Available online www.ijpras.com

International Journal of Pharmaceutical Research & Allied Sciences, 2017, 6(3):47-55

Research Article

ISSN : 2277-3657
CODEN(USA) : IJPRPM

Assessment of Microorganisms and Heavy Metals' Content in The Soils Of Arboretum Named After Nikolai Klyuev

Hamitowa S.M.[1,2]*, Glinushkin A.P.[2], Avdeev Y.M.[1,2], Nalyuhin A.N.[3], Beliy A.V.[1], Zavarin D.A.[1], Snetilova V.S.[1], Lebedeva M.A.[1], Danilova E.D.[1], Semykin V.A.[4], Pigorev I.Y.[4], Lichukov S.D.[5]

[1]Vologda State University, Vologda, Russia
[2]All-Russian Research Institute of Phytopathology, Moscow Region, Russia
[3]Vologda State Milk Academy, Vologda, Russia
[4]Kursk State Agricultural Academy, Kursk, Russia
[5]Belgorod State Agrarian University, Belgorod, Russia
**Corresponding Author E-mail: xamitowa. sveta @ yandex. ru*

ABSTRACT

The research concerns the dynamic results of heavy metal content investigation and microbiological characteristics of arboretum named after Nikolai Klyuev (Vitegra, Vologda Region, Russia). It defines the average proportion of the following heavy metals: copper, lead, zinc, mercury, silver, cadmium as well as microbiological indicators, i.e. the proportion of actinomycetes, bacteria and fungi in soil layers placed in three separate zones of the arboretum (conifer, broad-leaved and narrow leaved ones). The heavy metals proportion indicators and microbiological characteristics of soils and their comparative analysis are based on the laboratory research. The report also includes a literature review of soil research in Vologda region and suggests a better possible use of arboretum named after Nikolai Klyuev.

Keywords*: Soil, heavy metals, soil pollution, arboretum, urban green zones.*

INTRODUCTION

Soil is a separate natural unit in which protection is a top priority issue when it is used by industry or agriculture.

Soil as the most important part of the ecosystem is able to accumulate chemical contaminators. Heavy metals stand out of a great number of contaminators, for they can make chemical bonds with mineral and organic matter that causes the toxicity level rise but at the same time fulfils the function of a barrier for pollutants to penetrate the underground water basin.

Heavy metals are dangerous because they possess the ability for bioaccumulation, i.e. they can concentrate in the tissues of living beings and when excessive, they can be toxic. That is why the identification of heavy metal content in soils of urban areas comes out as an urgent task.

Present day society development reveals the expanding of urban system.

Urban environment is subjected to different changes because of the intensive anthropogenic influence. The soil surface of urban areas needs much attention as well as traffic influence consequences. Industrial and building sites have great impact on soil diverting its components (agrochemical and physical ones). It interferes with its important ecological function.

Microbiota, biochemical parameters of the soils, its biological activity – all are the first to change that is why they are considered by many explorers to be the most sensitive for pollution of soil layers [11-13].

Green areas play an important role for the urban population. The scientists often do not take that areas into consideration because their soils are traditionally believed not to be subjected to intensive anthropogenic influence and do not cause much of pollution and hereby are not dangerous.

Meanwhile, small recreational zones within cities are often influenced by industrial factors, as a result the vegetation and soils worsen though they play an important role of environmental recovery and fulfil recreational and sanitary functions.

The history of soil research works in Vologda Region dates back to late 19th – early 20th centuries

The staff members of Novoalexandrov Institute of Agriculture and Grass Farming investigated the soils of Belozersk, Kirillov, Ustyuzhna, Cherepovets, Velikiy Ustyug districts of Vologda region. Their reports were published in *"Soil Research Monitoring"*, where they give a brief soil characteristics of the areas, natural historical and economic conditions of their agriculture.

M.F. Kolokololov made the first descriptions of natural historical and economic conditions of agricultural areas, brief characteristics of soils in 1909-1912. In 20-s and early 30-s, the soil research of the North Eastern part of Russian plain were conducted by A.A. Krasyuk who did the zoning. Nowadays, much of these reports have a high historical value.

