of diseases, improve the mineral nutrition of plants, enhance soil fertility, and significantly reduce the pesticide load. Application of a new generation of biological products not only increases the productivity of the plants but also allows getting an earlier production, improving its safety.

### **Negative influence of anthropogenic factors on urban flora and biotechnological methods for solving ecological problems of dendroflora in Southern Kazakhstan**

**A.A. Yeshibayev**  
**sc. doctor of biology**

Nowadays, intensive growth of the technogenic burden on the urban ecosystem is the reason for the decline in the basic indicators of the human habitat. Dendroflora, being an important component of all urban ecosystems, plays the role of an accelerator of all biogeochemical processes in the soil and serves as a filter for purification of atmospheric air from toxic compounds.



However, the progressive growth of the burden on the urban ecosystem invariably leads to the emergence of various kinds of dendrological problems caused by abiotic and biotic factors. These negative factors in total lead to degradation of the dendroflora. In this regard, the solution of environmental problems of urban flora is an actual scientific task.

The above factors have caused the degradation of the unique community of dendroflora of southern Kazakhstan, which consists of 250 native and introduced species of trees. However, the basis of dendroflora is indigenous species that are well adapted to the arid conditions of the local climate. Since 2000, dendroflora began to mark the death of native species, especially six widespread trees. They began to be damaged by unknown pests and infectious diseases before that time.

The identification of the causes of this problem and the search for biotechnological solutions has become the goal of our research.

The research was conducted between 2012 and 2017. The communities of dendroflora of large settlements of the South Kazakhstan region and the city of Shymkent were investigated. Taxonomic analysis of the composition of the dendroflora was carried out using the illustrated determinant of the flora of Kazakhstan in 6 volumes, (1969), the life condition of the trees was assessed according to the method V.A. Kovdy, A.S. Kerzhentsev and V.A. Alekseeva. Species composition of pests was determined using the determinants of T.V. Paleeva (1994), A.S. Bogolyubov (2002), S.S. Izhevsky (2005). Species composition of phytopathogenic objects was determined using methods of incubating samples of affected parts of trees in the thermostats. Next, pure cultures of pathogens, their antagonists from natural substrates (micromycetes and bacteria) were isolated and identified by PCR analysis using the 16S rRNA gene and the ITS region of the nucleotide chain. Phytopathogenicity of isolated strains of microorganisms was checked by reinfection of parts of trees. The effectiveness of strains of antagonists against phytopathogens was evaluated according to the scheme of the pathogen-antagonist system, on an artificial nutrient medium using strains of pure cultures of microorganisms.

The results of the research establish that the cause of degradation of the dendroflora of Southern Kazakhstan are invasive pests and phytopathogenic microorganisms brought from the northern regions of Russia along with raw wood. Pests under new conditions were able to adapt well to local climate and other fodder substrate and caused the spread of phytopathogenic microorganisms of trees for southern Kazakhstan. Their joint livelihoods caused the degradation of the dendroflora of the region. Adult trees began to die from damage to phloem and rotting of the inner part of the trunk.

It has been established that the communities of six native species are most severely degraded: *Platanus orientalis* L., *Salix alba* L., *Salix babilonica* L., *Populus nigra* L., *Populus alba* L., *Ulmus pumila* L., *Ulmis macrocarpa* Hance .. The degree of pest damage and the disease burden of these tree species is 65-85%. The results of taxonomic analysis showed that of the 25 identified species of pests, the most harmful are six: xylophagous - *Monochamus urussovi* F., *Monochamus sutor* L., *Acanthocinus aedelis* L., *Cossus cossus* L., *Cetonia aureta* L., and leaf beetle - *Xanthogaleruca luteola* Muller . Of these pests, *Cossus cossus* L. belongs to the group of *lepidoptera*, and other species to the group of *Caleoptera*. The cause of putrefaction of the trunks of sick trees are bacteria from the family *Pseudomonadaceae*, the genera *Pseudomonas* and *Xanthomonas* - *Pseudomonas ulm* and *Xanthomonas ampelina*. In addition, phytopathogenic micromycetes *Fusarium solanium* and *Fusarium oxysporum* develop in damaged tree trunks, which destroy the phloem structure and deform the trunks.

