Advances in Science and Technology
Research Journal

Advances in Science and Technology Research Journal 2024, 18(1), 360–369
https://doi.org/10.12913/22998624/182983
ISSN 2299-8624, License CC-BY 4.0

Received: 2023.12.22
Accepted: 2024.01.11
Published: 2024.01.24

Evaluation of the Efficacy of Effective Microorganisms in the Reclamation of Degraded Soils

Grażyna Żukowska[1], Magdalena Myszura-Dymek[1*], Zofia Durczyńska[2*], Marta Bik-Małodzińska[1], Szymon Roszkowski[3]

[1] Faculty of Agrobioengineering, Institute of Soil Science, Environment Engineering and Management, University of Life Sciences in Lublin, ul. St. Leszczynskiego 7, 20-069 Lublin, Poland

[2] Collegium Medicum, Nicolaus Copernicus University, Jagiellonska 13/15, 85-067 Bydgoszcz, Poland

[3] Division of Biochemistry and Biogerontology, Collegium Medicum, Nicolaus Copernicus University, Jagiellonska 13/15, 85-067 Bydgoszcz, Poland

* Corresponding author's e-mail: magdalena.myszura-dymek@up.lublin.pl, zofia.durczynska@gmail.com

ABSTRACT

The aim of the research was to determine the reclamation effectiveness of the use of EM against the background of mineral wool and wool used together with sewage sludge or mineral fertilization with nitrogen, phosphorus, potassium (NPK) on soils degraded as a result of sulfur extraction. The effectiveness of reclamation was assessed on the basis of changes in the total organic carbon (TOC) content and the properties of humic compounds. The obtained results showed that the use of effective microorganisms in relation to mineral wool and wool used in combination with sewage sludge or NPK causes significant changes in the content and quality of soil organic matter. The observed decrease in the content of TOC, humins and an increase in the share of carbon of labile fractions and free and loosely bound with silicate-free (R_2O_3) and calcium (Ca) humus forms indicate, that in the soils in which effective microorganisms were used, mineralization processes dominated in the transformation of soil organic matter. The humification process resulted in more durable humic compounds with a higher content of humic acids and humic acids to fulvic acids ratio (C-HA:C-FA). Supplementing commonly used materials in soil reclamation with the introduction of effective microorganisms is a promising technology. Further research is necessary to determine the composition of these biofertilisers and the optimal doses at which they should be used.

Keywords: degraded soils, reclamation, effective microorganisms, C labile fractions, fractional composition of organic matter.

INTRODUCTION

The satisfaction of society's needs has led to the emergence of a peculiar paradox. Mankind, while striving to satisfy these needs, is in conflict with the natural environment, which leads to multifactorial processes of its transformation [1]. The cumulative processes of the degrading human impact on the environment translate into the emergence of global threats such as depletion of natural resources, pollution of all elements of the environment, and global warming [2], which create problems in ensuring access to clean water or sufficient quantities of good quality food [3]. Particular attention is focused on the soil system, as soils provide a range of services that are essential to the functioning of ecosystems and the UN Sustainable Development Goals [4-8]. Numerous studies and international organisations point to the need to protect soils and, when they are degraded, to take reclamation action [9].

Soil reclamation is a particular challenge in areas of multifactorial degradation and on devastated soils [10-14]. In order for soil reclamation in devastated areas to be effective, it is necessary not only to introduce a vegetation cover but also to apply treatments that initiate and intensify soil-forming processes [15]. A common scenario in the reclamation of degraded land is the use of waste with fertilising properties and mineral fertilisers [16-18].

The significant economic and environmental costs associated with the production and introduction of artificial fertilisers into the environment have increased interest in the use of biofertilizers [19, 20]. One concept for the production of biofertilizers is the use of microorganisms as an active agent [21-23], as numerous bacterial and fungal species promote plant growth while protecting plants from physical, chemical, and biological stresses.

One of the technologies used to improve soil properties is the use of effective microorganisms (EM) [23]. EM consists of numerous microorganisms, including photosynthetic bacteria (*Rhodopseudomonas palustrus, Rhodobacter spae*), lactic bacteria (*Lactobacillus casei, Streptococus lactis*), yeasts (*Saccharomyces albus, Candida utilis*), actinomycetes (*Streptomyces albus, S. griseus*) and moulds (*Aspergillus oryzae, Mucor hiemalis*) [24-26].

There are reports that the introduction of EM into the soil has a variety of positive effects, i.e., reducing decay processes, accelerating metabolism, increasing humus content, enhancing photosynthesis, detoxifying pesticide-contaminated soil, inhibiting plant pathogens, and increasing the biological quality of plant yields [27]. The EM formulation has been widely used in Japan for years and is gaining more and more interest with each passing year in Western Europe, the USA, and Brazil. Due to the positive effects observed as a result of its use, attempts are also being made to apply it in Poland [28-30].

According to a study by Tyburski and Łachacz [31], the EM preparation showed a favourable effect on available P content, earthworm colonisation, soil structure parameters, and maize yield. Different results were obtained by Jakubs et al. [32], indicating that with regard to most physicochemical properties, the preparation of the EM formula did not show significant effects of the EM preparation.

Studies show that the application of EM preparations accelerates the mineralisation of soil organic matter [33]. Clear reductions in soil organic carbon content have been shown in studies by Dziamba et al. [34] and Tołoczko et al. [35].

Given the wide range of predicted positive results associated with the application of EM, it seems highly justified to study the possibility of using EM in the reclamation of degraded soils. According to Badura [36], effective microorganisms perform their task on degraded, disturbed, and poor soils that lack microorganisms with protective functions. The EM preparation is intended to biologically activate the soil, being a microbial "vaccine".

In assessing the effects of reclamation, particular attention is paid to the transformation of soil organic matter [37, 38]. In the soil, organic matter with the participation of a numerous and diverse group of microorganisms, undergoes transformations and their rate depends on climatic conditions, soil structure, and the degree of environmental pollution. In rehabilitated soils, the soil environment is different from that of natural soils, so the transformation of humus compounds may take place in a different way, and the humus compounds formed may be characterised by different quality parameters [39]. There is no data in the literature on how the addition of EM affects the direction of the transformation of humus compounds.

