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Characteristics of the Soils with the *Bacillus thuringiensis* Entomopathogenic Bacteria Formed in the Belt of Wild Fruit Forests of the IlI and Zhetysu Alatau

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This paper presents the distribution areas of the Sievers apple (Malus niedzwetzkyana) in Central Asia, the history of its research and the profile structure, the composition and properties of the soils, under which they were formed, and the areas where the apple ermine moth and the *Bacillus thuringiensis* bacteria developed, the strains of these bacteria are the basis for the preparation of biological products against coleopteran pests. Formation of dark grey soils occurs in a neutral environment under the optimal hydrothermal conditions, which ensure the accumulation of significant amounts of humus. Heavy granulometric composition of the soils, consisting primarily of coarse dust (43-46%) and silt (21%), contributed to this process. In such conditions, a sufficient amount of labile forms of nutrients accumulates in the soil. Thus, a conjugate comparative morphological characteristic of the genetic horizons of the dark-grey soils profile, their composition and properties gives reason to believe that the conditions of heat and moisture supply with the increased amount of crop residues create optimal conditions for the development of microflora, including the *Bacillus thuringiensis* bacteria.

Key words: humus, entomopathogens, biopeticide, granulometry, profile, soils.

The concept of ecological security of Kazakhstan states: "The basis of an obvious imminent threat to biodiversity is a problem of forests disappearance, development of soil erosion, overuse of flora and fauna". It is exemplified by an ubiquitous sharp increase in the number of the apple ermine moth in the mountains of the Ili and Zhetysu Alatau in 2003-2005. This moth significantly damaged wild apple trees, resulting in 20-25% of trees dead or severely suppressed in growth and development. It is known that they are the world centre of modern cultural apple trees, and the Sivers tree is a genetic basis for all cultivated apple varieties in the world. In this regard, pests control, as well as anthropogenic stress control, are of great importance for the preservation of this natural phenomenon.

Recently, it has become apparent that the use of chemical pesticides leads to their accumulation in natural biocenoses and negatively affects wildlife and human health[1]. According to UNESCO, pesticides take the 8-9 place in the list of pollutants of the Earth biosphere[2]. Hence, the control over plant diseases and pests requires the development of innovative methods, which ensure compliance with the best environmental criteria of biocenoses. Biological methods of plant protection can be one of those.

In Kazakhstan, the methods of biological

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control over pests and plant diseases with the use of entomopathogenic microorganisms were used for the first time in 1962 by L.M. Mosievskaya. Biopesticides based on the *Bacillus thuringiensis* bacteria were used against arthropod pests. It is proved that plants, treated with this drug, are not affected by arthropods. The basis for the toxic action of bacteria is the protein of spores and the endotoxin crystals, which form the exotoxin and some toxic enzymes. Thus, currently, the drugs prepared from *Bacillus thuringiensis* are considered the safest and effective in the plant pests control, at the same time[3].

Bacillus thuringiensis bacteria are isolated from soil and arthropods[4, 5], as well as from other objects of the environment. Currently, 80 types of the *Bacillus thuringiensis* bacteria are known[6]. Among them, there are strains that have different effects on arthropods. On the biopesticides market, the share of drugs, composed of the spore-crystal *Bacillus thuringiensis* complexes, amounts to 90-95%. On the world level, around 20 samples of drugs are based on the *Bacillus thuringiensis* bacteria, in Russia more than ten kinds of these drugs are created[7]. During the selection of soil samples, it should be remembered that for example, thermophilic bacteria are not necessarily found only in thermal springs, and alkalophilic bacteria – in the soils with high pH values. Different strains of bacilli, capable of growing at different pH values, can be isolated from regular soil[8]. Louis Pasteur discovered the spore forming entomopathogenic bacteria during his research on silkworm diseases. However, the first reliable works on spore-crystal forming microorganisms were published in Japan. In 1901-1902, a Japanese scientist Ishiwata isolated a spore-forming bacterium from silkworms and described it as *Bacillus sotto*. Since 1908 Ivabushi had been writing about "*Sotto*" – the causative agent of paralysis in silkworms in Japan. In Europe, for the first time information about *Bacillus thuringiensis* appeared shortly after the delivery of the flour moth (*Ephestia kuehniella Zell*). So, in 1911-1915, in the province of Thuringia (Germany), a German scientist Berliner allocated a spore-forming bacterium from sick caterpillars and named it *Bacillus thuringiensis*[9]. Later, a similar strain was isolated and studied in detail by Matthes. He tried to use the bacteria against flour moth, but did not succeed[10].

In Europe, in the 50s only Hungarian researchers and the student of Goucha – Clement worked with *Bt*[11], Clement successfully used this bacterium against the Fall webworm.

In our country, except for the domestic drug "Ak kobelek", there are no biopesticides that are based on the strains of local entomopathogenic microorganisms. Although, this drug is included into the list of pesticides and toxic chemicals, due to lack of funding from the Government, it is produced in very limited quantities.

In regard with the aforementioned, the object of our research is to study the environmental conditions of the bacteria – dark grey soils that were formed in the belt of the wild fruit forests of the Ili and Zhetysu Alatau.