To solve this problem, it was necessary to create a bacterial biological preparation. We examined 550 strains of the bacterium of the *Bacillus* family isolated from various substrates of



our region. After preliminary tests, we isolated two strains of bacteria that showed a high degree of antagonistic activity against the main phytopathogens of dendroflora.

In the following stages of our work, it was found that the isolated prospective strains possess high enzymatic activity and very low toxicity for the organisms of experimental animals. And according to the phenotypic signs and the results of molecular genetic identification are the local strains of bacteria *Bacillus thuringiensis*. The results of studies of pathogenicity and allergenicity of these strains on the organism of warm-blooded animals and humans showed that only one strain complies with the standards permitting their use as a producer-strain for biologics. This strain, called *B. thuringiensis* 4ant B-RKM 0651, was deposited in the republican collection of promising microorganisms. Subsequently, on the basis of this strain, the biopreparation YUKOBAK-DENDRO was developed. This biopreparation was tested in conditions of dendroflora in southern Kazakhstan and introduced into production. YUKOBAK-DENDRO is developed in pasty and liquid formulations.

### **Research of medicinal plant raw materials for production of biological preparations PhD Aimenova Zhanar, SKSU**

*Nowadays biotechnology is one of the key directions of high-quality technology development in a number of industries of economy. In general, potential of opportunities and a range of application of biotechnology turned industry into the leading factor of economy development of the world community. In the Republic of Kazakhstan much attention is also paid to biotechnology development.*

Despite development of biotechnology in pharmaceutical and medicine industries, biological medicines, which possessed hemostatic action are not presented at the domestic market. The prevailing majority of the known medicines with hemostatic action are synthetic, possessing a number of side effects. Thus, research of opportunities of creation of the biological medicine possessing hemostatic action on the basis of vegetable raw materials is an urgent scientific task.

On the territory of Kazakhstan more than 6 000 species of plants grow. From them 1 058 species are included in the official list of the medicinal plants containing terpenoids, alkaloids, steroids and phenol compounds 1. In flora of South Kazakhstan there are more than 3 000 species of plants from which 553 are valuable natural sources of medicinal substances. However the majority of these species of plants still now remain little-studied 2. In the native pharmacological and cosmetic industries only a limited quantity of plants species is used. In this connection, the priority direction of researches is revealing of potential sources of biologically active compounds which will provide a long-term reliable raw-material base of the pharmacological and cosmetology industries of Kazakhstan. Revealing of new perspective species of plants will allow to use rationally vegetative resources and will give the chance to expand considerably the assortment of vegetative medicinal raw materials.

Species of *Lagochilus* genus concern number of the most perspective and littlestudied plants of South Kazakhstan which are known for the medicinal property from ancient times and were widely used in folk medicine of the East. From this genus, nowadays, the chemistry only of *Lagochilus inebriantis* Bunge species is well studied.

This species has been already used for more than 50 years in medicine as styptic and hypotensive remedy, other species of genus are little researched.

*Lagochilus inebriant* species is abundant in flat territories of area and in 70s years of the last century was cultivated in cultural agriculture as a source of medicinal raw materials. Areas of distribution of other species remain not investigated. According to T. I. Tsukervanik in world flora



there are 44 species of *Lagochilus* genus. According to taxonomy of this author 34 species grow on territory of the CIS. From them 25 species are on the territory of Uzbekistan, and in Kazakhstan — 14 species 6.

