JMBFS
Journal of Microbiology, Biotechnology and Food Sciences
International peer-reviewed scientific online journal
Published by Faculty of Biotechnology and Food Sciences

MATHEMATICAL MODEL FOR DETERMINING THE INFLUENCE OF SOME ABIOTIC FACTORS ON MICROBIOLOGICAL ACTIVITY IN TWO COMPOSTING VARIANTS

*Mariya Konsulova-Bakalova**[1], *Boyka Malcheva*[2], *Pavlina Naskova*[1]

Address(es):
[1] Technical University – Varna, Faculty of Manufacturing Engineering and Technologies, 1 Studentska Str., Varna, Bulgaria.
[2] University of Forestry, Faculty of Forestry, „Silviculture", 10 Kliment Ohridski Blvd., Sofia, Bulgaria.

*Corresponding author: mbakalova@tu-varna.bg

https://doi.org/10.55251/jmbfs.12077

ARTICLE INFO

Received 19. 11. 2024
Revised 9. 5. 2025
Accepted 20. 5. 2025
Published 1. 8. 2025

Regular article

OPEN ACCESS

ABSTRACT

Microbiological activity in composting is a complex process that depends on many abiotic factors. The mathematical model presented in this article is based on collected empirical data, on the basis of which the problem of predicting the values of several independent variables is solved. A standard statistical procedure of correlation matrix calculation and regression analysis is used. Two compost variants, differing in the microbiological agent added in one of them, were set. The amount of microorganisms was higher in the variant with microbial addition, and in both variants the amount and activity of microorganisms decreased during the thermophilic phase of composting. Actinomycetes take the major part of the total microflora, followed by non-spore-forming bacteria, except in the thermophilic phase, where the proportion of spore-forming bacteria is higher and mould fungi completely die out. The results of the laboratory tests determining the microbiological activity and the results of the regression models for each stage of the two composting treatments under study were obtained. The presented mathematical model can be used to predict microbiological activity during composting under different abiotic conditions and to optimize the process.

Keywords: composting, microflora, regression model

INTRODUCTION

The decomposition of organic matter involving soil biota is a process that has existed and been known since antiquity (**Billington, 1956; Gotaas, 1956; Diaz and De Bertoldi, 2007; Milena**). Composting converts raw organic materials into a biologically stable, humic-like product (**Ceglie and Abdelrahman, 2014; Graves and Hattemer, 2000**). Compost can be used in agriculture, horticulture and forestry, as well as landscaping, home gardens and in land reclamation as an organic conditioner (**Cooperband, 2000**).
Non-spore-forming bacteria and bacilli play a major role in the composting process, while actinomycetes and micromycetes are less represented (**Malcheva et al., 2018, Naskova *et al.*, 2024; Grigorova-Pesheva, 2019; Grigorova-Pesheva and Malcheva, 2023**). Actinomycetes develop much more slowly than bacteria and fungi and do not compete with them in the early stages of composting, but are more noticeable in the later stages of the process when they become abundant and the typical white or grey cast of actinomycetes is clearly visible at a depth of 10 cm from the surface of the composted mass (**Nozhevnikova, et al., 2019**). Streptomycetes such as *Streptomyces griseus*, *S. thermoviolaceus*, *S. globisporus*, *S. ruber* and *S. viridosporus* (**Chanal *et al.*, 2006**) and filamentous fungi of the genera *Mucor*, *Rhizopus* and *Aspergillus* (**Kango *et al.*, 2019**) are mainly found in composting. They play an important role in the decomposition of organic matter in compost mass. Not only bacteria (over 2000 species), fungi (over 100 species), actinomycetes, but also invertebrates are actively involved in the composting process (**Steel & Wim, 2012; De Gannes *et al.*, 2013; Sánchez *et al.*, 2017; Nozhevnikova *et al.*, 2019**).
The intensity of the composting process and the rate of oxidation of organic matter are influenced by the temperature, humidity, pH of the composted mass, t the multidispersity of the substrate that provides aeration, the presence of nutrients in an available form, the development of microflora, microfauna and macrofauna. Temperature is an important factor in the composting process and in differentiating the different phases of composting. In the thermophilic phase, at temperatures above 55 °C the fungi die, and once the temperature is lowered they again spread from colder areas throughout the volume (**Tuomela *et al.*, 2000**).
Statistical methods are often used to assess the influence of the above factors on the intensity of the composting process. In the study of **Chikae *et al.* (2006)**, for example, multiple regression is used to investigate the maturity of compost from food waste and wood chips. The idea proposed by the authors is to predict the so-called compost maturity (based on germination index (GI) value) by only some specified indicators without requiring large sized equipment. Multiple regression has also been used to investigate the effects of aeration, seeding, and agitation on compostability of plant wastes (**Chang *et al.*, 2006**). In addition to regression methods, some authors (**Yildiz & Degirmenci, 2015**) have proposed the use of neural networks. The inclusion of such methods, however, should be truly upon a proven need for them. Although, in recent years, they have been increasingly used in various fields, in most cases a good solution can have been achieved with simpler classical methods. The idea of this article is not only to present the results obtained in a specific experiment on the microbiological activity in two composting variants, but also to give a mathematical description of the variation of the amount of microflora in composts depending on several preselected factors. Usually, factors are quantities whose values are easily and accessibly measured without the need for specialized equipment. Thanks to such a mathematical model, the composition of the microflora in composts can be predicted by the values of the factors.
The aim of the study was to analyze the quantity, composition and activity of the compost microflora in the composting phases and to create a mathematical model to determine the influence of abiotic factors on the microbiological activity in the composting process.