The distribution area of the Sievers apple tree in the south and south-east of the Republic is on the Zhetysu Alatau – 48.8%, Ili Alatau – 25.7%, Karatau – 12.1%; Talas Alatau – 11.7%; Tarbagatay – 2%. The Sievers apple was first discovered in 1793 in the valley of the Urzhar River and described by the Russian botanist Johann Sievers. The Sievers apple is preserved in its original form in in the genetic reserves of the Zhetysu and Ili Alatau[12].

Objects and methods of research.

The objects of research are dark grey forest soils of the northern slopes of the Ili and Zhetysu Alatau. The Ili Alatau is the most northern ridge of the Tian Shan mountains and stretches the length of 260 km from east to west. The highest point is 4973 m; it is situated near the city of Almaty. On its northern slope, vertical belts of natural areas are clearly seen from the desert-steppe foothills to the mountain tundra. In one of the belts, a belt of deciduous forests is formed at a height of 1300-1600 m. Here, wild apple trees grow among aspens, birches and spruces. Under these apple trees, dark grey soils are formed[13]. The wild apple tree occupies 3064 hectares in the mountains of Trans-Ili Alatau, 2400 hectares are also covered with the high-density stand[14].

The Zhetysu Alatau is also a mountainous area, which extends to 400 km from the south-west to the north-east, reaches 5066 m in the upper reaches of the Cox River, forming a mountain junction. In the north east of this mountain junction, the Zhetysu Alatau is stretched as a single mountain range. In its northern part at a sea-level altitude of 1500-2000m, in the mountain-forest zone with a moderately warm climate and a relatively high amount of precipitation (about 1000 mm), the largest amount of which falls in the warm season, the Zhetysu Alatau forms a discontinuous belt of the deciduous forests of aspen, poplar, birch, ash, wild apple trees – sweet cherry, barberry et al., under which dark grey soils are formed on clay loam.

Soil profiles were studied by making cuts in a typical section, characterizing the distribution area of dark grey soils. In the description of cuts profiles, we used a traditional comparative morphological and genetic analysis with the allocation of genetic horizons. In the selected soil samples, we determined:

1. Humus by Tyurin I.V., GOST 26312-91 (The State Standard);
2. Total nitrogen by Kjeldahl I.G., GOST 26107-84;
3. Total phosphorus and potassium according to GOST 26264-84;
4. Readily hydrolyzable nitrogen by Tyurin – Kononova;
5. Labile phosphorus by Machingin;
6. Exchange potassium – on a flame photometer according to GOST 26205-84,85;
7. Composition of the aqueous extract according to GOST 2643-85, 26428-85, 26425-85, 26426-85, 26424-85, 25428;
8 pH according to GOST 26425-85;
9. Cation exchange capacity by Bobkov – Askinazi – Aleshin, GOST 17.4.4.01.

RESULTS AND DISCUSSION

The study on the composition and properties profiles of dark grey soil of the Ili and Zhetysu Alatau revealed that they are identical. Therefore, they are characterized based on studies, carried out on the Ili Alatau.

The cut was made on the northern slope of the mountain ridge between the gorges of Medeu and Butakovka at a sea-level altitude of 1140 m. The vegetation – birch, hawthorne, wild apple, aspen, trees, wild pear, black currant. The projected coverage is 90%.

The soil profile is clearly differentiated into the humus-accumulative (A) and the transitional (B1 and B2) horizons, underlain with the BC horizon and soil-forming rocks.

Despite adequate rainfall and plant residues, the soil depth is still small (105cm) that can be explained by the slope steepness.

The following features characterize the morphogenetic soil signs of the dark grey forest soils:

A 0-15 cm 15	Dark grey, moist, slightly compacted, crumbly and granular, penetrated by many roots, a significant part of the aggregates is linked into beads, heavy loam, many burrows and coprolite of earthworms, does not effervesce from 10% HCl, clear transition by colour.
B_1 15-30 cm 15	Brownish grey, very moist, in some places there is a water film on the surface of the aggreagtes, penetrated by small roots, some large roots are found, heavy loam, does not effervesce from 10% HCl, there are fungal hyphae, gradual transition by colour.
B_2 30-80 cm 50	Greyish-brown, heavily moistened, compacted, crumbly, heavy loam, slightly penetrated by small roots of herbaceous plants, burrows and coprolite of earthworms, the aggregates surface is covered by a humus-clay glossy film, does not effervesce from 10% HCl, the transition by composition and colour.
BC80-105 cm 25	Heterogeneously coloured, greyish-brown with yellowish spots, slightly moist, heavily compacted, heavy loam, poorly penetrated by roots, porous, oval voids 1 cm in diameter filled with coprolites of earthworms, burrows and coprolites of earthworms are found, does not effervesce from 10% HCl, gradual transition by colour.
$C_1$105-120 cm 15	Light brown with spots of carbonates, dense heavy loam.