Besides, extensive areas of distribution of representatives of *Lagochilus* genus are located in Iran, Afganistan, Mongolia, the Russian Altai, in a number of the countries of Caucasus and the Himalayas. By other authors it is established, that the flora of Central Asia where the natural habitat of *Lagochilus* covers Tien Shan and the Pamirs range is the richest in the specific relation. From the established number of species 20 are endemic ones. Widely used *L. inebrians* species is registered in the Red book and gathering of the raw material in natural areas of this species is completely

Flora of the South Kazakhstan is rich for herbs. A species *Lagochilus*, known as a source of vegetable raw materials for production of medicines of hemostatic action, contains several tens of types of plants from which only *Lagochilus inebrians* is well researched. Now many types from this genus unfairly remain out of interests of pharmaceutical and medicine biotechnology. According to literary data, it is known that *L.setulosus* made the practical interest for the purposes of pharmaceutical biotechnology. Except the known diterpene lagochilin, it also contains diterpene lagochirzin, which is active ingredient of hemostatic medicines.

*L.setulosus* plants, growing in the territory of the South Kazakhstan area, collected in a phase of mass blossoming and the beginning of fructification and also experimental animals which were under hemostatic activity of the biological medicine "Setulin" – white outbred mice and even-aged rabbits.

Objects of researches are natural diterpenes as a part of the phytomass of *L. setulosus* plants, methods of their extraction, influence of *L.setulosus* extract on a hemostasis of blood of warm-blooded animals, elements of technology of receipt biologic medicine product with hemostatic properties.

Research methodology – floristic route researches, scanning microscope method of macro- and microelements structure identification, high-quality and quantitative chemical analyses of *L.setulosus* biomass regarding determination of biologically active compounds, laboratory test methods of biological medicine regarding acute toxicity, influence on time of bleeding and the size of blood loss.

Scientific novelty consists in the following: for the first time there was established development of technology of receipt of biologic medicine product with hemostatic properties on the basis of extract of *L.setulosus* plants, way of extraction of the crushed phytomass of plants by the using of electrochemical processing (ECP) water (catholyte) as a result of which is established increasing of diterpenes lagochilin and lagochirzin on 50%.

Theoretical value of work is defined by the fact that object of research is perspective species of wild-growing flora of district which remain till this days not claimed for medical and pharmaceutical biotechnology. Research of biotechnological potential of wild-growing flora of southern region and perfection of existing technologies of reception of biological products from vegetative extracts represent the big scientific interest, because these results expand representation about possibility of use of new species of plants and isolation of highly effective medicinal substances from a vegetative biomass. Results, which were received in the course of performance of dissertational work bring the appreciable contribution to understanding of possibility of working out of biological preparations from the vegetative raw materials which efficiency is based on effect of influence of a complex of biologically active components of herbs phytomass. Results of research can be used for more detailed studying of biochemical structure and clinical effect of influences of some organic compounds *Lagochilus* genus plants phytomass, and also for studying of questions of successful cultural cultivation of perspective species.

Practical value consists in the specific structure of *Lagochilus* genus, growing in the South Kazakhstan area is investigated, areas of distribution and phytoresources of *Lagochilus setulosus* species are defined; the element and biochemical structure of phytomass of plants of this species is investigated, the complex of biologically active substances which together with diterpenes lagochilin and lagochirzin provide styptic properties of plants is defined; on a basis of extraction of this species plants the technology of reception of a biological preparation Setulin, which efficiency is proved by clinical tests for experimental animals is developed. For situation improvement in the market of manufacture of biological preparations on a vegetative basis it is necessary working out of the special program on research of biotechnological potential of medicinal species of grasses of wild flora of Kazakhstan including studying of structure of biologically active components of phytomass of perspective species, researches of possibility of introduction of new species in culture of flat agriculture, perfection of methods and ways of reception of biological preparations with hemostatic properties.

Also as a result of the carried out researches was established necessity of development of biological preparations on the basis of phyto-genesis, because the existing market styptic preparations is represented by synthetic preparations which have a number of side effects for an organism. Big pharmaceutical potential of local flora continues to remain not claimed. Studying of element and biochemical structure of phytomass of plants, perfection of process of diterpenes isolation in the extraction by electrochemically activated water and studying of their efficiency on indicators of a blood hemostasis of experimental animals has allowed to develop technology of reception of a biological preparation with styptic action.



