

## Final Project Report

### 1. Contestant profile

▪ Contestant name:	Dmitriy Faleyev
▪ Contestant occupation:	Leading Researcher of ecological biotechnology laboratory
▪ University / Organisation	Scientific-Research Institute of Ecology
▪ E-mail:	
▪ Phone (incl. country code):	
▪ Number of people in your team:	<b>5</b>

### 2. Project overview

Title:	Studying the role of arbuscular mycorrhiza in the formation of plant communities gravelly quarry near the village Baltabay
Contest:	Quarry Life Award
Quarry name:	Baltabay
Prize category: (select all appropriate)	<input type="checkbox"/> Education and Raising Awareness <input checked="" type="checkbox"/> Habitat and Species Research <input type="checkbox"/> Biodiversity Management <input type="checkbox"/> Student Project <input type="checkbox"/> Beyond Quarry Borders

## Abstract

Research career Baltabay flora and its surroundings found in the vicinity of the quarry vegetation consists mainly of semi-desert and steppe species (drought-tolerant xerophytes) and ephemera and ephemeroïds with short vegetation period. Here we had identified 40 plants. The vegetation in the vicinity of the quarry is exposed to road degression and intensive grazing.

On the territory of the quarry Baltabay despite the ongoing work there were identified about 20 species of plants. Basically it is undemanding to soil and pioneer weeds. Plants quickly begin to develop production facilities and sometimes even prevent the production, indicating a high potential for remediation of the local flora.

Arbuscular mycorrhiza Research has shown that in the vicinity of the quarry plant mainly poorly mycotrophic: the intensity of mycorrhizal infection does not exceed 1-1.5 points (on 5-point scale Selivanov), frequent nonmycorrhizal species.

On the territory of the quarry the vast majority of the samples studied plant root systems were nonmycorrhizal. Mycorrhizal plants are extremely rare, due to the absence of soil, flora poverty, prevalence of plants that in natural conditions are nonmycorrhizal or optional mycotrophy.

The presence of mycorrhiza indicates the status of the plant community - and can be used in the bioassay as the test marker. Mycorrhizal plants more resistant to adverse environmental factors, increasing resistance ka individual plants and plant communities and ecosystems.

Experiments with artificial mycorrhization on experimental plots career Baltabay showed that the introduction of inoculum arbuscular mycorrhizal fungi significantly increases the intensity of mycorrhizal infection and incidence of mycorrhiza in the roots of the test plants. In the experiment the plants were submitted to the high and medium mycotrophic instances, no mycorrhizal plants are extremely rare.

The experiments on the effect of arbuscular mycorrhizal inoculum, suspension of green algae soil and biogumus on the growth of seedlings - *Trifolium pretense* L. (Fabaceae); - *Lolium perenne* L., *Poa pratensis* L., *Festuca rubra* L. (Poaceae) have shown:

- Both separately and jointly making inoculum endomikoriz, algal suspension and biogumus improves germination, survival and growth of plants;

- Very effective it was the introduction (both together and separately) inoculum of vermicompost and arbuscular mycorrhizal fungi;

- The introduction of a suspension of algae is so pronounced effect had not, perhaps this is due to the negative influence of the dry climate;

- The highest intensity of mycorrhizal infection found in variants of the experiment with the introduction of inoculum mycorrhizal fungi, the joint introduction fungi and algae; fungi and liquid biogumus, it is obvious that making the suspension of algae and liquid biogumus helps stimulate the growth of arbuscular mycorrhizal fungi and plants mycorrhization;

- Joint and separate application of mycorrhizal fungi inoculum, a suspension of microscopic green algae and biogumus can be recommended to speed up the restoration of vegetation and soil cover in the open pit.

Further research in this area will create the preconditions for the development of modern, profitable biotechnology reclamation and rehabilitation of disturbed lands.

## Final report

Ecological problems are multifaceted, complex and their solutions accordingly requires multifaceted, comprehensive approach. In this regard, the development of biotechnology is very promising the restoration of degraded ecosystems with arbuscular mycorrhiza.

Arbuscular mycorrhiza (endomikorrhiza) - a mutually beneficial co-existence of the roots of higher plants with microscopic fungi Division *Glomeromycota*. Arbuscular mycorrhizal fungi are widespread in nature at the meeting of more than 80% of vascular plants, from arctic wastelands to deserts, tropical and equatorial forests, from the highlands to the coastal areas. Deep penetrating into the ground, penetrating it in different directions by a dense network of branches subtle, being the link between soil and root system of plants, soil hyphae of mycorrhizal fungi continuously supply the roots and aerial parts of the plant with water and nutrients.

