

Krzysztof FRĄCZEK<sup>1\*</sup>, Jacek GRZYB<sup>1</sup>  
and Dariusz ROPEK<sup>2</sup>

## MICROBIOLOGICAL PROPERTIES OF SOIL IN THE AREA OF THE MUNICIPAL WASTE DUMP FOR THE KRAKOW AGGLOMERATION

### WŁAŚCIWOŚCI MIKROBIOLOGICZNE GLEBY W STREFIE ODDZIAŁYWANIA SKŁADOWISKA ODPADÓW KOMUNALNYCH AGLOMERACJI KRAKOWSKIEJ

**Abstract:** The main aim of the research was the evaluation of the municipal waste dump impact on the quantity of bacteria and fungi in the soil environment in different distances from the collected waste. The field analyses were carried out for 12 months, from June to May, during 2 years. Analytical data show that 1 g of the soil's dry mass, collected 1230 meters from the municipal waste dump Barycz contains: 27 630 to 4 223 860 cfu of vegetative forms of bacteria and 4580 to 1 806 110 cfu of the bacterial spores. Ratio of bacterial spores and vegetative cells was between 1.3 and 98.6 %. *Microscopic fungi – Micromycetes* (yeasts and filamentous fungi) occurred in quantities between 1180 and 229 630 cfu/g of the soil's dry mass.

**Keywords:** soil, microorganisms, municipal waste dump

Among the natural environment, soil is the specific element, because of different types of collected pollutants, including microbiological ones, originating from the municipal waste dump. Moreover, there are many small, illegal objects in the area of the municipal waste dump, which very often increase the soil pollution [1–4]. Therefore, in most cases the waste dumps became the potential sources of long-term, various pollutions of the ground. Huge diversity of the waste dumps' pollutants evidences the fact that they are very dangerous source of the soil contamination. These potential pollutants are: organic compounds including petrochemicals, inorganic compounds including heavy metals and large amounts of microorganisms (including pathogens) [5–9].

<sup>1</sup> Department of Microbiology, University of Agriculture in Krakow, al. A. Mickiewicza 24/28, 30–059 Kraków, Poland.

<sup>2</sup> Department of Agricultural Environment Protection, University of Agriculture in Krakow, al. A. Mickiewicza 21, 31–120 Kraków, Poland, phone: +48 12 662 41 81.

\* Corresponding author: rfracze@cyf-kr.edu.pl

Having in mind the above mentioned issues, the aim of the research was evaluation of the influence of the municipal waste dump on the quantity of bacteria and fungi in the soil environment in different distances from the collected waste.

## Material and methods

The soil's microbiological analyses were carried out nearby the municipal waste dump Barycz in Krakow. The object is placed about 3,5 kilometers from Wieliczka in the area of the foredeep which originated in the exploited aquifers of the Bogucice sands, in the valley of the small Malinowka stream. The waste dump lies on the Southwestern-Northeastern axis and its coordinates are: 250 m AMSL to the North-East direction and 270 m AMSL to the South-West direction. The municipal waste dump Barycz in Krakow is the biggest and the longest exploited object in the Lesser Poland Voivodeship and one of the biggest in Poland. It started running in December 1974, its total area is 37 ha. It is estimated that in one year the Barycz waste dump collects about 175 thousand tones of the municipal waste. Alkaline and acidic brown soils are predominant in this area. These are mainly light and average loess, sometimes heavy and loam-loess soils.

For the analyses' purposes the soil samples were taken from June until May, monthly, from 10 analytical points within and in some distance from the municipal waste dump Barycz in Krakow. In order to do this, before starting the analyses, the area was evaluated and the representative analytical points were set for sampling. The analytical points were located in the following places:

POINT NO.:	POINT DESCRIPTION
1	In front of the III section, nearby the fence, surrounding the waste dump, to the South-West direction.
2	Nearby the III section, at its middle part level, to the South-West direction.
3	By the entrance, to the North direction (the entrance is between the II and III section)
4	Inside the II section – the active zone, until February.
5	Nearby the Malinowka stream, by the unused collector for the soaking water, next to the incoming road, to the North-East direction.
6	By the piezometer P-3, to the North-East direction.
7	By the Malinowka stream, on the fallow land, covered with grass and scrub, to the North direction from the borders of I section of the waste dump.
8	By the piezometer P-6, nearby the buildings, to the North direction from the borders of the II section of the waste dump.
9	By the piezometer P-8, by the Malinowka stream in the reed bed, to the North from the borders of the II section of the waste dump.
10	By the piezometer G, 1230 m to the North from the borders of the II section of the waste dump.