Therefore, research was carried out to determine the effectiveness of the use of EM against the background of Grodan mineral wool and mineral wool used together with sewage sludge or NPK in the reclamation of areas degraded as a result of sulfur extraction. The effectiveness of reclamation was assessed on the basis of changes in the organic carbon content and the properties of soil organic matter.

STUDY MATERIAL AND METHODS

The study was conducted on soils degraded as a result of sulphur extraction by underground smelting. The field experiment was carried out in two stages. The properties of the soil and materials used in the experiment and the first stage of reclamation including technical reclamation measures such as the dismantling of technical infrastructure and landscaping (levelling the surface) were described in detail in earlier works [40]. Subsequently, 100 $Mg \cdot ha^{-1}$ of lime tailings was introduced into the soil to neutralise the reaction. Sewage sludge at a rate of 100 Mg $d.m. \cdot ha^{-1}$, mineral wool at rates of 200, 400, and 800 $m^3 \cdot ha^{-1}$, and NPK (nitrogen, phosphorus, potassium) fertilisation were applied as fertilisers to aid reclamation. The experimental scheme is shown in Table 1 and Figure 1.

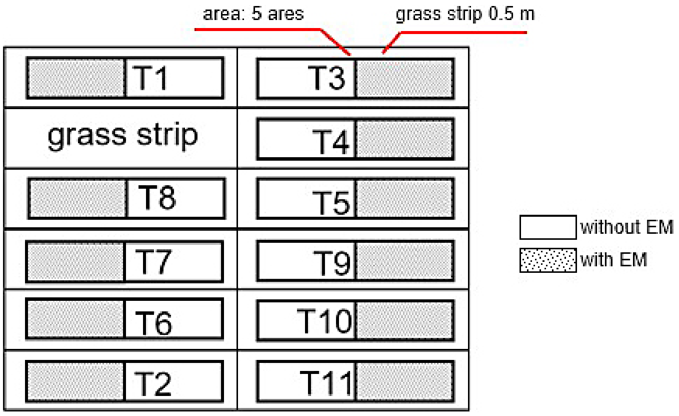

Fig. 1. Plot experiment scheme – stage 2

Table 1. Experimental scheme

Symbol	Fertilization variant
T1	Control I – soil + NPK
T2	Control II – soil +100 Mg d.m.·ha^{-1} sewage sludge
T3	Soil + mineral wool 200 m^3·ha^{-1}
T4	Soil + mineral wool 400 m^3·ha^{-1}
T5	Soil + mineral wool 800 m^3·ha^{-1}
T6	Soil + sewage sludge + mineral wool 200 m^3·ha^{-1}
T7	Soil + sewage sludge + mineral wool 400 m^3·ha^{-1}
T8	Soil + sewage sludge + mineral wool 800 m^3·ha^{-1}
T9	Soil + mineral wool 200 m^3·ha^{-1} + NPK
T10	Soil + mineral wool 400 m^3·ha^{-1} + NPK
T11	Soil + mineral wool 800 m^3·ha^{-1} + NPK

The test plants were a mixture of grasses (*Festuca pratensis, Festuca rubra, Lolium perenne, Lolium multiflorum, Dactylis glomerata, Trifolium pratense*). After 4 years of the experiment, a stage 2 experiment was started. For the stage 2 trials, each plot was divided into two parts. In one half of each plot, the experimental scheme was continued, while in the other half, EM spraying was applied (Figure 1). The spray used was a solution containing one part Emfarma Plus EM solution and nine parts water (ratio 1: 10). A rate of 20 litres of undiluted EM mixture was applied per ha. Soil samples for testing were taken for 3 consecutive years at the end of the growing season.

Laboratory analyses

Total organic carbon (TOC) was determined in dry soil samples by combustion using a TOC-VCSH [41] with an SSM-5000A module. The modified Kjeldahl method [42] with a Kjeltech TM 8100 distillation was used to determine the total nitrogen (TN) content. Labile fractions of organic matter (CL) was determined using the modified Andrzejewski and Myśków method [43]. CL was extracted from soil with 0.05 mol $NaOH \cdot dm^{-3}$ (1:10w/v) for 1 hour. Fractional composition of humus in the soil samples was determined using the modified method of Kononova-Bielchikova [44]. During extraction, the following were determined in the individual fractions: carbon content of the solutions after decalcification ($C_{deca.}$) – extraction with 0.05 mol $H_2SO_4 \cdot dm^{-3}$ (1:10 w/v); free and loosely bound with silicate-free (R_2O_3) and calcium (Ca) humus forms – extraction (24 h) of 0.1 mol $Na_4P_2O_7$ + 0.1 mol $NaOH \cdot dm^{-3}$ (1:10w/v) ($C_{extracted}$); carbon content of humic acids (C–KH) – after precipitation with 6 mol $H_2SO_4 \cdot dm^{-3}$ from an extract of 0.1 mol $Na_4P_2O_7 \cdot dm^{-3}$ + 0.1 mol $NaOH \cdot dm^{-3}$, followed by dissolution with a solution of 0.1 mol $NaOH \cdot dm^{-3}$; carbon content of fulvic acids (C–KF) was calculated from the difference: C–KF = $C_{extracted}$ – C–KH. The proportion of non-extractable carbon – humins (C–Humins) was calculated from the difference: C–Humins = TOC – ($C_{extracted}$ + $C_{deca.}$). The carbon content of the labile fractions and the fractional composition were expressed in $g \cdot kg^{-1}$d.m. of the soil sample and as the percentage of each fraction in the TOC pool (%TOC).

The results obtained were statistically processed by analysis of variance using the Tukey test. Statistical calculations were performed using the StatSoft STATISTICA 10 programme.