Mycorrhizal plants are more resistant to various adverse factors of the environment: a lack of nutrients and water, poisoning by heavy metals and other toxic substances of human origin, soil salinization, etc. The high role of arbuscular mycorrhizal fungi and as a component for events remediation and recovery, land as endomikorrhiza enhances both stability and viability mycotroph and biogeocoenose stability as a whole. Arbuscular mycorrhiza has been actively involved in the cycle of minerals: promotes biological weathering, development of soil-forming processes.

### Study mikosimbiotrofizma herbaceous plants

For the intensity of mycotic infections taken root system of plants, with a depth of 15-20cm, since it is at this depth is found most of the structures of mycorrhizal fungus. Samples taken carefully cleaned of adhering soil particles by shaking, and then washed in water. Roots collected immediately fixed in 4% formalin solution.

Samples of root systems, after removal of formalin, washed well with water and subjected to maceration: roots refluxed for 3.5 hours on a steam bath in 10% KOH solution.

Macerated specimens stained with trypan blue in lactic acid. A solution of trypan blue in lacto-glycerol is prepared according to the recipe males: Trypan blue - 0.1 g; lactic acid - 50 g; distilled water - 100 grams, gliitsirin (chemically pure) - 50 Washed g. from alkali roots macerated is placed in a solution of trypan blue for 30 minutes without heating, and then washed by water and placed for the dye color differentiation in laktoglitserin (50% solution (reagent pure) gliitsirina in distilled water). Then stained roots were washed with water.

For microscopy roots of glycerol and placed in a pressure prepare drugs and examined under a microscope at 120x magnification times. In each field of view is determined the intensity of mycorrhizal infection mycorrhizal fungus in points (on a five-point system).

To determine the number of mikosimbioticheskogo differentiation, the number of high-, medium-, low- and nemikotrofnyh copies in points (on a five-point system). By vysokomikotrofnyh include plants with mycorrhizal infection intensity of 3.5 points, srednemikotrofnyh - 2-3.5 points, slabomikotrofnyh - to 2 points.

### The study of vegetation, the selection of the root systems of herbaceous plants samples to study arbuscular mycorrhizal fungi quarry Baltabay and its surroundings

The climate of the study area is sharply continental. Summers are hot and dry. Vegetation is typical for steppes and semi-deserts. The plant associations a large proportion of the plants and ephemera ephemeroids characterized by a short period of vegetation. In connection with the above, many of the plants wither in late May - early June. In connection with this, our first trip to the quarry Baltabay was held in early May: 05. 05. 2016.

In the course of the studies were conducted: geobotanical description and photographing point sampling patterns of root systems of herbaceous plants to study mikosimbiotrofizma (arbuscular mycorrhizal fungi).

The bulk of the soil structures of arbuscular mycorrhizal fungi is located at a depth of 15-20 cm, so the root systems of plants samples were collected from this depth. Selected plant roots gently freed from the soil particles and promptly delivered to the laboratory for fixing.

To clarify the taxonomic composition of the plant has been assembled a herbarium.

### Flora neighborhoods quarry Baltabay

During the research the role of arbuscular mycorrhizal fungi in shaping vegetation quarry Baltabay and the surrounding area has been studied taxonomy of herbaceous plants.

Plant communities formed under conditions of lack of moisture, but in terms of having a close groundwater that feed a nearby river Turgen. Most of the plants represented by ephemeral and ephemeroïds - species with a short growing season, as well as xeromorphic (drought-resistant) species and mesophytes.

Ephemera and ephemeroïds presented species such as *Papaver pavoninum* Schrenk., *Kochia prostrata* (L.) Schrad., *Alyssum desertorum* Stapf., *Allium* sp., *Bromus tectorum* L., *Bromus* sp., *Poa bulbosa* L., *Eremopyrum* sp., *Lapulla* sp. and others. Xerophytes - *Artemisia terrae-albae* Krasch, *Artemisia* sp. and others. Mesophytes - *Ranunculus* sp, *Taraxacum* sp.. And others. In total during the course of this study, we detected and identified about 40 species.