In 1932-1941, the scientists of the Leningrad department of the All-Union Institute of Fertilization and soil research made their first exploration of the soil layers in Vologda region. Most districts got "soil maps" and "soil descriptions" that included the soil formation characteristics as well as the carbonate and underground water's effect. The exploration ended in soil map by N.L. Blagovidov.

Soil zoning in Vologda region was made in proportion of 1:500 000. A brief description of southern soils can be found in articles by A.G. Trutnev (1936-1939) that are purely descriptive having little to do with analytics. During the same period, A.M. Archangelskiy did the parent rock and soil research.

In 1954-1955, the Soviet Academy of Science (V.V. Docuchayev Central Museum) staff when exploring the soils of the middle part of the region made stationary observations of the seasonal changeability of the organic matter and microbiological changeability of soils and then published the articles of A.A. Baranovskaya (1957), A.A. Zavalishin and V.P. Firsova (1960).

O.V. Butuzova (1957) was the first to systematize the Vologda region soil data in the publication "The Nature of Vologda region" [22]. The author considers the main soil formation factors (climate, vegetation, parent rock, human activity) and shows the morphological descriptions of basic soil types.

The soil research of Vologa, Sokol, Chopsara districts contributed with structural and chemical composition data. The reports of L.L. Dvornikova characterize the humidification peculiarities and their conditions [23]. L.L. Dvornikova and A.P. Petrov (1970) in their report highlight the diversity of soil according to their structural composition and the humidification peculiarities of the territory. The authors describe the morphological characteristics of the basic types of non-agricultural and agricultural soils, and the humidification conditions and some of their agricultural features [33].

The report by K.A. Gavrilov describes the soils formed on the rock's different origin and brings in the data of structural and chemical composition of soils in different districts of the region. This is one of the research works that helps to analyze some of the soil types formed on the rock of various origins which contain the data of structural and chemical composition of soils in different districts of the region and that helps to analyze some soil types fully [24].

From 1953 to 1971, Vologda land management department expedition led a big research work of soils on all agricultural sites of the region. This resulted in soil maps and agrochemical measurement data and the recommendation for better use and greater fertility of these lands. Vologda State Pedagogical University, Physical Geography department staff explored the soils in the southern part of the Vologda region midlands. T.K. Tolokonnikova, V.V. Komissarov, N.P. Antipov, N.D. Avdoshenko, K.I. Usoltseva, A.A. Lyapkina, I.N. Shaizhina, L.B. Galkina were among the research leaders and N.D. Avdoshenko [31], V.V. Komissarov, A.A. Lyapkina [30], T.K. Tolokonnikova [26], K.I. Usoltseva and L.N. Weinberg [28, 29] wrote articles based on those research works.

Starting from 1964, regional agrochemical laboratory (now State Federal Agrochemical Service Department "Vologodskiy") has been leading a systematical observation of the agrochemical features of arable soils conducting the regular measuring of acidity, humus proportion and active forms of phosphorus and potassium, hydrological acidity, the amount of absorbed chemical bases but most of these data are not structurally systematized.

Having a lack of microelements concentration data, State Federal Agrochemical Service Department "Vologodskiy" did the research of active forms of serum, copper, zinc, manganese and boron amount of all agricultural lands of the region.

At the same time, they investigated the amount of such dangerous contaminators/ pollutants as heavy metals. From 1992 to 1997, they conducted a complete monitoring of average proportion of copper, lead, nickel, cobalt, zinc, manganese, cadmium, chrome, arsenic of all agricultural lands on 736,2 thousand hectares of total area.

State Federal Agrochemical Service Department "Vologodskiy" and Federal State Regional Plant Protection Station did the identification research of chlorine organic pesticides and herbicides in arable areas. To estimate the radioactive level, they measured such radioactive elements as strontium-90, cesium-137, potassium-40, thorium-232, radium-226 in soils and plants on the sample areas of Gryazovets, Cherepovets, Ustyuzhna, Belozersk, Kirillov, Sokol, Vologda,Totma, Kaduy districts.

Recently the interest for soil layer as the main factor of regional ecosystem formation has risen greatly. Scientists study the soil influence on the buffer capacity of waters for acidification and the role the soils play in agricultural use, accumulation and distribution of heavy metals and their introduction into a hydrographical net [18].