MATERIAL AND METHODS

Used variants and experimental methods

Two spring compost variants (compost 1 without additive and compost 2 with additive) were created and analyzed under indoor composting (in composters) located at the experimental field of the Technical University - Varna. Both composts contain shredded plant residues from: lemons, potatoes, cucumbers, tomatoes, zucchini, wheat, fruit tree leaves. Soil was used as starter. A microbial additive (solution concentration 1:100, 25 l per 1 m^3) containing lactic acid and photosynthetic bacteria, nitrogen-fixing bacteria, yeasts of the genus *Saccharomyces* was added to the one compost.
In the composting phases, temperature, humidity and pH were monitored in the compostable mixtures. For the microbiological analysis of compost variants (after dilution), a method with three times inoculation of solid nutrient media followed by counting of colony- forming units (CFU) in 1 g of absolutely dry compost was used (Mishustin and Emtsev, 1989; Malcheva and Naskova, 2018; Nustorova and Malcheva, 2020). Systematic and physiological aerobic groups of microorganisms were determined - spore-forming bacteria (bacilli, including lactobacilli) and non-spore-forming bacteria (on mesopeptone agar), micromycetes (mould fungi) - on Chapek-Dox agar, actinomycetes and mineral nitrogen-utilizing bacteria (on Actinomycetes isolation agar). The total microflora was calculated as the sum of

the studied groups of microorganisms (non-spore-forming bacteria, bacilli, actinomycetes, mould fungi). The mineralization coefficient was calculated as the ratio between bacteria utilizing mineral nitrogen and the sum of non-spore-forming bacteria and bacilli (Mishustin & Runov, 1957; Malcheva & Naskova, 2018).

Statistical processing of the experimental data

The aim of the study is to search for a mathematical model describing the amount of microflora in composting. Essentially, the problem is to predict the values of several independent variables based on previously collected empirical data from the experiments described above. Usually, solving such a problem requires conducting a mathematical analysis with finding dependencies between preselected factors. In this case these are temperature, humidity and pH of compost variants. For simplicity, let us name these factors in the following with x_1 - temperature, x_2 - pH, x_3 - humidity, respectively. Here $X = [x_1 ; x_2 ; x_3]$ is a matrix of independent variables and Y is a matrix of microflora quantity. For the purpose of the study, it is assumed that there is no statistical dependence between the factors in the X matrix. The problem of finding and eventually describing a mathematical relationship between the elements x_i (i=1,2,3) of the matrix of independent variables X and the components of the matrix of dependent variables Y is posed. Typically, correlation or covariance matrices are used to find a statistical relationship. The covariance indicates the direction of the linear relationship between the variables, while the correlation measures both the strength and direction of the linear relationship between two variables. In other words, the correlation analysis task is to establish the degree of influence of factors on the trait. Correlation analysis allows to manifest the unknown relationships between factors and the trait, to determine the principal components - the factors that have the greatest influence on the variation of trait values. The actual procedure of calculating the correlation matrix and the interpretation of its elements will not be discussed in detail here, as it is a standard statistical procedure (Gogtay, & Thatte, 2017; Senthilnathan, 2019). In the case as a measure by whose interpretation the presence of statistical dependence will be judged, the one described in (Senthilnathan, 2019). will be used. If the correlation coefficient is R_{YX}, then: For $0 \leq R_{YX} < 0.3$, the correlation is considered as weak; At $0.3 \leq R_{YX} < 0.5$, the correlation is moderate; At $0.5 \leq R_{YX} < 0.7$, the correlation is significant; At $0.7 \leq R_{YX} < 0.9$, the correlation is strong; At $0.9 \leq R_{YX} < 1.0$, the correlation is very strong; At $R_{YX} = 1$, the correlation relation becomes functional.
The next step is usually regression analysis (Draper & Smith, 1998). Through regression analysis, possible functional relationships between two or more random variables can be studied and evaluated. The main questions answered by the analysis are whether a functional dependence exists between two dependent random variables and, if so, to find a function that describes it sufficiently accurately.
The dependence between random variables in real conditions can be different. If the dependence between X and Y is so strong that if it is known what value one quantity X has taken, can be obtained the exact value of Y, then the relationship between X and Y is functional. At the same time, the relationship between random variables may often not be strictly functional. Such examples are particularly characteristic of such areas of science and practice as biology (Naskova et al., 2017), medicine, agricultural engineering, economics, etc., where the development of various processes and phenomena, as a rule, depends on many factors that can hardly be accounted for in their completeness. In such situations, when the change of one quantity affects another only statistically, it is customary to speak of statistical dependence between quantities. In particular, statistical dependence manifests itself in the fact that a change in one of the quantities changes the average value of the other; in this case, statistical dependence is called correlation. The existence of a correlation relationship means that observed changes in the values of one quantity correspond changes in the values of the other quantity.
The functional relationship between X and Y is called the regression equation:

$$y=f(x_1, x_2, ..., x_m), \quad (1)$$

where m - number of independent factors.
If the regression function is linear, it is said to be a linear regression model. Otherwise, the regression model is called nonlinear. In the present problem we assume that there is linear relationship and therefore in the following exposition we will discuss the features of a linear regression analysis. If the variable y depends on only one independent variable x or m=1, we have the first-degree linear regression, which in practice can be achieved very rarely. However, from a practical point of view, the situation where the quantity y depends on a set of m variables ($x_1, x_2, ..., x_m$) is much more realistic. The regression is called multivariable linear regression. The regression analysis or finding the regression equation means on the calculation of the β coefficients in equation (1), also called regression coefficients:

$$y=\beta_0+\beta_1 x_1+\beta_2 x_2+...+\beta_m x_m, \quad (2)$$

larger values of the β coefficients are associated with greater change in the dependent variable. Negative values mean that an increase in the corresponding independent variable leads to a decrease in the value of the dependent variable and vice versa - when the corresponding coefficient is greater than zero, its increase leads to an increase in Y as well. However, when comparing the coefficients by absolute value, the variance of the variables must also be taken into consideration. A prerequisite for creating regression models is determining the static significance of the estimated coefficients in the regression equation. In (Naskova, Konsolova, Plamenov, & Malcheva 2017) is described the procedure of finding quality measures of the regression model.
Here we turn our attention to one of the regression analysis results on which one can reasoned about the adequacy of the resulting models, namely the difference between the value predicted by the model and the experimental observation - the so-called residual. In establishing regression relationships, a comparison can be made between the empirical data and those obtained by the regression model. The difference between them is:

$$e_i = y_{ipred} - y, \quad (3),$$

where y_{pred} are the values for the dependent variables obtained from the regression model or so-called predicted outcomes, and e_i are the residuals for the i-th observation. In this sense, the matrix notation of the regression equations is:

$$y=X.\beta+e, \quad (4)$$

where y is a vector of measurements, f is a vector of residuals, and X is a vector of independent variables.
Traditionally, the residuals, and more precisely the sum of the squares of the respective residuals, and the minimization and are used in assessing the adequacy of regression models. In this article, the values of the respective residuals are used to compare the models obtained for different composting variants.

RESULTS AND DISCUSSION

Experimental data

Table 1 shows the quantity of the respective groups of microorganisms and their mineralization activity during the different composting phases. Two composting variants, with and without additive, were analyzed.
Actinomycetes take a major share of the total microflora, with the highest levels in all phases of composting. This is followed by non-spore-forming bacteria in the mesophilic, maturation and maturation phases. In the thermophilic phase, the amount of bacilli is higher than that of non-spore-forming bacteria, since bacilli as spore-forming bacteria survive to a higher extent at the elevated temperature in the thermophilic phase. Raising the temperature to 55-60 °C in the thermophilic phase leads to complete death of the mould fungi. In the mesophilic and maturation phases, the amount of mould fungi is higher than that of bacilli, while in the maturation phase the amount is close. The rate of mineralisation of the organic matter in terms of mineralisation coefficient values is higher in the maturation and mesophilic phases and significantly lower in the thermophilic phase. In general, non-spore-forming bacteria and bacilli are more active in the initial stages of organic matter destruction, whereas actinomycetes and mould fungi play a more active role in the final stages of organic matter degradation. When analyzing other compost variants, the highest amount of non-spore-forming bacteria was found in the composting phases (**Malcheva *et al.*, 2018; Grigorova-Pesheva, 2019; Grigorova-Pesheva and Malcheva, 2023; Grigorova-Pesheva *et al.*, 2023; Naskova *et al.*, 2024**).
Soil and soil-compost microbiome composition and activity are dependent on a complex of anthropogenic and environmental factors (**Malcheva, 2021; Malcheva *et al.*, 2018a; 2018b; 2019a; 2019b; 2020; Naskova *et al.*, 2015; 2016; Plamenov *et al.*, 2016; Yankova *et al.*, 2016**). Humidity and temperature are major factors influencing the development and activity of soil and soil-compost microflora (**Koleva *et al.*, 2024**). **Konsulova *et al.* (2017)** developed a statistical model for the recognition and prediction of soil microbiological activity by indirect signs, incorporating into the model some of the main factors influencing the composition and activity of microorganisms: soil temperature and moisture, sampling depth.

Statistical analyses in composting without additive

The calculated correlation coefficients for composting without additive are presented in Table 2.
The correlation is considered to be significant at values between 0.5 and 0.7, and above these values it is strong (0.7÷0.9) and very strong (0.9÷1). In this case, the data on the correlation coefficient show that the amount of bacteria and bacilli utilizing mineral nitrogen is only slightly affected by humidity. Based on the results obtained from the regression analysis, it may be assumed that the humidity factor could be excluded from the mathematical model.
Using linear regression, regression models were made for the groups of microorganisms indicated in Fig. 1. It can be clearly seen that the regression models repeat the pattern of variation of the experimental data, but for some of them there is a significant shift in the values. Table 3 gives the values for R^2 (coefficient of determination) and F (Fisher's criterion).