### **Flora quarry Baltabay**

Ecosystems (plant communities) of dry steppes and semi-deserts is easier to break down as a result of technogenic influence and recover for a long time. However, within 3 years after the removal of topsoil (overburden), for the extraction of the quarry, there are relatively frequent emergence of the plants - the pioneers.

In our career revealed *Poligonum aviculare* L., *Poligonum* sp., *Artemisia annua* L., *Artemisia* sp., *Xanthium strumarium* L., *Convolvulus* sp., *Carex* sp., *Papaver pavoninum* Schrenk., *Bromus tectorum* L., *Bromus* sp., *Poa bulbosa* L., *Phragmites communis* Trin., *Sisymbrium altissimum* L., *Herniaria* sp. and others. Only about 20 species.

The presence of such vegetation even prevents the further conduct of work on mining in the quarry. This fact is indicative of the high recultivation potential of disturbed land at the expense of resources of the species diversity of local (native) plant communities (ecosystems), providing a high level of natural recovery technogenic disturbed areas: in this case, due to mechanical problems (destruction) and subsequent removal of soil, up to expose the bedrock.

In the course of research on arbuscular mycorrhizal grasses career Baltabay and the surrounding area we studied more than 450 samples of root systems of herbaceous plants in 12 plant communities.

Arbuscular mycorrhiza Research has shown that in the vicinity of the quarry plant mainly poorly mycotrophic: the intensity of mycorrhizal infection does not exceed 1-1.5 points (on 5-point scale Selivanov), frequent nonmycorrhizal species.

On the territory of the quarry the vast majority of the samples studied plant root systems were nonmycorrhizal. Mycorrhizal plants are extremely rare, due to the absence of soil, flora poverty, prevalence of plants that in natural conditions are nonmycorrhizal or optional mycotrophy.

The presence of mycorrhiza indicates the status of the plant community - and can be used in the bioassay as the test marker. Mycorrhizal plants more resistant to adverse environmental factors, increasing resistance ka individual plants and plant communities and ecosystems.

### **Setting a field experiment on the recultivation career with arbuscular mycorrhizal**

At the edge of the open pit quarry Baltabay, at a safe distance from the work area, we set up an experiment to study the possibilities of restoring damaged lands technologically quarry with soil microflora: inoculum of arbuscular mycorrhizal fungi, in conjunction with the introduction biogumus and the suspension of microscopic soil green algae.

The results of such studies can be the basis for the creation of highly productive ecosystems biotechnology on anthropogenic disturbed areas, with the lost vegetation and soil cover.

It was sprinkled on the soil (overburden layer) from the quarry to the experiment. The soil was removed for extraction at the quarry.

Preparations were soil mix: overburden the soil and washed coarse sand from the quarry, sawdust at a ratio of 7: 2: 1. Soil mix was applied to a layer of parent rock (gravel and sand) - layer thickness of 15-20 cm.

Variants of the experiment: 1 - the control; 2 - suspension of soil green algae genus *Chlorella*, *Scenedesmus* (ChS); 3 - biogumus (BG) (5% by volume of soil mix); 4 - inoculum of arbuscular mycorrhizal fungi (M); 5 - M + ChS; 6 - M + ChS + BG; 7 - M + BG; 8 - M + AExBG a liquid biogumus (aqueous extract); 9 - AExBG.

Seeded grass families, which in nature are mycorrhizal: legumes (Fabaceae) - *Trifolium pretense* L.; cereals (Poaceae) - *Lolium perenne* L., *Poa pratensis* L., *Festuca rubra* L.

The ultimate objective of these studies - Biotechnology Development recultivation in open pits with arbuscular mycorrhizal fungi, biogumus and soil green algae.

**Watering, making a suspension of microscopic green algae soil and extract biogumus (aqueous extract).**

Algae were cultured in plastic containers, with occasional stirring, with 12 hour lighting. Culturing was carried out using Knopp nutrient medium and adding carbon dioxide.

The density of the algae suspension -  $1,76 \times 10^6$  (*Chlorella*) and  $4,5 \times 10^6$  units/ml (*Scenedesmus*). Suspension application rate of  $2 \text{ l/m}^2$ .

Adding green algae Suspension improves germination of seeds and spores of mycorrhizal fungi, and improves their growth, increases soil fertility and crop yields, thus contributing to the restoration of vegetation and soil cover, the development of ecosystems in quarries.