RESULTS AND DISCUSSION

Total organic carbon and total nitrogen content

The organic carbon (humus) content of soils is one of the criteria for determining the degree of their degradation, and it becomes fundamental when the other properties fall within the ranges for average soil conditions or are more favourable than them [45]. At the same time, soil organic carbon content is an indicator of soil quality and fertility [46] as it positively influences the overall soil properties and thus determines the ability of soils to perform ecosystem functions [47].

The organic carbon (TOC) content of the soil on which the experiment was conducted varied greatly and was 2.1 g·kg^{-1} in the 0–20 cm layer and 1.14 g·kg^{-1} in the 20–40 cm layer [40]. In the soil of the control object (T1), the TOC content in the 5th year after reclamation was 12.4 g·kg^{-1} (Table 2). Assessing the effectiveness of reclamation based on organic carbon content, it can be concluded that the neutralisation of the reaction, introduction of grasses, and application of supplementary mineral fertilisation was an effective method of reclamation. The results obtained may indicate that the improvement of the reaction of the reclaimed soil (after the application of liming) reduced the rate of mineralisation of organic matter. Furthermore, these results confirm that the application of mineral fertilisation at the optimum level, under slightly acidic and near-neutral reaction conditions, allows the humus content of the soil to be maintained or increased [48, 49].

The TOC content of the reclaimed soil was very favourably affected by the sewage sludge. In the soil of the reclamation variant (T2), the TOC content was significantly higher compared to the soil of the T1 object and amounted to 14.4 g·kg^{-1}. The favourable effect of the sewage sludge on the TOC content is related to the introduction of significant amounts of exogenous organic matter into the soil [50, 51]. The fertilisation of the soil with mineral wool only, especially at rates of 400 and 800 m^{3}·ha^{-1}, also had a beneficial effect on the organic matter balance. In the soil of these objects, the TOC content was higher compared to the soil with the addition of sewage sludge. The beneficial effect of wool on the TOC content should be linked to the improvement of the physical properties of the soil, which has a significant effect on the direction of the transformation of soil organic matter [52], and to the indirect effect of wool on the yield of plants whose residues are a source of organic carbon in the soil [53]. The results showed a very favourable effect (statistically confirmed) of Grodan mineral wool applied together with sewage sludge on the increase in TOC content. In the soil of these objects, the TOC content ranged from 21.8 g·kg^{-1} (wool at 200 m^{3}·ha^{-1}) to 13.1 g·kg^{-1} (wool at 400 m^{3}·ha^{-1}).

The application of effective micro-organisms, against the background of the reclamation variants evaluated, had a significant effect on the transformation of organic matter in the soil, as manifested by changes in TOC content (Table 2). In the soil

Table 2. Content of TOC, NT and C of labile fractions (CL)

Fertilization variant	TOC		NT		CL			
	g·kg^{-1} d.m				g·kg^{-1} d.m		% TOC	
	0	EM	0	EM	0	EM	0	EM
Control I – soil + NPK	10.5	96	1.20	1.07	3.8	3.3	47.22	52.31
Control II – soil + sewage sludge	14.4	13.0	1.47	1.24	3.9	3.9	27.00	34.17
Soil + mineral wool 200 m^{3}·ha^{-1}	11.4	11.7	2.29	1.35	4.8	5.2	38.82	43.20
Soil + mineral wool 400 m^{3}·ha^{-1}	15.3	10.6	1.27	0.91	4.4	4.9	39.15	45.33
Soil + mineral wool 800 m^{3}·ha^{-1}	15.8	9.7	1.49	1.08	5.8	6.8	35.92	41.16
Soil + sewage sludge + mineral wool 200 m^{3}·ha^{-1}	21.8	14.2	2.29	1.35	4.3	6.7	21.95	39.15
Soil + sewage sludge + mineral wool 400 m^{3}·ha^{-1}	13.1	9.9	1.27	0.91	3.6	5.7	20.27	38.06
Soil + sewage sludge + mineral wool 800 m^{3}·ha^{-1}	14.2	11.3	1.49	1.08	3.0	4.9	20.32	33.44
Soil + mineral wool 200 m^{3}·ha^{-1} + NPK	14.9	12.7	1.57	1.21	4.8	4.8	40.00	43.04
Soil + mineral wool 400 m^{3}·ha^{-1} + NPK	14.0	11.6	1.39	1.11	5.3	5.0	39.64	45.65
Soil + mineral wool 800 m^{3}·ha^{-1} + NPK	15.3	13.9	1.54	1.33	5.0	4.7	39.25	45.58
Average	14.61	11.65	1.57	1.15	4.43	5.08	33.59	41.92
NIR between: - fertilization variant - series (without EM and with EM)	0.4** 0.4**		0.05** 0.05**		2.99 0.70		14.45** 3.39**	

Note: NPK – mineral fertilization N (nitrogen), P (phosphorus), K (potassium); TOC – total organic carbon; TN – total nitrogen; CL – carbon of labile fractions.

enriched with the EM mixture, a significant reduction in TOC content was found in most sites. In the soil of the EM – enriched control object, the TOC content was 9.6 g·kg^{-1} and was about 1 g·kg^{-1} lower compared to the soil without their addition. A statistically non-significant higher TOC content in the EM – enriched soil was only found in the reclaimed soils with mineral wool at a rate of 200 m^3·ha^{-1}. The results obtained did not confirm the thesis known from the literature about the beneficial effect of EM inoculation on organic carbon content [54-56]. Studies by Schenck zu Schweinsdberg-Mickan and Müller [57] showed no effect of EM on TOC content.

In soil remediated with waste, a significant effect of the applied remediation treatments on total nitrogen (NT) content was found. In the soil of the control object fertilised with NPK, the NT content was 1.20 g·kg^{-1} (Table 2). Fertilisation with sewage sludge at a rate of 100 Mg·ha^{-1} significantly increased NT content (to 1.47 g·kg^{-1}). In all fertilisation variants with mineral wool , a significant increase in total nitrogen content was found. In soil fertilised with mineral wool (applied without additives), the NT content ranged from 1.25 g·kg^{-1} (at a wool dose of 200 m^3·ha^{-1}) to 1.75 g·kg^{-1} (at a wool dose of 400 m^3·ha^{-1}).