Table 1 Quantity (cfu/g), composition and activity of compost microflora

Variants	Total microflora	Non-spore-forming bacteria	Bacilli	Lacto-bacilli	Actino-mycetes	Micro-micetes	Bacteria assimilating mineral nitrogen	Minera-lization coefficient
Mesophilic phase - 2nd day								
Compost 1	3783000	1415200	180000	50000	1852200	285600	800200	0,486
Compost 2	4400600	1685000	198400	63200	2154000	300000	950000	0,488
Mesophilic phase - 12th day								
Compost 1	3784000	1415800	180200	50000	1852400	285600	800400	0,486
Compost 2	4401600	1685400	198600	63600	2154000	300000	950400	0,488
Mesophilic phase - average values								
Compost 1	3783500	1415500	180100	50000	1852300	285600	800300	0,486
Compost 2	4401100	1685200	198500	63400	2154000	300000	950200	0,488
Thermophilic phase - 13th day								
Compost 1	1615200	28200	135000	12000	1440000	0	27800	0,159
Compost 2	1702600	30000	158200	14400	1500000	0	29200	0,144
Thermophilic phase - 15th day								
Compost 1	1614600	27600	135000	12000	1440000	0	27400	0,157
Compost 2	1701800	29400	158200	14200	1500000	0	29000	0,144
Thermophilic phase - average values								
Compost 1	1614900	27900	135000	12000	1440000	0	27600	0,158
Compost 2	1702200	29700	158200	14300	1500000	0	29100	0,144
Cooling phase - 17th day								
Compost 1	3221400	1142000	162000	42300	1656500	218600	528500	0,393
Compost 2	3535600	1385200	175000	52000	1685000	238400	722000	0,448
Cooling phase - 28th day								
Compost 1	3222800	1142600	162400	42400	1656600	218800	528600	0,392
Compost 2	3538400	1386200	175200	52200	1686000	238800	722400	0,448
Cooling phase - average values								
Compost 1	3222100	1142300	162200	42350	1656550	218700	528550	0,392
Compost 2	3537000	1385700	175100	52100	1685500	238600	722200	0,448
Ripening phase - 32nd day								
Compost 1	3536000	1360000	256000	48000	1600000	272000	864000	0,519
Compost 2	3968800	1508800	328000	65600	1738400	328000	951200	0,500
Ripening phase - 51st day								
Compost 1	3523400	1351000	255000	47200	1599000	271200	865000	0,523
Compost 2	3966200	1508000	327000	65400	1738200	327600	950800	0,500
Ripening phase - average values								
Compost 1	3529700	1355500	255500	47600	1599500	271600	864500	0,521
Compost 2	3967500	1508400	327500	65500	1738300	327800	951000	0,500

Table 2 Correlation coefficients

Indicators	Non-spore-forming bacteria	Bacilli	Lacto-bacilli	Actino-mycetes	Micro-micetes	Bacteria assimilating mineral nitrogen
Temperature (°C) - average	-0,871	-0,780	-0,866	-0,633	-0,887	-0,946
pH	-0,969	-0,556	-0,969	-0,715	-0,959	-0,892
Humidity (%)	0,541	-0,252	0,558	0,879	0,534	0,373

Table 3 Values for R^2 and F when creating regression models of microflora in composting without additive

Indicators	Non-spore-forming bacteria	Bacilli	Lactobacilli	Actinomycetes	Micromicetes	Bacteria assimilating mineral nitrogen
R^2	0,968793234	0,924555039	0,968234999	0,913236946	0,964794415	0,949207982
F	186,2659972	73,52817417	182,8871337	63,15385889	164,4275055	112,1287977
Pr > F	<0,0001	<0,0001	<0,0001	<0,0001	<0,0001	<0,0001

The closer the coefficient of determination is to 100%, the better the model is. According to it, the best results are obtained for non-spore-forming bacteria and lactobacilli.

The reason for graphical results offset could be the inclusion of a non-significant factor in the relevant mathematical models. On this basis and the previously calculated correlation coefficients, a new regression analysis procedure was conducted with a reduced number of factors for some of the microflora species. The resulting regression models are shown in Fig. 2 and significantly better describe the variation of the amount of microflora in the compost.