Experiments with artificial mycorrhization on experimental plots career Baltabay showed that the introduction of inoculum arbuscular mycorrhizal fungi significantly increases the intensity of mycorrhizal infection and incidence of mycorrhiza in the roots of the test plants. In the experiment the plants were submitted to the high and medium mycotrophic instances, no mycorrhizal plants are extremely rare.

The experiments on the effect of arbuscular mycorrhizal inoculum, suspension of green algae soil and biogumus on the growth of seedlings - *Trifolium pretense* L. (Fabaceae); - *Lolium perenne* L., *Poa pratensis* L., *Festuca rubra* L. (Poaceae) have shown:

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- Joint and separate application of mycorrhizal fungi inoculum, a suspension of microscopic green algae and biogumus can be recommended to speed up the restoration of vegetation and soil cover in the open pit (Tab. 1-3).

Table 1 - dry weight of the aerial parts of plants in a field experiment in the open pit Baltabay

plant species	Variants of the experiment / dry weight of the aerial parts of the plants (g / m <sup>2</sup> )								
	1	2	3	4	5	6	7	8	9
	K	ChS	BG	M	M+ChS	M+ChS +BG	M+BG	M+GBG	GBG
<i>Trifolium pretense</i> L.	2,40	13,60	2,70	25,10	33,60	9,50	3,40	153,60	213,60
Poaceae: <i>Lolium perenne</i> L., <i>Poa pratensis</i> L., <i>Festuca rubra</i> L.	17,10	66,40	51,50	91,70	4,00	37,60	8,50	78,90	52,40
<i>Amarant sp.</i>	195,06	269,10	501,60	95,90	211,50	511,60	664,90	144,30	37,40
other types of herbaceous plants	17,70	18,24	10,50	14,00	0,00	30,80	0,00	25,20	32,10
<b>Total</b>	<b>232,24</b>	<b>367,34</b>	<b>566,30</b>	<b>226,70</b>	<b>249,10</b>	<b>589,50</b>	<b>676,80</b>	<b>402,00</b>	<b>335,50</b>

Table 2 - mycorrhizal status of plants in a field experiment in the open pit Baltabay

plant species	Variants of the experiment / mycorrhizal plant status								
	1	2	3	4	5	6	7	8	9
	K	ChS	BG	M	M+ChS	M+ChS +BG	M+BG	M+GBG	GBG
Trifolium pretense L.	+	++	+	+++	+++	++	++	+++	+
Poaceae: Lolium perenne L., Poa pratensis L., Festuca rubra L.	++	±	-	+++	+	++	++	++	++

- - no mycorrhizal plants,  
± - weak mycotroph or no mycorrhizal  
+ - weak mycotroph,  
++ - medium mycotroph,  
+++ - high mycotroph.

Table 3 - Dry mass of the aerial part of the plant in a field experiment in pot plants

plant species	Variants of the experiment / dry weight of the aerial parts of the plants (g / m2)								
	1	2	3	4	5	6	7	8	9
	K	ChS	BG	M	M+ChS	M+ChS +BG	M+BG	M+GBG	GBG
Trifolium pretense L.	58,212	174,048	676,2	540,96	152,88	176,4	664,44	170,52	294
Poaceae: Lolium perenne L., Poa pratensis L., Festuca rubra L.	11,76	105,84	511,56	352,8	188,16	258,72	341,04	123,48	117,6
<b>Total</b>	<b>69,97</b>	<b>279,89</b>	<b>187,76</b>	<b>893,76</b>	<b>341,04</b>	<b>435,12</b>	<b>1005,48</b>	<b>294,00</b>	<b>411,60</b>

Further research in this area will create the preconditions for the development of modern, profitable biotechnology reclamation and rehabilitation of disturbed lands.

### Background anthropogenic natural recovery of disturbed lands quarries

In extreme conditions of dry steppes and semi-desert plant communities (ecosystems) are more vulnerable to external influences of various ecological factors, including exposure to tehnogenniks. These plant communities (ecosystems) is easier to break down and are restored relatively long time - tens and hundreds of years.

Biological diversity, particularly diversity of flora, environs mining quarries - the basis for the start-up and acceleration of the processes of natural restoration of ecosystems in the territory of the quarries themselves after finishing their work.

A major role in the formation of plant communities and whole ecosystems play arbuscular mycorrhizal fungi (endomycorrhiza). Arbuscular mycorrhiza increase resistance to adverse environmental factors as the individual plants and whole plant communities.