The effect of the conditioner (EM mixture) was revealed by a reduction in total nitrogen content in the reclaimed soils (Table 2). The observed reduction in NT content under the influence of EM was due to the utilisation of nitrogen by the higher demand of soil microorganisms [58].

Labile organic matter fractions

The reclamation technologies used should be effective and their effects sustainable [59]. With regard to soil organic matter, the determination of organic carbon content is insufficient [60]. Reclaimed soils have different environmental conditions than natural soils, hence the transformation of humus compounds may be disturbed and newly formed humus compounds may have different properties [37].

Labile humic substances are a diverse group of humus compounds, and their common characteristic is that they exist in the free state or are weakly associated with the mineral fraction of the soil. Labile humic substances are a product of the early stages of humification, the so-called 'young' form of humus, or are products of degradation processes within stable humus forms [62].

The content of labile organic matter fractions (CL) in the control soil of the experiment was 42.7% TOC (Table 2), and this content was typical for light mineral soils [43]. In the soil fertilised with sewage sludge, the organic matter fractions in question accounted for 27.0% TOC. These results are confirmed by earlier studies on the effect of sewage sludge fertilisation on CL content in light soil [63]. The proportion of CL in soil reclaimed with mineral wool and mineral wool + NPK was similar to their proportion in the control soil. In soil fertilised with wool at a rate of 200 m^3·ha^{-1} against sludge, CL accounted for 21.95% TOC and this was significantly lower compared to the NPK control. Increasing the dose of wool applied together with the sludge had no effect on changes in the proportion of the fraction described.

The effective microorganisms applied against the evaluated reclamation variants increased the share of labile fractions of organic matter (Table 2). The highest share of labile C fractions, in soil with EM addition, was found in soil fertilised with mineral wool (on average about 43% TOC) and mineral wool applied together with NPK (on average about 44% TOC). The increase in the proportion of CL in the soil of these sites is a result of the acceleration of soil organic matter mineralisation processes under the influence of EM [64]. In the soil of the wool + sewage sludge variants, the proportion of C in labile fractions also increased under the influence of EM (by 16% on average). In this case, this can be considered a favourable effect because, in soils treated with sewage sludge, the proportion of labile carbon fractions is generally low, and an increase in the C share of these fractions indicates that the organic matter introduced with the sludge becomes involved in the soil carbon cycle [59].

Fractional composition of organic matter

The content and properties of soil humus compounds are shaped by typological processes and are characteristic of individual soil types. The properties of soil humus are mainly determined by humic acids [65, 66]. The results of studies on the influence of anthropogenic factors on the fractional composition of humus clearly indicate that the proportion and properties of humus compounds are modified by fertilisation [39, 53, 67].

The proportion of low – molecular – weight humus compounds attributed to fulvic acid affinity ($C_{deca.}$) in soils rehabilitated with mineral wool and sewage sludge varied only slightly (Table 3) and gave values from about 5.3% TOC to 6.8% TOC. The effective microorganisms significantly reduced the percentage share of described humus compounds.

In the soil of the control object free and loosely bound with R_2O_3 and Ca humus forms, i.e., $C_{extracted}$, accounted for an average of 52.70% TOC (Table 3). The fertilisation of the reclaimed land with sewage sludge at a rate of 100 Mg d.m.·ha^{-1} reduced the proportion of the described humus connections to about 37% TOC. The results obtained are confirmed by earlier studies [68]. The lowest share of free and loosely bound humus connections with R_2O_3 and Ca was found in fertiliser variants with mineral wool and NPK. The described connections in the soil of these fertiliser variants accounted for 30.50 to 31.25% TOC. In sites where wool was applied without additives, the content of the described combinations was between 32.35 and 33.82% TOC. In soil rehabilitated with mineral wool with sludge additives, the proportion of humus compounds free and bound to half – oxides and Ca ranged from 39.02 to 31.45% TOC and decreased with increasing mineral wool dosage.

An evaluation of the effect of effective microorganisms on the fractional composition of organic matter showed that EMs applied against the reclamation variants under evaluation significantly increased the percentage share of humus fraction extracted by a mixture of 0.1 mol $Na_4P_2O_7 \cdot dm^{-3}$ + 0.1 mol $NaOH \cdot dm^{-3}$ (Table 3). The application of EM in the control soil increased the proportion of this fraction to 67.31% TOC. In the soil of the other fertilisation variants, the average range of increase in the percentage share of the fraction in question was about 7% in the soil with mineral wool, 9% in the soil + mineral wool + NPK, and about 10% in the soil of the wool + sewage sludge variant.

Table 4 shows the content and the percentage share of humic acids (C–HA) and fulvic acids (C–FA) in humus compounds extracted with a mixture of 0.1 mol $Na_2PO_7 \cdot dm^{-3}$ + 0.1 mol $NaOH \cdot dm^{-3}$. In the soil of the control object, fertilised with NPK, the content of fulvic acids was significantly higher than humic acids and the C–HA:C–FA ratio had a value of 0.44. Such a low C–HA:C–FA ratio indicates a very poor quality of humus connections [44].

Soil fertilisation with sewage sludge had a beneficial effect on the quality of humus compounds. Within the humus connections of free and loosely bound half-oxygen and Ca, fulvic acids still predominated over humic acids, but the value of the C–HA:C–FA ratio increased and was 0.59. In all the fertilisation variants in which Grodan mineral wool was applied, compared to the control soil + NPK, there was no significant effect on the values of the C–HA:C–FA ratio.