MATERIALS AND METHODS

In 2014, in 130th anniversary of Nickolay Kluyev (a local poet), Vologda government and Vitegra administration created a arboretum named after Nickolai Kluyev, where they planted trees, flowers, bushes mostly depicted in the poets rhymes, having been completed with plant compositions and small architectural constructions in Kluyev's poetry style.

Creating such an arboretum is a unique experience that allows memorizing one of the most talented Russian poets, to combine cultural, historical and ecological elements. Such method develops the mind by creating deep appreciation of nature.

The total area of the arboretum is 1 hectare and it is divided into 4 sectors: narrow-leaved, broad-leaved, a conifer one and paradise garden, each one planted with a peculiar set of tree species.

The main objective of this research is the soil estimation at arboretum from the point of view of heavy metal proportion and microbiological constituents.

The subject matter of the research is the arboretum soils (samples of 2014-2015).

The samples for heavy metals amounts and microbiological composition were taken in May 14-15, 2015.

The analyses were made using the common method of microbiological analyses of soils.

We measured soil fungi and bacteria by reproduction and insemination method by placing the soil suspension into dense nutrient medium.

For the soil fungi amount analyses, we used such medium as agar of Czapek.

The medium composition (g per 1 L of distilled water):

- Sucrose – 20 g;
- $NaN0_3$ – 3 g;
- K_2HPO_4 – 1 g;
- $MgS0_4$ – 0.5 g;

- KCl – 0.5 g;
- $FeS0_4$ – 0.1 g;
- Agar – 15 g.

For actinomycetes amount analyses, we used starch-ammonium medium.

The medium composition (g per 1 L of distilled water):

- Soluble starch – 10 g;
- $(NH_4)_2SO_4$ – 2 g;
- KH_2PO_4 – 1 g;
- $MgSO_4$ – 1 g;
- NaCl – 1 g;
- $CaCO_3$ – 3 g;
- Agar – 20 g.

For the total bacteria amount analyses, we used peptone medium.

The medium composition (g per 1 L of distilled water):

- Peptone – 5 g;
- Yeasts extract – 0,2 g;
- Agar – 20 g.

The prepared media were sterilized in autoclave under the atmospheric pressure for 1 hour.

The preparing of the soil suspension

A soil sample (10 g) was put into a retort with distilled sterilized water (90gr in each) and shaken for 10 min. Then it was given an hour to stabilize (so that more solid soil part goes down) for 1 min, 1 ml of suspension from each next retort used for the following solution. The insemination in the Petri dishes was made in proportion of 10^{-2}.

The suspensions in the Petri dishes were filled with the corresponding nutrient medium (cooled down to 45^0C) and were carefully agitated. After agar was fixed, the Petri dishes were put into thermostat and incubated with the temperature of 27^0C before the check time.

The measuring of microorganisms was done visually after 4 days passed. After the colonies amount calculation, they were grouped according to cultural signs.

The microflora analyses were based on the starch-ammonium agar, the colonies calculated after 7-10 days.

We measured the amount of colonies. All colonies calculated in a Petri dish, they were grouped according to their coloring and the consistency of aerial mycelium.

The total amount of fungi in agar of Czapek

The amount of colonies was calculated after 2-3 days. After 7 days, we defined their generic composition.

The sampling of soils for heavy metals presence was made in accordance with the requirements of GOST (Federal Standards of Certification). The size of the sample area was 10/10 meters. The soil was taken by the "envelope" method: the soil samples were bulked onto a plastic fill and thoroughly mixed up; the ground soil was evened in the form of an envelope and was divided into 4 parts. The two opposite parts were thrown away, the two rest mixed up.

After the procedure, it was evened, divided into 6 parts and from the center of each, we took a sample and put it into a linen bag weighing about 1 kg.

RESULTS AND DISCUSSION

Our research of the heavy metals can be found in Tables 1 and 2. The amount of the explored heavy metals within arboretum does not exceed maximum marginal concentration and variable marginal concentration [42, 43] in the upper soil layers.

In dynamics, the heavy metal proportion in soils is not subjected to any changes, the data being stable say that the soil layer within arboretum is not prone to any harmful anthropogenic changes which can cause the penetration of heavy metals into soli layers to increase the pollution.

We revealed dependence of the content of copper on depth of the soil (Figure 1). The direct curvilinear interrelation is reflected in drawing: with increase in depth of the soil, the content of copper increases.