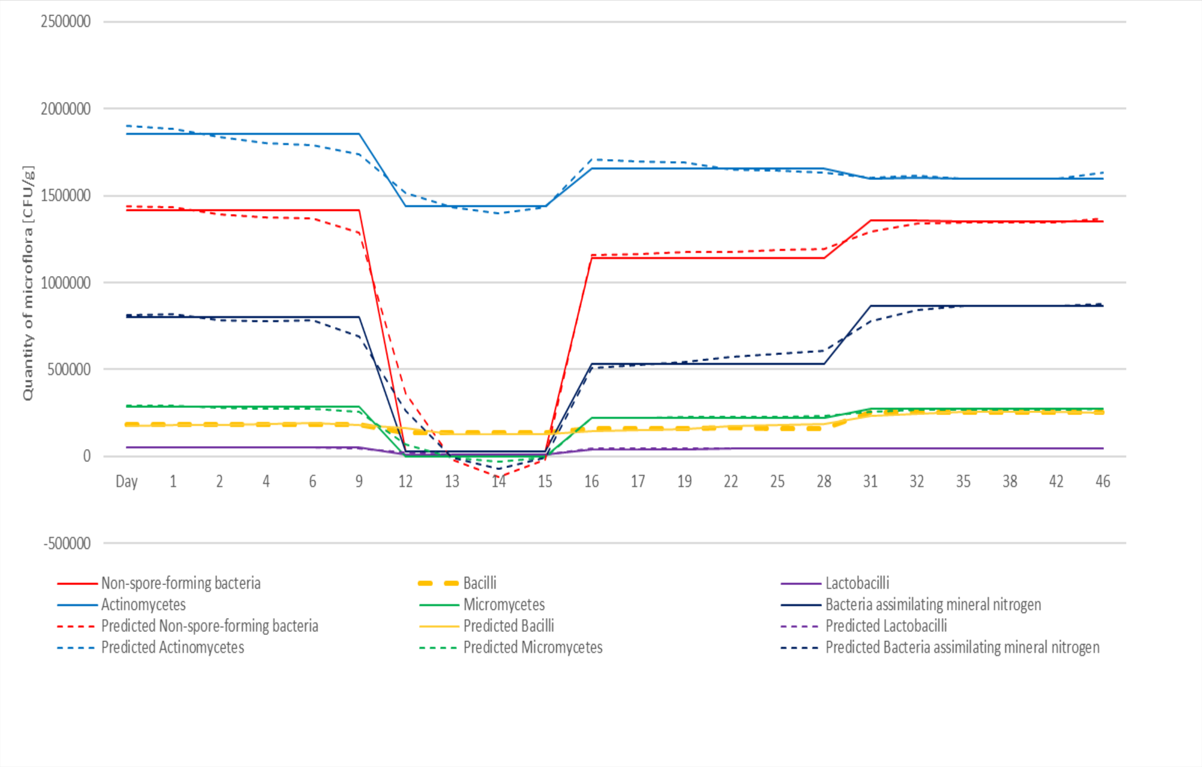


Figure 1 Graphical results for experimental microbiota quantities and those obtained by regression model for composting without additive