Table 3. Fractional composition of organic matter

Fertilization variant	Cdeca.				Cextracted				Humins	
	g·kg^{-1}d.m.		% TOC		g·kg^{-1} d.m.		% TOC		% TOC	
	0	EM	0	EM	0	EM	0	EM	0	EM
Control I – soil + NPK	0.6	0.4	6.76	5.77	4.7	4.2	52.70	67.31	47.30	32.69
Control II – soil + sewage sludge	1.0	0.7	6.67	5.73	5.4	6.0	37.50	52.08	62.50	47.92
Soil + mineral wool 200 m^3·ha^{-1}	0.8	0.7	6.86	5.50	4.1	4.8	33.82	40.00	66.18	60.00
Soil + mineral wool 400 m^3·ha^{-1}	0.7	0.6	6.38	5.83	4.0	4.4	35.11	41.11	64.89	58.89
Soil + mineral wool 800 m^3·ha^{-1}	1.1	0.9	6.43	5.62	5.3	6.4	32.35	38.41	67.65	61.59
Soil + sewage sludge + mineral wool 200 m^3·ha^{-1}	1.2	1.0	6.10	5.36	7.7	8.4	39.02	49.30	60.98	50.70
Soil + sewage sludge + mineral wool 400 m^3·ha^{-1}	1.1	0.8	6.00	5.65	6.0	6.4	33.33	42.74	66.67	57.26
Soil + sewage sludge + mineral wool 800 m^3·ha^{-1}	1.0	0.8	6.85	5.74	4.7	5.8	31.45	39.34	68.55	60.66
Soil + mineral wool 200 m^3·ha^{-1} + NPK	0.7	0.6	6.00	5.71	3.7	4.1	30.50	37.50	69.50	62.50
Soil + mineral wool 400 m^3·ha^{-1} + NPK	0.9	0.6	6.47	5.71	4.2	4.4	31.25	40.22	68.75	59.78
Soil + mineral wool 800 m^3·ha^{-1} + NPK	0.8	0.6	6.60	5.81	4.0	4.4	31.13	43.02	68.87	56.98
Average	0.90	0.70	6.47	5.68	4.89	5.39	35.29	44.64	64.71	55.36
NIR between: - fertilization variant - series (without EM and with EM)	 0.22** 0.05**		 0.90 0.21**		 1.26** 0.29**		 9.14** 2.14**			

Note: Cdeca. – carbon decalcification; Cextracted – carbon extracted.

Table 4. The content and the percentage share of humic acids (C–HA), fulvic acids (C–FA) and the C–HA:C–FA ratio

Fertilization variant	C–HA				C–FA				C–HA:C–FA	
	g·kg^{-1}		% TOC		g·kg^{-1}		% TOC			
	0	EM	0	EM	0	EM	0	EM	0	EM
Control I – soil + NPK	1.44	1.44	16.21	23.08	3.24	2.67	36.40	44.24	0.44	0.52
Control II – soil + sewage sludge	2.01	2.40	14.00	20.83	3.38	3.60	23.50	31.25	0.59	0.67
Soil + mineral wool 200 m^3·ha^{-1}	1.24	1.92	10.19	16.00	2.89	2.88	23.63	24.00	0.43	0.67
Soil + mineral wool 400 m^3·ha^{-1}	1.24	1.82	11.06	16.89	2.71	2.61	24.05	24.22	0.46	0.70
Soil + mineral wool 800 m^3·ha^{-1}	1.53	2.49	9.41	15.07	3.74	3.86	22.94	23.34	0.41	0.65
Soil + sewage sludge + mineral wool 200 m^3·ha^{-1}	2.59	3.36	13.17	19.71	5.08	5.04	25.85	29.59	0.51	0.67
Soil + sewage sludge + mineral wool 400 m^3·ha^{-1}	2.01	2.59	11.20	17.41	3.98	3.76	22.13	25.33	0.51	0.69
Soil + sewage sludge + mineral wool 800 m^3·ha^{-1}	1.63	2.30	10.96	15.73	3.04	3.45	20.49	23.61	0.54	0.67
Soil + mineral wool 200 m^3·ha^{-1} + NPK	1.05	1.44	8.80	13.04	2.60	2.70	21.70	24.46	0.40	0.53
Soil + mineral wool 400 m^3·ha^{-1} + NPK	1.34	1.53	10.00	13.91	2.85	2.90	21.25	26.31	0.47	0.53
Soil + mineral wool 800 m^3·ha^{-1} + NPK	1.24	1.53	9.81	14.98	2.71	2.90	21.32	28.13	0.46	0.53
Average	1.57	2.07	11.35	16.97	3.29	3.31	23.93	27.68	0.47	0.62
NIR between: - fertilization variant - series (without EM and with EM)	1.27** 0.19**		2.88** 0.68**		0.68** 0.16		8.14** 1.91**		0.20 0.05**	

Note: C–HA – carbon of humic acids; C–FA – carbon of fulvic acids; C–HA:C–FA – ratio of carbon of humic acids to carbon of fulvic acids.

Regardless of the reclamation variant, an increase in the ratio of humic acids to fulvic acids was found after EM application (Table 4), indicating a positive effect of EM on soil fertility. At the same time, the greatest increase in this ratio was recorded in the reclamation variants with mineral wool. The introduction of EM into the soil significantly increases the activity of enzymes involved in the decomposition of cellulose, which may indicate the mineralisation processes of organic matter, and at the same time, it may be the beginning of the humification process [29]. The humification process involves polycondensation and polymerisation of lower molecular weight compounds, which result from the mineralisation of compounds with complex structures (cellulose, lignin) [69]. The results obtained, including the increase in the share of C–HA in soils with the addition of EM, indicate a significant role for EM in activating soil organic matter transformation processes.

The content and percentage share of humins (extraction residues) was inversely related to the previously discussed content of the free and loosely bound to half – oxygen and Ca fractions. In the soil of the control object, compounds constituting the extraction residue (humins) accounted for 47.30% TOC (Table 3) In the soil fertilised with sewage sludge, humins accounted for 62.4% TOC. A further increase in the share of humins was found in the soil with the addition of mineral wool. The percentage share of humins in soil + wool + sludge series ranged from about 64% TOC to about 68% TOC. Such a favourable effect of mineral wool on the share of humis should be linked to an improvement in the water and air conditions in the soils of these sites, which in turn had a favourable effect on the direction of organic matter transformation – increased humification at the expense of organic matter mineralisation [65, 70]. Effective microorganisms influenced a reduction in the percentage share of humins in the reclaimed soil (Table 3).