From the description of the soil profile it is clear that it has a sufficiently deep (30 cm) humus horizon (A + B_1), composed of water-stable aggregates, bonded by the root system of herbaceous plants. The B_1 horizon is characterized by the higher prevalence of brown colour and a bigger depth (50 cm), BC – by weak soil-forming processes. The latter is a wintering place for insects and earthworms. They are also found in the soil-forming rocks of the C horizon. Soil formation occured under the conditions of the soil washout regime, as evidenced by an absence of

Table 1 . Chemical composition of dark grey soils of wild fruit forests of the Ili Alatau

Depth of sampling, cm	Humus, %	Total nitrogen, %	Total phosphorous, %	Water pH	The absorption capacity mgEq per 100 g	Absorbed bases mgEq per 100 g				Labile forms mg/100 g		
						Ca^{2+}	Mg^{2+}	Na^+	K^+	Hydrolysable nitrogen	P_2O_5	K_2O
0-15	10.57	0.569	0.25	7.2	23.35	19.60/84	3.20/13.7	0.54/2.3	0.01/0.04	9.80	4.16	74.40
15-25	2.98	0.192	0.21	7.0	19.32	16.40/85	2.40/12.4	0.46/2.4	0.06/0.31	3.70	1.80	23.04
40-50	2.42	0.162	0.18	7.0							1.45	19.20

Table 2. Composition of the aqueous extract of dark grey soils of wild fruit forests in the Ili Alatau, mgEq /%

Depth of sampling, cm	Alkalinity HCO_3^-	CO_3^{2-}	Cl^-	SO_4^{2-}	Ñà$^{2+}$	Mg^{2+}	Na^+	K^+	Amount of salts, %	Solids, %
0-15	0.52	0.00	0.10	0.00	0.30	0.15	0.05	0.12	0.049	0.072
	0.032	0.000	0.004	0.000	0.006	0.002	0.001	0.005		
15-25	0.26	0.00	0.10	0.00	0.20	0.10	0.04	0.03	0.027	0.048
	0.016	0.000	0.004	0.000	0.004	0.001	0.001	0.001		
40-50	0.24	0.00	0.10	0.00	0.15	0.10	0.07	0.03	0.025	0.036
	0.015	0.000	0.004	0.000	0.003	0.001	0.002	0.001		

effervescence signs from HCl, it indicates that the profile is leached from carbonates.

One of the features of the chemical composition and physico-chemical properties of dark grey soils is a high content of humus (10.57%) in the A horizon, then the humus content sharply reduces in the other horizons (to 2.70%) (Table 1). Such a change in the content of humus in the upper horizons is due to the predominance of the fibrous root system of herbaceous plants in the upper part of the profile and large tree roots in the lower part of the profile. The nitrogen content in the humus horizon is very high (0.569%) with a sharp decline (to 0.19%) in the $\hat{A}_1$ horizon. Here there is a direct connection between the content of humus and nitrogen and it is due to the elemental composition of humic and fulvic acids. The content of carbon and nitrogen are in the ratios 56:4 and 42:3, respectively[15].

From the description of the soil profile it is clear that it has a sufficiently deep (30 cm) humus horizon (A + B_1), composed of water-stable aggregates, bonded by the root system of herbaceous plants. The $\hat{A}_1$ horizon is characterized by the higher prevalence of brown colour and a bigger depth (50 cm), BC – by weak soil-forming processes. The latter is a wintering place for insects and earthworms. They are also found in the soil-forming rocks of the C horizon. Soil formation occured under the conditions of the soil washout regime, as evidenced by an absence of effervescence signs from HCl, it indicates that the profile is leached from carbonates.

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Elevated levels of calcium magnesium are explained by their fixation in the soil profile in the form of biogenic carbonates. The fact that the amount of solids is 1.5 times greater than the amount of salts indicates that in the liquid phase there are other soluble compounds except for salts, which are apparently represented mainly by organic substances.

Soil moisture has a direct impact on the soil-forming processes through their water regime. Large amount of precipitation (850 mm), a significant part (70%) of which which falls in spring and summer, creates a high relative moisture content of air and soil, thus leading to the wash-out water regime of the soil profile. Examination of the autumn state of field moisture of the dark-grey soils profile showed extremely high moisture (33%) in the 0-25 cm layer (near the maximum field moisture capacity) with a gradual decrease by depth, reaching a minimum (13.4%) in the BC horizon; the lower limit of which determines the depth of rainfall penetration

CONCLUSIONS

1. Formation of dark grey soils occurs under grass and deciduous forests of the northern and north-western slopes of the Ili and Zhetysu Alatau under the optimum hydrothermal carbonate-free conditions with the “wash-out” water regime, which determine the fresh water soil solution.
2. Soil profiles of dark grey soil are clearly differentiated into the humus accumulative A horizon with a low depth (0-15 cm), but high humus content (10%), underlain by the $\hat{A}_1$ horizon with a low depth (15-33 cm) and moderate humus content (2.7%). The $\hat{A}_1$ horizon is followed by the $\hat{A}_2$ coprolite horizon; the latter gradually passes to the forest soil-forming rocks through the BC horizon.
3. Hydrothermal regimes of the A and $\hat{A}_1$ dark grey soils horizons are optimal for the development of the *Bacillus thuringiensis* bacteria.

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