It needs to be stressed that the 8 to 10 analytical points were outside of the protective area – 500 meters from the waste dump's borders. The samples were transported to the laboratory of Department of Microbiology, University of Agriculture in Krakow, where the humidity and pH tests were carried out, together with the microbiological analyses, using the serial dilution method. The following media were used for the purpose of the quantitative analyses: nutritious agar for the vegetative cells and spores of bacteria and Malt Extract Agar (MEA) for microscopic fungi. After the incubation period, colony forming units (cfu) were counted and then calculated for 1 gram of the soil's dry mass.

## Results and discussion

Apart from the naturally existing microorganisms, the foreign microbes can be found in the soil environment, which migrate from the municipal objects. Microbiological contamination of soils nearby the municipal waste dumps is mostly caused by the bioaerosols and microorganisms' spreading by the wild animals, rodents, flies and other insects. Soils may also become polluted during the delivery and unloading waste, incorrect waste dump exploitation and incorrect water export from the object [2, 10–13].

Analytical data presented in the Table 1 shows that in 1 g of the soil dry mass within and nearby the municipal waste dump Barycz in Krakow (up to 1230 m North from the borders of the section II) from 27 630 to 4 223 860 cfu of vegetative forms and from 4580 to 1 806 110 cfu of the bacterial spores can be found. The percent ratio of the spores to the general quantity of bacteria – vegetative forms, was from 1.3 to 98.6 %. Maximum amount of the bacterial vegetative cells was found in June by the entrance to the waste dump and similar amount was found in the same period inside the II section. These places were profusely inhabited by these microorganisms in different periods of the soil sampling. The lowest amount of the vegetative forms was found in January above the III section – about 570 m from the gate to the waste dump (analytical point No. 1). The highest amount of the bacterial spores was found in December inside the II section of the waste dump (analytical point No. 4) and in June in the sample from the 5 analytical point – by the unused soaking water collector and by the P-3 piezometer, point no. 6 (Table 2). Low amounts of the general spores' number were observed in June and January in the soil taken from the point no. 7, located between piezometers P-3 and P-6 (about 300m from the borders of the I section) and in November and December in the point no. 10 (1230 m from the borders of the II section) nearby the G piezometer. It needs to be noted that a large amount of the bacterial spores (over 90 %) were found in relation to the vegetative cells nearby the waste dump's entrance and around the old soaking water collector and the incoming road (Krzemieńska Street) in the different time periods. Difficult environmental conditions for the development of bacteria were found in the area of the municipal waste dump, therefore very often spore formation was observed. It may be the result of the fact that microorganisms change their metabolism and the microbiocenotic composition as the

response for numerous stress and stimulating factors, which can occur in the soil environment [2, 14].