CONCLUSIONS

The application of effective microorganisms (EM), against the background mineral wool and wool applied together with sewage sludge or NPK, influenced significant changes in the content and quality of soil organic matter. Mineralization processes prevailed in the transformation of humus compounds of the soils on which effective microorganisms were applied, as indicated by the recorded decrease in the content of TOC, humins, and an increase in the percentage share of labile fractions and humus compounds free and loosely bound to R_2O_3 and Ca. More stable humus compounds were formed during the humification process, as evidenced by the observed increase in the percentage share of humic acids and the C–HA:C–FA ratio in the soil on which effective microorganisms were applied. Supplementing commonly used materials in soil remediation with the introduction of effective microorganisms is a promising technology. Further research is needed to determine the composition of these biofertilizers and the optimal doses at which they should be applied.

REFERENCES

1. Hariram N.P., Mekha K.B., Suganthan V., Sudhakar K. Sustainalism: An integrated socio-economic-environmental model to address sustainable development and sustainability. Sustainability 2023; 15(13): 10682.
2. Intergovernmental Panel on Climate Change (IPCC). Summary for policymakers. In: Pachauri, R.K., Meyer, L.A. (Eds.), Contribution of Working Groups I, II, and III to the Fifth Assessment Report of the Intergovernmental Panel on Climate Change. Climate Change 2014 Syntyhesis Report. Geneva: IPCC.
3. Chu E.W., Karr J.R. Environmental impact: concept, consequences, measurement. Reference Module in Life Sciences, 2017.
4. Lal R., Horn R., Kosaki T. (Eds.). Soil and Sustainable Development Goals, Schweizerbart, Stuttgart, 2018.
5. Helming K., Daedlow K., Paul C., Techen A., Bartke S., Bartkowski B., Kaiser D. B, Wollschläger U., Vogel H. J. Managing soil functions for a sustainable bioeconomy – assessment framework and state of the art. Land Degradation & Development 2018; 29: 3112-3126.
6. Bouma J. Soil security as a roadmap focusing soil contributions on sustainable development agendas. Soil Security 2020; 1:100001.
7. Bonfante A., Basile A., Bouma J. Targeting the soil quality and soil health concepts when aiming for the United Nations Sustainable Development Goals and the EU Green Deal. Soil, 2020, 6, 453-466.
8. Löbmann M. T., Maring L., Prokop G., Brils J., Bender J., Bispo A., Helming K. Systems knowledge for sustainable soil and land management. Science of The Total Environment 2022; 822: 153389.
9. Veerman C., Correia T. P., Bastioli C., Biro B., Bouma J., Cienciala E., Emmett B., Frison E.A., Grand A., Filchew L.H., Kriaučiūnienė Z., Pogrzeba M., Soussana J.F., Olmo C.V., Wittkowski R. Caring for soil is caring for life-ensure 75% of soils are healthy by 2030 for food, people, nature and climate. European Commission, Brussels, 2020.
10. Sheoran V., Sheoran A.S., Poonia P. Soil reclamation of abandoned mine land by revegetation. International Journal of Soil, Sediment and Water 2010; 3, 13.
11. Wang J., Guo L., Bai Z., Yang, L. Using computed tomography (CT) images and multi-fractal theory to quantify the pore distribution of reconstructed soils during ecological restoration in opencast coal-mine. Ecological Engineering 2016; 92: 148–157.
12. Feng J., Wang Z., Bai L. Reading effects of surface coal mining and land reclamation on soil properties: A review. Earth-Science Reviews 2019; 191: 12–25.
13. Myszura M., Żukowska G., Kobyłka A., Mazurkiewicz J. Enzymatic activity of soils forming on an afforested heap from an opencast sulphur mine. Forests 2021; 12(11): 1469.
14. Bik-Małodzińska M., Żukowska G., Paśmionka I., Rybczyńska-Tkaczyk K., Jakubczyk A. Assessment of the effectiveness of soil reclamation techniques degraded by the sulfur industry. Advances in Science and Technology Research Journal 2022; 16(5): 324-334.
15. Pihlap E., Vuko M., Lucas M., Steffens M., Schloter M., Vetterlein D., Endenich M., Kögel-Knabner I. Initial soil formation in an agriculturally reclaimed open-cast mining area – the role of management and loess parent material. Soil and Tillage Research 2019; 191: 224-237.
16. Larney Francis J., Angers Denis A. The role of organic amendments in soil reclamation: A review. Canadian Journal of Soil Science 2012; 92(1): 19-38.
17. Babcock-Jackson L., Konovalova T., Krogman J.P., Bird R., Díaz L.L. Sustainable fertilizers: publication landscape on wastes as nutrient sources, wastewater treatment processes for nutrient recovery, biorefineries, and green ammonia synthesis. Journal of Agricultural and Food Chemistry 2023; 71(22): 8265-8296.
18. Kurniawati A., Toth G., Ylivainio K. Opportunities and challenges of bio-based fertilizers utilization for improving soil health. Organic Agriculture 2023; 13: 335–350.
19. Nazranov K.M., Didanova E.N., Khalishkhova L.Z. Formation and development of the biopreparations market. IOP Conf. Series: Earth and Environmental Science 2021; 699: 012037.
20. Priya A.K., Alagumalai A., Balaji D., Song H. Bio-based agricultural products: a sustainable alternative to agrochemicals for promoting a circular economy. RSC Sustainability 2023; 1: 746-762.
21. Singh S.P., Ekanem E., Wakefield T., Comer S. Emerging importance of bio-based products and bio-energy in the U.S. economy: information dissemination and training of students. Int Food Agribus Manag Rev 2023; 5: 1–15.
22. Pylak M., Oszust K., Frąc M. Review report on the role of bioproducts, biopreparations, biostimulants and microbial inoculants in organic production of fruit. Reviews in Environmental Science and Bio/Technology 2019; 18: 597–616.
23. Talaat N.B. Effective microorganisms: An innovative tool for inducing common bean (*Phaseolus*

vulgaris L.) salt-tolerance by regulating photosynthetic rate and endogenous phytohormones production, Scientia Horticulturae 2019; 250: 254-265.