The microbiological research data can be observed in Tables 3 and 4.

Soil samples taken from different parts of the forest have different amount of microorganisms, the dynamic changes being absent.

The amount of actinomycetes in the "narrow leaved forest" (p1) is lower than in the soil of the "broad leaved forest" (p2) and "conifer forest" (p3).

The bacilli amount in "narrow leaved forest" (p1) is higher than in the soil of the "broad leaved forest" (p2) and "conifer forest" (p3).

As for the amount of fungi, one can notice the higher amount of Fusarium spp. in the soil of "narrow leaved forest" (p1).

In the soil of the "broad leaved forest" (p2), there is more Trichoderma sp. than in other samples, where there were no such fungi defined by this method.

Penicillium and Aspergillus predominate in the soils of the "broad leaved forest" (p2) and "conifer forest" (p3) unlike in the soil of "narrow leaved forest" (p1).

CONCLUSION

As far as there is no information of industrial pollution with heavy metals and microbiological research within arboretum, our new data broaden the knowledge of an average level of pollution of urban soils a help to make a database of complex ecological scope of the environment.

The results of our research can be used in working out the recommendations for restoring and saving of urban environments; biomonitoring and biodiognosing the soil conditions of urban areas; measuring the effect of different factors on the environment; agricultural planning; taking different ecological and industrial measures; and study process.

REFERENCES

1. Shoba S.A., 2009. Soil science horizons: results and perspectives. Soil science, 5:515-520.
2. Alloway B.J., 1990. Heavy metals in soils. London: John Wiley & Sons, 339 p.
3. Horckmans L. et al, 2005. Local background concentrations of trace elements in soils: a case study in the Grand Duchy of Luxembourg. Cat ena., 59: 279-304.
4. Bech J. et al, 2005. Baseline concentrations of trace elements in surface soils of the Torreles and Sant Climent municipal districts (Catalonia, Spain). Environmental Monitoring and Assessment, 108: 309-322.

5. Ghambashidze G. et al, 2006. Heavy metals in soils. Annals of Agrarian Science, 4: 7–11.
6. Baize D. et al, 2001. Of the necessity of knowledge of the natural pedogeochemical background content in the evaluation of the contamination of soils by trace elements. Science of the Total Environment, 264: 127–139.
7. Lado L. et al, 2008. Heavy metals in European soils: a geostatistical analysis of the FOREGS Geochemical database. Geoderma, 148: 189-199.
8. Urushadze T. et al, 2007. Soil contamination with heavy metals in Imereti region (Georgia). Bulletin of the Georgian National Academy of Sciences, 175: 122-130.
9. Siegel F.R., 2001. Environmental geochemistry of potentially toxic metals. Berlin: Springer, 218 p.
10. Sierra M. et al, 2007. Baselines for trace elements and evaluation of environmental risk in soils of Almería (SE Spain). Geoderma, 139: 209-219.
11. Helzer Y.G. et al, 1990. Bioactiivity data in soil research. Soil science, 9: 47-60.
12. Rubenchik L.I., 1972. Microorganisms as biological indicators. Kiev: Naukova Dumka, 165 p.
13. Skvotsova I.N. et al, 1990. Microbial communities’ composition changes as one of the ecological monitoring indicators. Moscow, 84 p.
14. Gabbasova I.M. et al, 2006. Agroecological estimation of urban parks. Orenburg University Science letters, 10:362-367.
15. Fomina N.V., 2012. Microbiological characteristics of soils of Krasnoyarsk recreational areas. Conteporary agroscience issues, 9-11.
16. Dabahov M.V. et al, 2007. Heavy metals in urban parks soil. Urbanized areas ecology, 3: 41-46.
17. Vorobey G.A. (ed), 2007. Nature of the Vologda region. Vologda: Vologda publishing house, 440 p.
18. Vodyanitski Y.N., 2005. Heavy metal study in soils. Moscow, 110 p.
19. Parahonski E.V., 2001. Stable development principles of the region during recession period. Vologda, 431 p.
20. Davidov L.K., 1970. Natural zoning of Vologda region for agricultural Purposes. Vologda, 286 p.
21. Krasyuk A.A., 1923. The description of soils and waters along the Vologda- Archangels railways line from the station of Vandiga to Vologda. Agriculture and transport, 9(12): 113-166.
22. Krasyuk A.A., 1922. Vologda region, Garkopol soils of 1921. North Krai, 3:3-20.
23. Commissarov V.V., 1987. Soil Vologda region, rational use and protection: a tutorial. Vologda: 76 p.
24. Zavalishin A.A., 1973. Research genesis gray forest and podzolic soils: selected works. Leningrad: 300 p.
25. Zavalishin A.A., 1939. Soils of the forest zone, their formation and properties. Leningrad: Publishing House of the USSR Academy of Sciences, 111 p.
26. Antipov N.P. et al, 1957. The nature of the Vologda region: a collection of articles. Vologda: Regional Publishing House, 328 p.
27. Dvornikova L., 1964. Soil cover the western part of the Vologda region. Leningrad, 17 p.
28. Pestryakov W.K. (ed) et al, 1973. Soil fertility and crop. Leningrad, 255 p.
29. Plavinsky V.A., 2007. The soils of the Vologda region, their classification and properties. Vologda, 91 p.
30. Tolokonnikova T.K. (ed), 1977. Natural conditions and resources of the north of the European part of the USSR: a collection of articles. Vologda: Vologda Pedagogical Institute, 143 p.
31. Frolov E.D. (ed), 1981. Philosophical Dictionary. Moscow: Politizdat, 445 p.