The Table 3 presents the results from the whole analytical period about the average number of bacteria in the soil samples from the analytical points. They show that the average quantity of the vegetative cells of bacteria was from 344 550 to 961 657 cfu/g of the soil dry mass. The highest number was found in the soil samples taken by the waste dump's entrance – about 10 m from the gateway and slightly lower number was found inside the III section. In the mentioned points, the average number of vegetative bacteria cfu was over 800 000 (by the III section), and even over 900 000 (entrance, section II) in 1 gram of the soil dry mass. On the basis of the gained results it was ascertained that the highest average number of the bacterial spores (314 983 cfu/g of the soil's dry mass) and their highest percentage (62.6 % in comparison to the vegetative cells) were found in the soil by the old soaking water collector (point No. 5). The average number of spores in this analytical point was over 2 times higher than in the point No. 1. In the other tested soil samples the average quantity was from 89 594 to 241 393 cfu/g of the soil dry mass, which counted for 19.9 to 41.1 % in relation to the vegetative forms of the bacteria isolated from the tested field. The quantitative analyses of the bacteria occurrence in 10 analytical points showed that their quantity was various, however it was still higher in the soil samples by the waste dump's entrance, in the II active section and near the middle of the III section (it was being built during the research) and by the P-3 piezometer nearby the old, unused soaking water collector. The carried out research revealed that the occurrence of the vegetative forms of bacteria as well as their spores is strongly influenced by the location of the analytical point, time of sampling, direction of the wind, soil reaction and the tested soils' humidity. It is usual that the bacterial contamination and suspensions are being transported to relatively short distances from the waste dumps. The range of their infiltration depends on their survival rates in the aquifer layer and the distance of the ground water flow. It is assumed that they can reach from several to 100 meters from the waste dump. While investigating the seasonal changes, the maximum quantity of vegetative forms of bacteria was found in most points in August and minimal quantity was found in the period from November until February (Table 1). However, large numbers of bacterial spores were found in June, November and in January, which was related to high or low air temperatures (Table 2).

Soil biomass consists mostly of fungi, which are very common in this environment. In the neutral conditions, with high concentration of the hydrogen ions, especially with high content of organic matter, fungi are much more profuse than bacteria. Thanks to their individual biochemical properties, *eg* Ability to produce slime, water accumulation, production of organic acids and releasing from the soil minerals such nutrients as: potassium, phosphorus *etc* their role in pedogenesis and plant nutrition is very important [1, 15, 16].

On the basis of the mycological research, it was ascertained that the microscopic fungi – *Micromycetes* (yeasts and filamentous fungi) occurred in quantities from 1180 to 229 630 cfu/g of the tested soil dry mass (Table 4). The lowest number was noted in September in the soil sample taken from the bank of the Malinowka stream in the reed

Table 1  
 Relationship between the bacterial vegetative forms [cfu in 1 g of the soil d.m.] and the sampling period and the sampling point in the area of the municipal waste dump Barycz in Krakow

Point	Sampling dates											
	June	July	August	September	October	November	December	January	February	March	April	May
1	212 540	525 240	630 150	667 600	125 640	480 400	180 680	27 630	390 130	84 690	422 210	766 850
2	2 082 840	2 222 710	2 105 550	57 670	309 290	352 850	224 500	449 730	96 760	252 910	37 100	1 494 340
3	4 223 860	48 430	424 000	1 607 700	518 970	1 141 740	311 780	915 040	250 560	478 000	903 000	716 800
4	4 114 350	264 710	298 440	2 872 760	306 320	527 310	489 000	451 350	678 870	569 850	236 870	136 460
5	2 337 610	100 400	262 020	458 070	489 410	133 340	390 120	584 000	294 000	509 600	234 000	243 500
6	2 300 280	198 260	1 008 150	266 840	404 710	260 330	318 410	242 260	202 660	367 890	153 230	727 500
7	149 800	802 000	706 340	571 330	508 640	216 430	74 530	91 730	123 600	421 700	743 200	198 470
8	436 930	520 170	1 925 680	944 880	433 850	45 230	65 830	537 640	390 860	746 700	654 300	412 400
9	843 880	561 450	363 860	110 240	278 650	251 720	118 570	226 370	309 700	164 300	413 560	492 300
10	515 640	401 660	420 450	483 950	1 360 770	44 730	38 320	1 273 300	891 300	352 000	397 830	431 000

Table 2

Relationship between the bacterial spores [cfu in 1 g of the soil d.m.] and the sampling period and the sampling point in the area of the municipal waste dump Barycz in Krakow