24. Higa T. Effective microorganisms – their role in kyusei nature farming and sustainableagriculture. In: Proceedings of the Third International Conference on Kyusei NatureFarming. Ed. J.F. Parr et al., USDA, Washington, 1996, USA:20-24.
25. Goessler W., Kuehenelt D. Analytical methods for the determination of arsenic and arsenic compounds in the environment. In: William T, Frankenberger Jr William (eds) CRC Press, 2002: 27–50.
26. Zakaria Z., Gairola S., Shariff N.M. Effective microorganisms (EM) technology for water quality restoration and potential for sustainable water resources and management. In: Proc. of the 2010 International Congress on Environmental Modelling and Software Modelling for Environment's Sake, Fifth Biennial Meeting, Ottawa, ON, Canada, 5–8 July 2010.
27. Han H.S., Supanjani E., Lee K.D. Effect of co-inoculation with phosphate and potassium solubilizing bacteria on mineral uptake and growth of pepper and cucumber. Plant, Soil and Environment 2006; 52(3): 130–136.
28. Pranagal J., Ligęza S., Smal H. Impact of effective microorganisms (EM) application on the physical condition of haplic luvisol. Agronomy 2020; 10: 1049.
29. Piotrowska A., Boruszko D. Analysis of the influence of the application of effective microorganisms on the dynamics of spring wheat. Emergence 2021; 23: 684-693.
30. Pszczółkowski P., Sawicka B., Barbaś P., Skiba D., Krochmal-Marczak B. The use of effective microorganisms as a sustainable alternative to improve the quality of potatoes in food processing. Applied Sciences 2023; 13: 7062.
31. Tyburski J., Łachacz A. The effectiveness of agents improving heavy soils in organic farms. In: Summary of Results of Studies on Organic Farming Realized in 2009. Ministry of Agriculture and Rural Development: Warsaw, Poland, 2010; 318: 267–276.
32. Jakubus M., Gajewski P., Kaczmarek Z., Mocek A. Impact of addition of organic additives and EM-A preparation on physical, chemical and structural state of the arable-humus soil horizon. J Res Appl Agric Engin 2013; 58(3): 220–225.
33. Zhao Y., Li W., Zhou Z., Wang L., Pan L., Zhao L. Dynamics of microbial community structure and cellulolytic activity in agricultural soil amended with two biofertilizers. European Journal of Soil Biology 2005; 41: 21–29.
34. Dziamba S., Pranagal J., Wielgosz E. Some properties of Haplic Luvisol after one-year application of effective microorganisms (EM). Advances of Agricultural Sciences Problem 2009; 542: 139–145.
35. Tołoczko W., Trawczyńska A., Niewiadomski A. Content of organic compounds in soils fertilized with EM preparation. Soil Science Annual 2009; 60: 97–101.
36. Badura L. Soil microorganisms and their importance in ecosystems degraded by humans. Advances of Agricultural Sciences Problem 2005; (505):15-23.
37. Bao N., Wu L., Ye B., Yang K., Zhou W. Assessing soil organic matter of reclaimed soil from a large surface coal mine using a field spectroradiometer in laboratory. Geoderma 2017; 288: 47-55.
38. Woś B., Józefowska A., Chodak M., Pietrzykowski M. Recovering of soil organic matter and associated C and N pools on regenerated forest ecosystems at different tree species influence on post-fire and reclaimed mine sites. Geoderma Regional 2023; 33.
39. Dębska B., Długosz J., Piotrowska-Długosz A., Banach-Szot M. The impact of a bio-fertilizer on the soil organic matter status and carbon sequestration – results from a field-scale study. Journal of Soils and Sediments 2016; 16: 2335–2343.
40. Baran S., Urban D., Wójcikowska-Kapusta A., Bik-Małodzińska M., Żukowska G. Phytoindicative evaluation of habitat conditions of soilless formations reclaimed with flotation sludge, sewage sludge and used mineral wool under the influence of the Jeziórko Sulphur Mine. Journal of Elementology 2015; 20(1): 7-18.
41. PN-EN 15936:2013:02. Available online: https://sklep.pkn.pl/pn-en-15936-2013-02p.html (accessed on 10 January 2024).
42. International Organization for Standardization. Soil Quality. In: Determination of Total Nitrogen Content by Dry Combustion; International Organization for Standardization: Geneva, Switzerland, 1998.
43. Myśków W., Zięba S. The influence of long term fertilization on the biological activity and organic substances of soil. Polish Journal of Soil Sciences 1981; 14(2): 141-151.
44. Kononova M.M. Soil Organic Matter: its Nature, its Role in Soil Formation and in Soil Fertility. Pergamon Press Ltd. Oxford, 1966.
45. Obalum S.E., Chibuike G.U., Peth S., Ouyang Y. Soil organic matter as sole indicator of soil degradation. Environmental Monitoring and Assessment 2017; 189(4): 176.
46. Franzluebbers J. Soil organic matter stratification ratio as an indicator of soil quality. Soil and Tillage Research 2002; 66(2): 95-106.
47. Cotrufo M.F., Lavallee J.M. Chapter One – Soil organic matter formation, persistence, and functioning: A synthesis of current understanding to inform its conservation and regeneration, Editor(s): Donald L. Sparks, Advances in Agronomy, Academic Press 2022; 172: 1-66.
48. Balík J., Kulhánek M., Černý J., Sedlář O., Suran P., Procházková S., Asrade D.A. The impact of the long-term application of mineral nitrogen and sewage sludge fertilizers on the quality of soil organic matter. Chemical and Biological Technologies in Agriculture 2022; 9: 86.