*L.setulosus*

### **Influence of soil microorganisms on soil structure** **prof. Mamitova Aigul, SKSU**

Microorganisms in soil are important because they affect soil structure and fertility. Soil microorganisms can be classified as bacteria, actinomycetes, fungi, algae and protozoa. Each of these groups has characteristics that define them and their functions in soil.

Up to 10 billion bacterial cells inhabit each gram of soil in and around plant roots, a region known as the rhizosphere. The composition of the rhizobiome can change rapidly in response to changes in the surrounding environment.

*Soil microorganisms are responsible for:*

Transforming raw elements from one chemical form to another. Important nutrients in the soil are released by microbial activity are Nitrogen, Phosphorus, Sulfur, Iron and others.



Breaking down soil organic matter into a form useful to plants. This increases soil fertility by making nutrients available and raising CEC levels.

Degradation of pesticides and other chemicals found in the soil.

Suppression of pathogenic microorganisms that cause diseases. The pathogens themselves are part of this group, but are highly outnumbered by beneficial microbes.

Bacteria- more dominant group of microorganisms in the soil and equal to one half of the microbial biomass in soil. Majority are Heterotrophs. (Common soil bacteria - Arthrobacter, Bacillus, Clostridium, Micrococcus). Bacteria is the most abundant microorganisms in the soil, and serve many important purposes, including nitrogen fixation.

Biochemical processes

One of the most distinguished features of bacteria is their biochemical versatility. A bacterial genus called Pseudomonas can metabolize a wide range of chemicals and fertilizers. In contrast, another genus known as Nitrobacter can only derive its energy by turning nitrite into nitrate, which is also known as oxidation. The genus Clostridium is an example of bacterial versatility because it, unlike most species, can grow in the absence of oxygen, respiring anaerobically. Several species of Pseudomonas, such as Pseudomonas aeruginosa are able to respire both aerobically and anaerobically, using nitrate as the terminal electron acceptor.

Nitrogen fixation

Bacteria are responsible for the process of nitrogen fixation, which is the conversion of atmospheric nitrogen into nitrogen-containing compounds (such as ammonia) that can be used by plants. Autotrophic bacteria derive their energy by making their own food through oxidation, like the Nitrobacters species, rather than feeding on plants or other organisms. These bacteria are responsible for nitrogen fixation. The amount of autotrophic bacteria is small compared to heterotrophic bacteria (the opposite of autotrophic bacteria, heterotrophic bacteria acquire energy by consuming plants or other microorganisms), but are very important because almost every plant and organism requires nitrogen in some way.

**Anaerobic digestion of agricultural waste for biogas and fertilizer production.  
Production of bio fertilizers on the base of soil microorganisms  
prof. Mutaliev Botagoz, SKSU**

In the many countries the quantity of traditional type of energy are insufficient for ensuring of human by fuel.

How will human beings generate energy when all the oil resources we benefit from today are fully consumed? There is as yet no clear answer to this question. But regardless of what the answer may be, it is clear that the mankind will always want to continue building huge inventories of energy. With the declining quantity of fossil fuels it is critical today to focus on sustained economic use of existing limited resources and on identifying new technologies and renewable resources, e.g., biomass, for future energy supply.

Anaerobic fermentation of organic wastes to produce energy in the form of biogas is, arguably, the most likely option to be commercially interesting, provided that the economics were favourable.

A recent review, however, has demonstrated that the use of anaerobic fermentation for the treatment of the organic waste would reduce the emission of carbon dioxide. Therefore, in the light of the emission reductions agreed at the Kyoto Summit, environmental considerations may be of greater significance than economics.



Anaerobic fermentation converts a major part of organic nitrogen to ammonia, which is then directly available, though also potentially phytotoxic, for plants as a nitrogen source. It was found that in addition to nitrogen, the fermented residue as a fertilizer could satisfy the plants' need for phosphorus, whereas potassium needs to be added through fertilization. Consequently, this work will investigate the anaerobic digestion of agricultural waste for biogas and fertilizer production.