Point	Sappling dates											
	June	July	August	September	October	November	December	January	February	March	April	May
1	106 270	292 000	52 050	371 000	46 650	239 190	68 900	22 600	85 300	69 810	66 380	164 440
2	539 150	64 600	90 140	38 840	232 850	315 210	64 800	7 480	86 500	175 790	29 730	278 340
3	130 490	47 260	399 500	220 040	35 410	684 150	124 300	112 460	164 000	196 000	81 500	97 000
4	118 190	99 120	264 250	1 179 170	171 150	231 130	198 000	84 960	236 000	134 700	103 580	76 460
5	1 806 110	90 590	167 610	407 840	91 070	125 200	85 200	336 830	246 870	152 840	135 900	133 730
6	1 352 190	116 220	195 060	205 740	5 080	83 500	114 320	72 920	174 320	83 400	93 400	196 350
7	4 580	173 440	41 330	371 180	13 130	56 380	43 700	6 620	45 280	81 360	153 800	84 330
8	85 090	127 090	509 380	289 200	348 980	30 150	43 220	423 940	87 200	153 200	111 400	63 200
9	831 940	375 670	16 840	60 950	16 910	11 930	22 350	31 870	96 500	84 200	65 980	84 600
10	226 450	42 180	405 060	160 170	468 080	4 710	7 400	1 038 160	59 600	37 000	131 430	189 000

Table 3

Average number of microorganisms [cfu in 1 g of the soil d.m.] in the area of the municipal waste dump Barycz in Krakow

Tested microorganisms	Sampling points									
	1	2	3	4	5	6	7	8	9	10
Bacterial vegetative forms	376 147	807 188	961 657	912 190	503 006	537 543	383 980	592 873	344 550	550 913
Bacterial spores	132 050	160 286	191 010	241 393	314 983	224 375	89 594	189 340	141 650	230 770
Fungi	33 613	75 623	35 844	26 380	60 915	39 618	17 495	33 350	27 173	28 430

bed in the wetland (point no. 9) and the highest number was found in the same period in the unused soaking water collector (point No. 5). The amount of microscopic fungi was found to be influenced by the area and period of sampling as well as by the soil reaction and the environment's humidity. The often noticed acidic reaction and high soil humidity created very good conditions for the growth and development of *Micro-mycetes*. Comparison of the average amount of fungi, presented in the Table 3 showed the significant domination of the microscopic fungi (75 623 cfu/g of the soil dry mass) in the soil nearby the III section of the waste dump, which was being built at that time and nearby the old soaking water collector (60 915 cfu/g of the soil dry mass). The increased air movement is known to promote the transport and falling of the fungi spores. According to Kulig [17] the dangerous objects cause negative effects to the environment mostly during their exploitation. During their building and after closing them, the municipal objects become smaller threat to the environment. In the other places, average number of the tested microorganisms was on the level from 17 495 cfu to 39 618 cfu/g of the soil's dry mass. The highest average was found in the analytical point no. 6 (by the P-3 piezometer about 70 m from the unused soaking water collector) and the lowest was found in the point No. 7 (above the P-3 piezometer, 300 meters from the borders of the II section – the oldest one and long time ago recultivated). According to Drzał et al [11] and Kulig [17] the microbiological contamination of the soils is observed nearby different waste dumps in various distances and with different intensity.

The Pearson correlation coefficient and its statistical significance was calculated to evaluate the influence of pH and humidity on the quantity of the tested soil microorganisms (Table 5). In most cases this correlation was negative and not significant for the soil samples from the municipal waste dump Barycz in Krakow. Only in the analytical point No. 10, the number of vegetative forms of bacteria was highly positively correlated with the soil humidity. In the other points no significant correlation was found between the soil humidity and the number of the tested microorganisms. On the other hand, the occurrence of the vegetative cells of bacteria and fungi was negatively correlated with the soil's pH in the points no. 3, 5 and 10.

The presented research as well as the results gained by the other authors confirm the differentiation of the microorganisms' occurrence in the soil in the area of the municipal objects as well as various environment contamination in that area [14, 18–20].

## Conclusions

1. On the basis of the research it may be ascertained that the municipal waste dump Barycz in Krakow causes negative effects on the soil environment, especially during the exploitation, but also during the building period.
2. After comparing all the results, it may be concluded that the waste dump does not have negative influence on the general bacteriological and mycological state of the environment only in the distance longer than 300 meters North from the borders of the waste dump's sections.