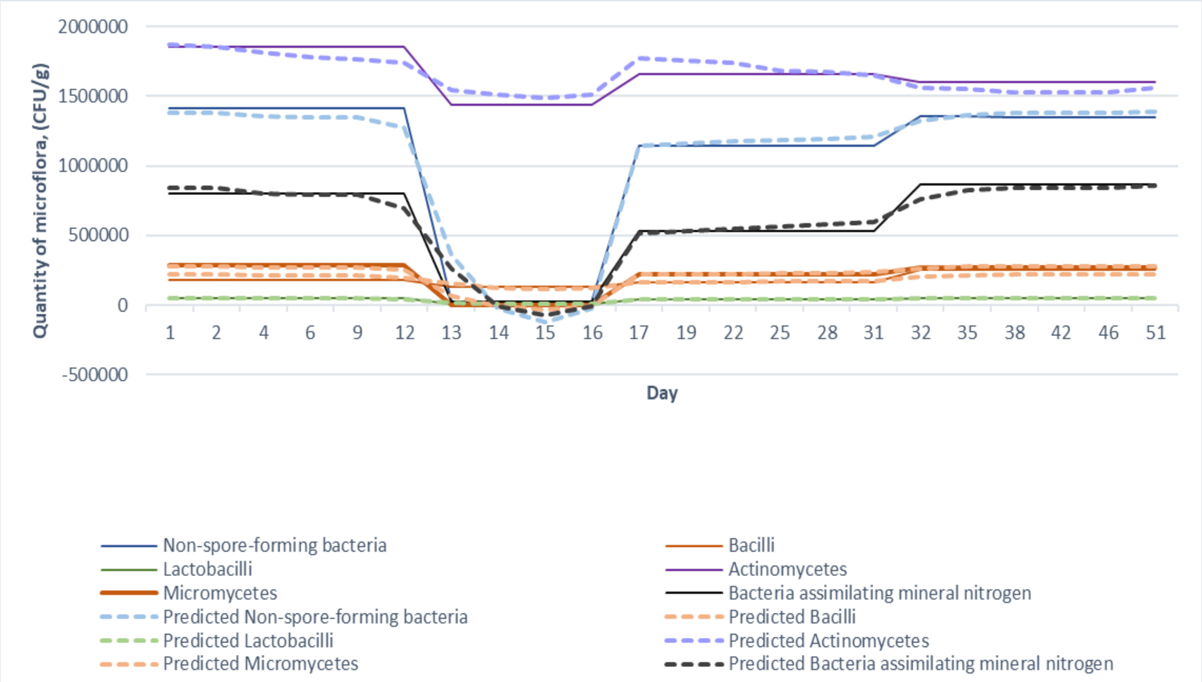
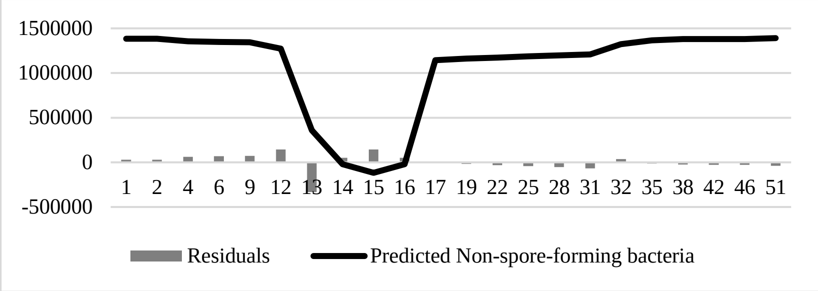
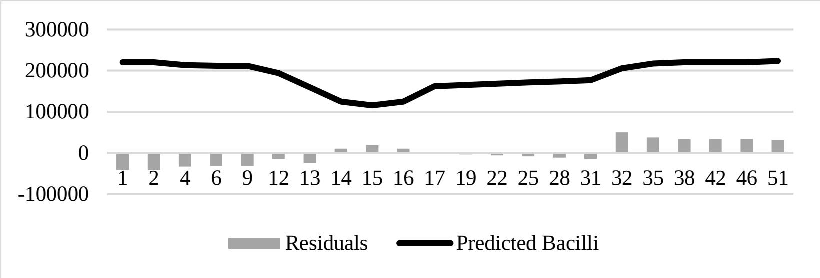
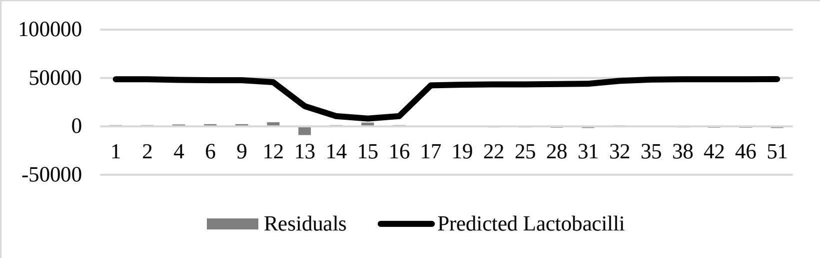
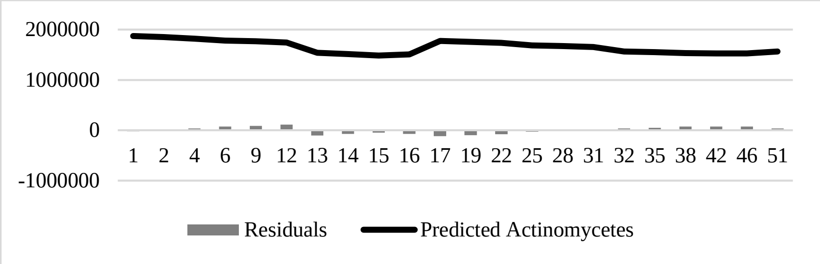
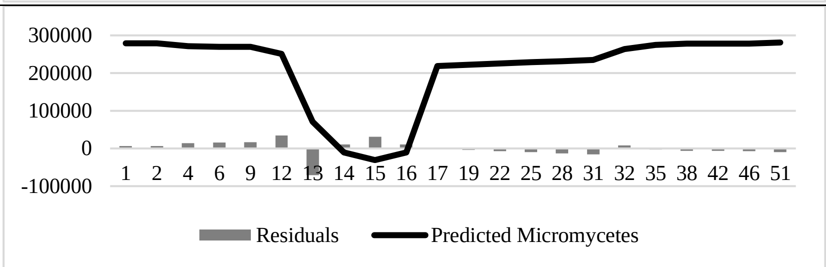
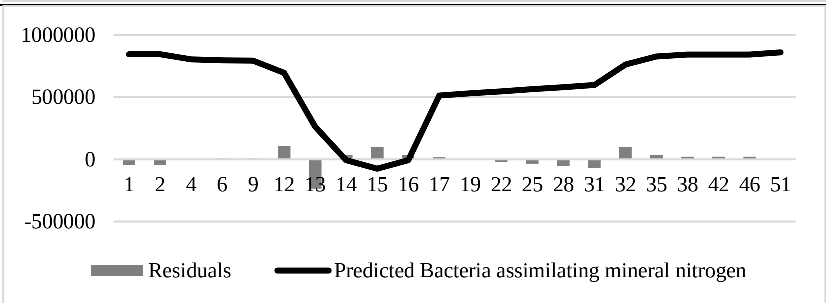


Figure 2 Graphical results for experimental amounts of microflora and those obtained by regression model for composting without additive with adjustment in the independent factors included

The final regression equations have the form shown in Table 4, where the corresponding graphical results are also presented. In addition, the residuals can be seen on the graphs when the values are calculated by the regression model.

Table 4 Regression models for composting without additive

Microorganisms / Regression equation	Graphical results from the regression model
Non-spore-forming bacteria $Y = -11562*x_1-20286*x_2+1793516$	
Bacilli $Y = -2913*x_1+290542$	
Lactobacilli $Y = -298*x_1-558*x_2+59516$	
Actinomycetes $Y=17399*x_1+1195883$	
Micromycetes $Y=-2944*x_1-3811*x_2+374520$	
Bacteria assimilating mineral nitrogen $Y=-16503*x_1-6103*x_2+1281214$	

where x_1 - temperature, x_2 – pH, x_3 - humidity, Y – the corresponding amount of microflora
In the graphs: horizontal axis - consecutive day of composting, vertical axis - the quantity of microflora (CFU/g)

Statistical analyses in composting with additive

Similar calculations were made for composting with additive.