49. Körschens M., Albert E., Armbruster M., Barkusky D., Baumecker M., Behle-Schalk L., Bischoff R., Čergan Z., Ellmer F., Herbst F., Hoffmann S., Hofmann B., Kismanyoky T., Kubat J., Kunzova E., Lopez-Fando C., Merbach I., Merbach W., Pardor M.T., Rogasik J., Rühlmann J., Spiegel H., Schulz E., Tajnsek A., Toth Z., Wegener H., Zorn W. Effect of mineral and organic fertilization on crop yield, nitrogen uptake, carbon and nitrogen balances, as well as soil organic carbon content and dynamics: results from 20 European long-term field experiments of the twenty–first century. Journal Archives of Agronomy and Soil Science 2013; 59: 8.
50. Woźniak M.M., Siebielec S., Siebielec G., Bojarszczuk J., Gałązka A., Urbaniak M. Microbially modified effect of exogenous organic matter on soil chemical and biological indices and plant responses. Journal of Soils and Sediments 2024; 24: 70–85.
51. Achkir A., Aouragh A., El Mahi M., Labjar N., Bouch M.E., Ouahidi M.L., Moussaoui T.E. Implication of sewage sludge increased application rates on soil fertility and heavy metals contamination risk. Emerging Contaminants 2023; 9(1): 100200.
52. Haddix M.L., Gregorich E.G., Helgason B.L., Janzen H., Ellert B.H., Francesca Cotrufo M. Climate, carbon content, and soil texture control the independent formation and persistence of particulate and mineral-associated organic matter in soil. Geoderma 2020; 363: 114160.
53. Bolinder M.A., Crotty F., Elsen A., Frac M., Kismányoky T., Lipiec J., Tits M., Tóth Z., Kätterer T. The effect of crop residues, cover crops, manures and nitrogen fertilization on soil organic carbon changes in agroecosystems: a synthesis of reviews. Mitigation and Adaptation Strategies for Global Change 2020; 25: 929–952.
54. Valarini P.J., Díaz M.C., Gascó J.M., Guerrero F., Tokeshi H. Assessment of soil properties by organic matter and EM-microorganisms incorporation. Revista Brasileira de Ciência do Solo 2003; 27: 519–525.
55. Wu S.C., Cao Z.H., Li Z.G., Cheung K.C., Wong M.H. Effects of biofertilizer containing N-fixer, P and K solublizers and AM fungi on maize growth: a greenhouse trial. Geoderma 2005; 125: 155–166.
56. Nisha R., Kaushik A., Kaushik C.P. Effect of indigenous cyanobacterial application on structural stability and productivity of an organically poor semi-arid soil. Geoderma 2007; 138: 49–56.
57. Schenck zu Schweinsberg-Mickan M., Müller T. Impact of effective microorganisms and other biofertilizers on soil microbial characteristics, organic matter decomposition, and plant growth. Journal of Plant Nutrition and Soil Science 2009; 172: 704–712.
58. Li X., Guo Q., Wang Y., Xu J., Wei Q., Chen L., Liao L. Enhancing nitrogen and phosphorus removal by applying effective microorganisms to constructed wetlands. Water 2020; 12: 2443.
59. Żukowska G., Baran S., Pawłowski A., Myszura M., Wójcikowska-Kapusta A., Wesołowska S., Pawłowska M. Reclamation of drill cuttings landfill. Annual Set the Environment Protection 2016; 18: 988-1006.
60. Woś B., Pająk M., Pietrzykowski M. Soil organic carbon pools and associated soil chemical properties under two pine species (*Pinus sylvestris* L. and *Pinus nigra* Arn.) introduced on reclaimed sandy soils. Forests 2022; 13(2): 328.
61. Haynes R.J. Labile organic matter fractions as central components of the quality of agricultural soils: an overview. Advances in Agronomy 2005; 85: 221-268.
62. Żukowska G., Myszura M., Zdeb M., Pawłowska M. Carbon sequestration in soil as a sustainable way of greenhouse effect mitigation. Problems of Sustainable Development 2020; 15(2): 195-205
63. Żukowska G., Flis-Bujak M., Baran S. Changes in the content of labile fractions of organic matter in light soil fertilized with sewage sludge. Folia Universitatis Agriculturae Stetinensis 1999; (77): 429-436.
64. Joshi H., Choudhary S.P., Mundra S.L. Role of effective microorganisms (EM) in sustainable agriculture. International Journal of Current Microbiology and Applied Sciences 2019; 8(3): 172-181.
65. Orlov D.S. Humus acids of soils, 1st edn. A.A. Balkema, Rotterdam 1986.
66. Wiesmeier M., Urbanski L., Hobley E., Lang B., von Lützow M., Marin-Spiotta E., van Wesemael B., Rabot E., Ließ M., Garcia-Franco N., Wollschläger U., Vogel H., Kögel-Knabner I. Soil organic carbon storage as a key function of soils – A review of drivers and indicators at various scales. Geoderma 2019; 333: 149-162.
67. Yang Z.H., Singh B.R., Sitaula B.K. (2004) Soil organic carbon fractions under different land uses in Mardi Watershed of Nepal. Commun Soil Sci Plant Anal 2004; 35:615–629.
68. Żukowska G., Baran S., Wójcikowska-Kapusta A. Organic carbon content and fractional composition of organic matter in soil reclaimed with sewage sludge. Przemysł Chemiczny 2012; 91(6): 1267-1269.
69. Mayer J., Scheid S., Widmer F., Fließbach A., Oberholzer H.R. How effective are effective microorganisms (EM)? Results from a field study in temperate climate. Applied Soil Ecology 2019; 46(2): 230-239.
70. Lal R., Bouma J., Brevik E., Dawson L., Field D. J., Glaser B., Hatano R., Hartemink A. E., Kosaki T., Lascelles B., Monger C., Muggler C., Ndzana G. M., Pan N. X., Paradelo R., Reyes-Sánchez L. B., Sandén T., Singh B. R, Spiegel H., Yanai J., Zhang J. Soils and sustainable development goals of the United Nations: An International Union of Soil Sciences perspective. Geoderma Regional 2021; 25.