Table 4

Relationship between the fungi [cfu in 1 g of the soil d.m.] and the sampling period and the sampling point in the area of the municipal waste dump Barycz in Krakow

Point	Sampling dates											
	June	July	August	September	October	November	December	January	February	March	April	May
1	38 190	49 850	58 700	69 560	9 950	12 050	12 800	10 900	16 850	39 300	37 200	48 000
2	101 810	111 740	24 000	32 370	106 110	96 600	42 600	47 100	68 350	31 600	98 300	146 890
3	54 050	60 680	10 000	75 120	25 540	31 460	34 300	19 800	27 700	12 680	24 200	54 600
4	260	8 210	28 000	62 260	56 800	70 030	2 400	40	1 600	12 800	34 500	39 680
5	92 350	4 620	72 400	229 630	29 740	22 600	34 800	23 800	36 400	56 680	72 700	55 260
6	63 270	32 320	26 500	23 580	16 510	98 000	54 300	13 400	20 260	31 260	43 720	52 300
7	16 770	27 940	29 000	26 980	9 070	1 190	3 680	5 000	18 460	36 870	15 330	19 650
8	27 760	18 330	31 400	83 200	41 550	11 800	35 400	9 800	12 300	23 000	67 300	38 400
9	9 250	8 900	6 500	1 180	18 680	57 860	36 450	26 410	39 780	40 670	67 800	12 600
10	47 800	10 260	9 400	71 860	59 000	14 120	12 430	18 380	22 100	17 300	18 340	40 200



Table 5  
Correlation between the number of microorganisms and the soil humidity and pH

Number of microorganisms	Sampling points									
	1	2	3	4	5	6	7	8	9	10
	Humidity									
Bacterial vegetative forms	-0.35	-0.46	0.35	0.56	-0.01	-0.19	-0.39	0.06	-0.44	0.57
Bacterial spores	-0.22	-0.14	0.11	-0.01	-0.05	-0.37	-0.32	0.40	-0.34	0.79*
Fungi	-0.57	-0.06	-0.42	-0.51	-0.19	-0.43	-0.47	-0.40	0.16	-0.15
	pH									
Bacterial vegetative forms	-0.22	0.48	-0.65*	0.30	-0.17	-0.03	0.02	0.14	0.18	-0.17
Bacterial spores	-0.16	0.09	-0.16	0.32	-0.15	-0.06	-0.39	0.16	0.06	-0.15
Fungi	-0.13	-1.12	-0.32	-0.57	-0.73*	0.21	-0.05	-0.45	0.12	-0.62*

3. The gained results indicate the need to carry out the long-term analyses of the microbiocenotic content of the soils nearby the waste dumps. Only this can improve the knowledge about the dynamics and the range of the biological contamination and it can create the effective environment protection strategy.

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**WŁAŚCIWOŚCI MIKROBIOLOGICZNE GLEBY W STREFIE ODDZIAŁYWANIA  
SKŁADOWISKA ODPADÓW KOMUNALNYCH AGLOMERACJI KRAKOWSKIEJ**<sup>1</sup> Katedra Mikrobiologii<sup>2</sup> Katedra Ochrony Środowiska Rolniczego

Uniwersytet Rolniczy im. Hugona Kołłątaja w Krakowie

**Abstrakt:** Głównym celem przeprowadzonych badań była ocena wpływu składowiska odpadów komunalnych na kształtowanie się liczebności bakterii i grzybów w środowisku glebowym w różnej odległości od składowanych odpadów. Badania terenowe związane z tematem pracy prowadzono przez 12 kolejnych miesięcy, od czerwca do maja w okresie 2 lat. Z danych analitycznych wynika, że w 1 g suchej masy gleby na terenie i w odległości do 1230 m od granic składowiska odpadów komunalnych Barycz w Krakowie występuje; od 27 630 do 4 223 860 jtk/g suchej masy gleby form wegetatywnych i od 4580 do 1 806 110 jtk form przetrwalnych bakterii. Procentowy stosunek przetrwalników do liczby bakterii form wegetatywnych wynosił od 1,3 do 98,6 %. Natomiast grzyby mikroskopowe – *Micromycetes* (drożdże i grzyby strzępkowe) występowały w ilościach od 1180 do 229 630 jtk/g s.m. badanych gleb.

**Słowa kluczowe:** gleba, mikroorganizmy, składowisko odpadów komunalnych