Table 5 Correlation coefficients - compost with additive

Indicators	Non-spore-forming bacteria	Bacilli	Lacto-bacilli	Actino-mycetes	Micro-mycetes	Bacteria assimilating mineral nitrogen
Temperature (°C) - average	-0,886	-0,689	-0,941	-0,684	-0,941	-0,940
pH	-0,917	-0,348	-0,886	-0,629	-0,879	-0,890
Humidity (%)	-0,803	-0,771	-0,870	-0,236	-0,891	-0,851

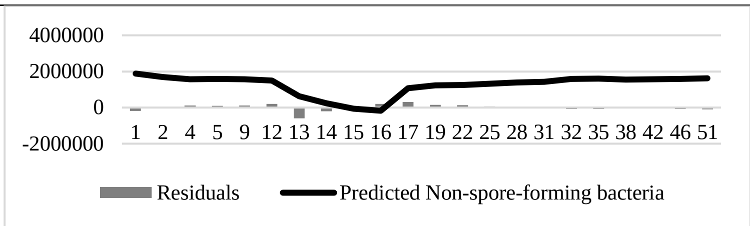
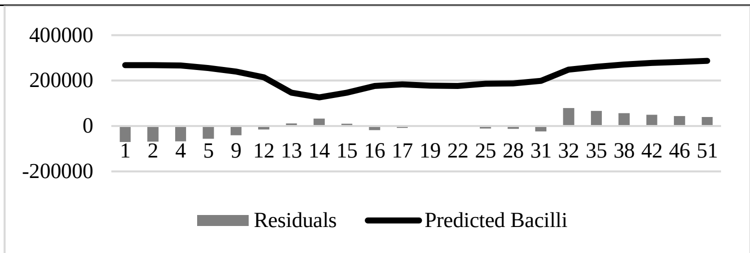
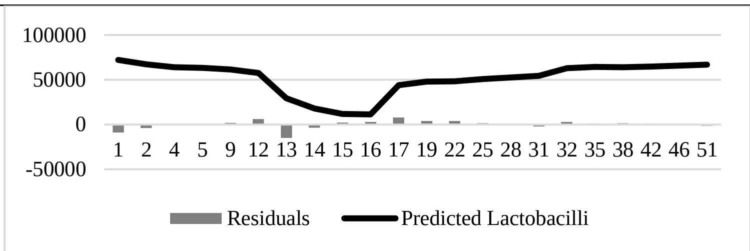
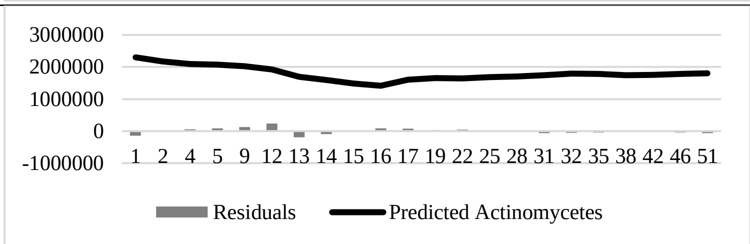
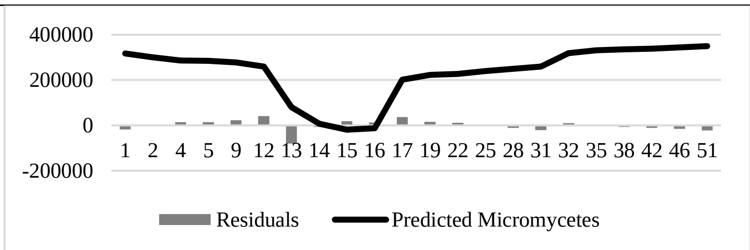
Table 6: Values for R² and F when creating regression models of microflora in composting with additive

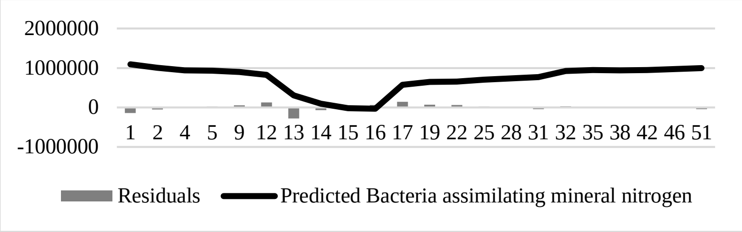
Indicators	Non-spore-forming bacteria	Bacilli	Lactobacilli	Actino-mycetes	Micro-mycetes	Bacteria assimilating mineral nitrogen
R^2	0,904911135	0,835712723	0,949036347	0,850361576	0,955724789	0,942861444
F	57,09887076	30,52139163	111,7309652	34,09665313	129,5160113	99,00790345
Pr > F	<0,0001	<0,0001	<0,0001	<0,0001	<0,0001	<0,0001

When composting with an additive, R^2 values are slightly lower than when composting without an additive.