Implementation of the researches on the development of technologies of the obtaining of the alternative source energy – biogas by way of anaerobic fermentation of the production wastes will help to solve the problems of management of home and industrial wastes in light of the priority directions in Concepts of ecological safety of the Republic of Kazakhstan. The optimization of given process will allow to provide ecological safety and stable development of the country. The wastes from various types of production capable to give significant profit at the reasonable use. The priority of the utilization of wastes above their liquidation is obvious, that's why the investigations in the field of the repeated use of wastes are perspective and actual.

Results of analysis of literature shown that the biogas yield dependence from the contents of co-ferments in agricultural plants.

In general, all types of biomass can be used as substrates as long as they contain carbohydrates, proteins, fats, cellulose, and hemicellulose as main components.

By co - substrates added to the liquid manure the content of organic substrate is increased, hence the yield of biogas. From an economic point of view it is only profitable, however, when the materials are sourced from a location within a distance of 15 – 20 km. In general, the content of dry matter (liquid manure and co - substrates) in the substrate should be below 2 – 12% to ensure the functionality of standard pumps and a proper mixing in the bioreactor, what is important for an efficient transformation process. But the addition of co - substrates possess a higher hygienic risk. If the residue from the fermentation process is to be used as fertilizer for agricultural areas, the co - substrates should meet the national laws and should not pose any hazard from exposure, e.g., they must be free of pathogens. Some co - substrates, like residues from separated fat, leftovers, and flotated material from a fat removal tank, contain nitrogen - rich nutrients. When distributing this to fields, the upper limit for nitrogen can be exceeded, given e.g. The limit is set at 210 kg ha<sup>-1</sup>, which corresponds to the quantity of nitrogen in excreta of 3.5 GVE ha<sup>-1</sup>. In this case, the residue from fermentation may not be used on the agricultural areas from which the residue came, but must be transported to, e.g., cattle - less agricultural enterprises in the neighbourhood, what often involves additional costs.

The following table show the biogas yield at the usage the various wastes from livestock of husbandry as a substrate.

Table 1 - Biogas yield at the usage the various wastes from livestock of husbandry as a substrate

Substrate for biogas production	DM [%] oDM in DM [%]	Biogas yield [m <sup>3</sup> kg oTS] Retention time [d]
Liquid manure from cattle	6-11	0,1 – 0,8
Excreta from chicken	10 – 29 67 – 77	0.3 – 0.8
Excreta from sheep (fresh)	18 -25 80-85	0,3 – 0,4
Excreta from pigs	20 – 25 75 – 80	0.27 – 0.45



Literature data shows that technology of biogas production based on processing of agriculture waste is very well-known in the world. But despite on this fact use of technologies have some problems. One of the problem – is high cost of equipment. Usually turn of bioreactor reach up 25-30 days. This involves use of big volume of reactors that essentially influence on the cost of all device. Therefore high cost of equipment for processing of waste is important and basic deterrent factor.

We putted the objective to develop technology of accelerated methanogenesis to decrease the bioreactor turn up to 7 days.

For this it is necessary to improve construction of bioreactor for increase of methane yield. One of way of increase reactor productivity and decrease of Hydraulic Retention time is density increase, that is achieved by microorganisms immobilization. We developed new construction of bioreactor including immobilized installation.

Acceleration of methanogenesis occurs for account of methanogenic microflora fixing in apparatus on polymeric carriers, which provides more efficiency fermentation of agriculture waste. Immobilization allows to accumulate methanogenic bacteria as a biofilm on the carriers surface. This provides storage of biomass independently from Hydraulic Retention time, and immobilized microorganisms is not exposed to stress and damaging by gas bubbles.

In addition, we developed the optimal composition of nutrient medium for methanogenic bacteria. Also time of fermentation depends on pre-processing of raw material, and before fermentation raw material is processed by electrical charge (pulse method). This method allows to crush material, also pathogenic microflora is perished.

So, term of bioreactor turn is decreased, and volume of processed raw material is increased.

Therefore, usage of this bioreactor with immobilized device allows to decrease of raw material processing time, to produce biogas and high-quality biofertilizers.