Research arbuscular mycorrhiza herbaceous plants growing in the pit and adjacent territories - this is the first step towards the development and optimization of biotechnologies aimed at restoring degraded lands career, improve the ecological environment and the subsequent return of the land into circulation, with the prospect of the return of land for economic use; for example, their use in crop or livestock production. The presence of arbuscular mycorrhiza in plant communities contributes to the floristic and biological diversity of ecosystems quarries.

The data can be embedded in the educational process in the universities of Kazakhstan in the field of "Ecology", "Biology", "Biotechnology", and can also be used when reading the articles "Environment", "Environmental Biotechnology" and others. The research results will be used during the classes with students in the classroom mug "Environmental biotechnology". In addition, the data may be of interest to specialists and biologists ecologists working in the field of remediation and reclamation of disturbed lands.

The data obtained in the course of endomikoriz research can be used as a long-term research in order to optimize and cleaner production at the quarry. Biotechnology reclamation and rehabilitation of disturbed lands developed using arbuscular mycorrhiza will optimize costs in the environmental field, due to the conservation and restoration of violated acceleration processes during the production processes of plant and soil, which as a result leads to a reduction of costs.



**To be kept and filled in at the end of your report**

<p><b>Project tags (select all appropriate):</b></p> <p>This will be use to classify your project in the project archive (that is also available online)</p>	
<p><b>Project focus:</b></p> <ul style="list-style-type: none"> <li><input checked="" type="checkbox"/> Biodiversity management</li> <li><input type="checkbox"/> Cooperation programmes</li> <li><input type="checkbox"/> Education and Raising awareness</li> <li><input checked="" type="checkbox"/> Endangered and protected species</li> <li><input checked="" type="checkbox"/> Invasive species</li> <li><input checked="" type="checkbox"/> Landscape management - rehabilitation</li> <li><input checked="" type="checkbox"/> Rehabilitation</li> <li><input checked="" type="checkbox"/> Scientific research</li> <li><input checked="" type="checkbox"/> Soil management</li> <li><input checked="" type="checkbox"/> Urban ecology</li> <li><input type="checkbox"/> Water management</li> </ul> <p><b>Flora:</b></p> <ul style="list-style-type: none"> <li><input type="checkbox"/> Conifers and cycads</li> <li><input type="checkbox"/> Ferns</li> <li><input checked="" type="checkbox"/> Flowering plants</li> <li><input checked="" type="checkbox"/> Fungi</li> <li><input type="checkbox"/> Mosses and liverworts</li> </ul> <p><b>Fauna:</b></p> <ul style="list-style-type: none"> <li><input type="checkbox"/> Amphibians</li> <li><input type="checkbox"/> Birds</li> <li><input type="checkbox"/> Dragonflies &amp; Butterflies</li> <li><input type="checkbox"/> Fish</li> <li><input type="checkbox"/> Mammals</li> <li><input type="checkbox"/> Reptiles</li> <li><input type="checkbox"/> Spiders</li> <li><input checked="" type="checkbox"/> Other insects</li> <li><input checked="" type="checkbox"/> Other species</li> </ul>	<p><b>Habitat:</b></p> <ul style="list-style-type: none"> <li><input type="checkbox"/> Cave</li> <li><input type="checkbox"/> Cliffs</li> <li><input checked="" type="checkbox"/> Fields - crops/culture</li> <li><input type="checkbox"/> Forest</li> <li><input checked="" type="checkbox"/> Grassland</li> <li><input checked="" type="checkbox"/> Human settlement</li> <li><input type="checkbox"/> Open areas of rocky grounds</li> <li><input type="checkbox"/> Recreational areas</li> <li><input type="checkbox"/> Screes</li> <li><input type="checkbox"/> Shrubs &amp; groves</li> <li><input checked="" type="checkbox"/> Soil</li> <li><input checked="" type="checkbox"/> Wander biotopes</li> <li><input type="checkbox"/> Water bodies (flowing, standing)</li> <li><input type="checkbox"/> Wetland</li> </ul> <p><b>Stakeholders:</b></p> <ul style="list-style-type: none"> <li><input checked="" type="checkbox"/> Authorities</li> <li><input checked="" type="checkbox"/> Local community</li> <li><input checked="" type="checkbox"/> NGOs</li> <li><input checked="" type="checkbox"/> Schools</li> <li><input checked="" type="checkbox"/> Universities</li> </ul>