32. Usoltseva K.I., 1962. Soil cover of the southwestern part of the collective farm. Scientific notes VSPI, 27: 79-90.

33. Usoltseva K.I. et al, 1966. Bog soils, their economic value and use. Nature, agriculture and culture of the Vologda region, pp. 139-148.

34. Lyapkin A.A., 1966. Prisuhonskoy lowland floodplain soils. Nature, agriculture and culture of the Vologda region, pp. 123-138.

35. Avdoshenko N.D., 1956. The soil cover of the collective farm "Russia". Scientific notes LGPI, 116: 159-217.

36. Baranovsky A.V. et al, 1957. The results of observations of the seasonal variability of soil Vologda Region. Collection of works of the Central Museum of Soil Science, 2: 194-227.

37. Dvornikova L.L. et al, 1970. Soil Vologda Region. Natural zoning of the Vologda region for agriculture, pp. 115-168.

38. Tepper E.Z. (ed) et al, 1979. Workshop on microbiology. Moscow: Kolos, 216 p.

39. State Standard 17.4.3.01-83. Protection of Nature. Soils. General requirements for sampling.

40. State Standard 17.4.4.02-84. Protection of Nature. Soils. Methods of sampling and sample preparation for chemical, bacteriological, helminthological analysis.

41. State Standard 28168-89. Soils. Sample selection.

42. Hygienic standards 2.1.7.2511-09. Roughly allowable concentrations of chemicals in the soil.

43. Hygienic standards 2.1.7.2041-06. Maximum allowable concentrations of chemicals in the soil.

Table 1 - Heavy metal composition in the arboretum soils, 2014

Heavy metals composition data, mg/kg					
Cu	Hg	Ag	Cd	Pb	Zn
Conifer forest					
13,4 ±0,31	0,023±0,005	1,5±0,01	0,5±0,002	16,2±0,53	51,5±1,70
Broad leaved forest					
14,6 ±0,17	0,03±0,017	1,34±0,02	0,6±0,003	16,4±0,78	52,5±1,39
Narrow leaved forest					
15,9 ±3,1	0,02±0,006	1,4±0,02	0,5±0,04	15,4±0,88	56,7±1,86

Table 2 - Heavy metal composition in the arboretum soils, 2015

Heavy metals composition data, mg/kg					
Cu	Hg	Ag	Cd	Pb	Zn
Conifer forest					
15,3 ±0,4	0,03±0,004	1,9±0,01	0,8±0,002	17,0±0,53	52,5±1,8

Broad leaved forest					
14,7 ±0,15	0,03±0,012	1,44±0,1	0,6±0,004	16,3±0,8	52,1±1,5
Narrow leaved forest					
16,8 ±3,4	0,02±0,04	1,5±0,04	0,5±0,03	15,6±0,7	56,3±1,9

Table 3 - The amount of actinomycetes, bacteria and fungi dominating in the soil layers of 3 sectors (conifer, broad-leaved and narrow leaved), 2014

Microorganisms	Colony type	Microorganisms amount(CFU/g of soil)		
		p_1 (narrow leaved forest soil)	p_2 (broad leaved forest soil)	p_3 (conifer forest soil)
Actinomycetes	-	$0,3x10^3$	$0,67x10^3$	$0,67x10^3$
Bacteria	Bacillus mycoides	$0,4x10^3$	$0,14\ x10^3$	$0,24\ x10^3$
	Bacillus spp.	$0,61\ x10^3$	$0,3\ x10^3$	$0,35\ x10^3$
Fungi	Penicillium	$0,53\ x10^3$	$0,46\ x10^4$	$0,3\ x10^4$
	Fusarium spp.	$0,32\ x10^3$	$0,03\ x10^3$	$0,06\ x10^3$
	Aspergillus	$0,23\ x10^3$	0	$0,25\ x10^3$
	Trichoderma sp.	0	$0,2\ x10^3$	0
	Mycellia sterylia	0	$0,11\ x10^3$	$0,21\ x10^3$

Table 4 - The amount of actinomycetes, bacteria and fungi dominating in the soil layers of 3 sectors (conifer, broad-leaved and narrow leaved), 2015

Microorganisms	Colony type	Microorganisms amount(CFU/gr of soil)		
		p_1 (narrow leaved forest soil))	p_2 (broad leaved forest soil)	p_3 (conifer forest soil)

Actinomycetes	-	0,4x10^3	0,76x10^3	0,68x10^3
Bacteria	Bacillus mycoides	0,3x10^3	0,16 x10^3	0,26 x10^3
	Bacillus spp.	0,6 x10^3	0,3 x10^3	0,32 x10^3
Fungi	Penicillium	0,5 x10^3	0,49 x10^4	0,33 x10^4
	Fusarium spp.	0,3 x10^3	0,02 x10^3	0,06 x10^3
	Aspergillus	0,14 x10^3	0	0,24 x10^3
	Trichoderma sp.	0	0,2 x10^3	0
	Mycellia sterylia	0	0,1 x10^3	0,2 x10^3

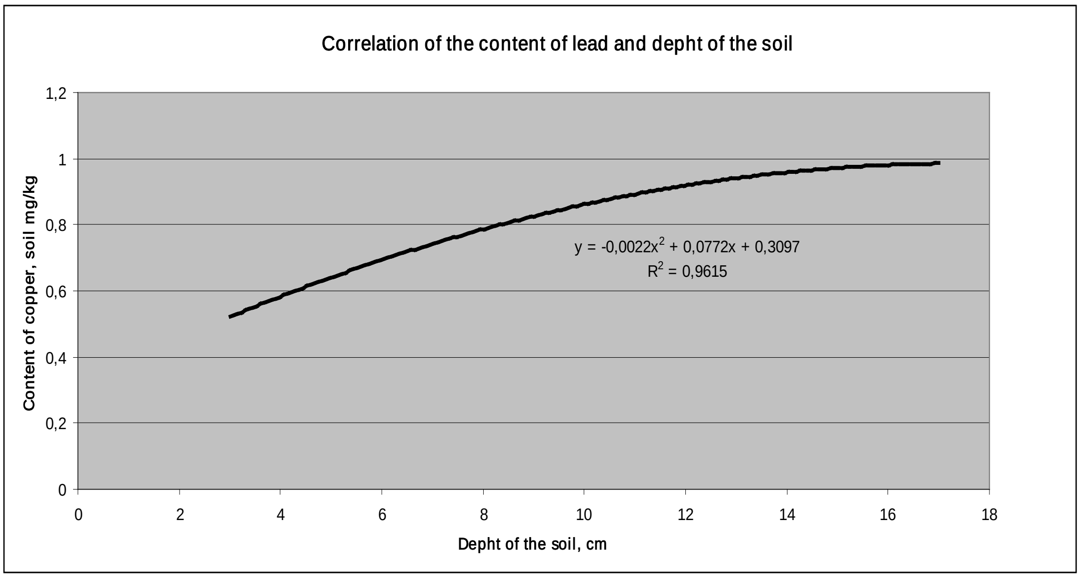

Figure 1 - Correlation of the content of lead and depth of the soil