The change in the amount of microflora and the corresponding dependencies obtained by regression analysis can be seen in Fig. 3, and the regression equations themselves are presented in Table 7.

Table 7 Regression models for additive composting

Microorganisms / Regression equation	Graphical results from the regression model
Non-spore-forming bacteria $Y = -19826*x_1-857421*x_2+7981248$	
Bacilli $Y = -5377*x_1+60560*x_2$	
Lactobacilli $Y = -968*x_1-16963*x_2+206701$	
Actinomycetes $Y=-21058*x_1-320602*x_2+19903*x_3+4096800$	
Micromycetes $Y=-4586*x_1-75332*x_2+-2774*x_3+1025404$	

Bacteria assimilating mineral nitrogen $Y=-17454*x_1-322468*x_2+3626541$	

where x_1 - temperature, x_2 – pH, x_3 - humidity, Y – the corresponding amount of microflora
In the graphs: horizontal axis - consecutive day of composting, vertical axis - the quantity of microflora (CFU/g)

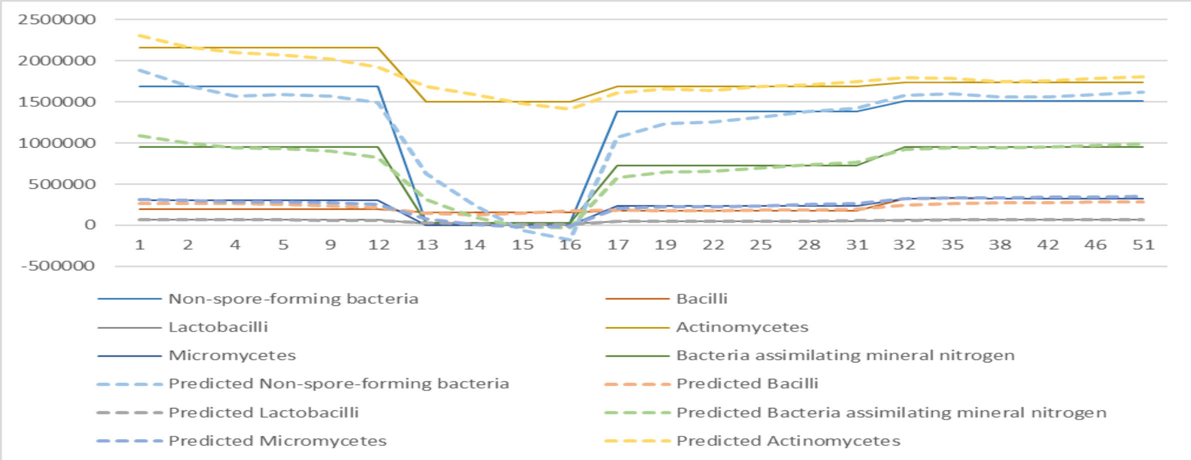


Figure 3 Graphical results for experimental microflora quantities and those obtained by regression model for composting with additive

A further comparison between the mathematical models in the two cases of compost with and without additives and the different composting phases can be made based on the residuals obtained in the regression analysis. The larger the residual, the greater the difference between the experimental data and those obtained by the model.

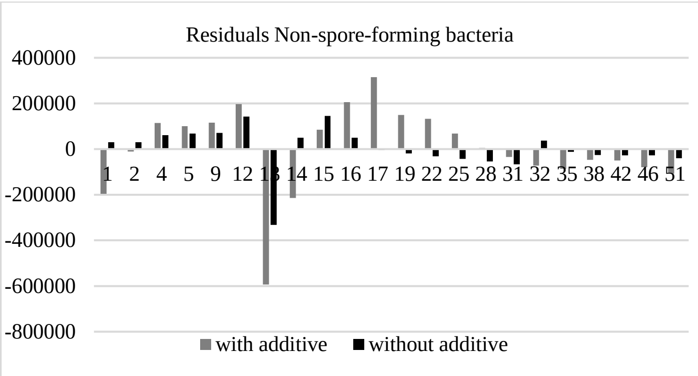


Figure 4. Comparison of residuals when creating a regression model of the quantity of non-spore-forming bacteria

For non-spore-forming bacteria, a more accurate match between the model and experimental data was observed after day 25 (during the aging phase) for both composting treatments, with and without additive (Fig. 4).

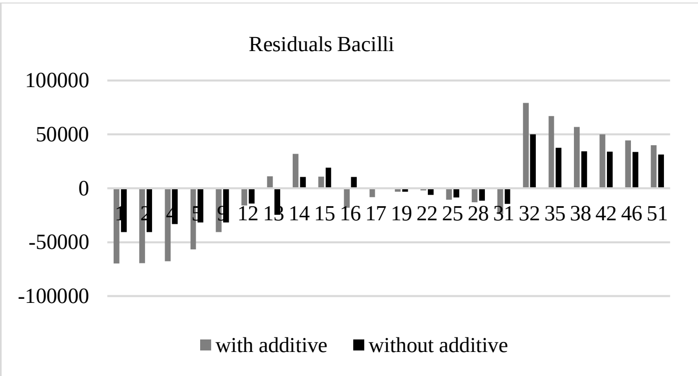


Figure 5 Comparison of residuals when creating a regression model of bacilli quantity

The regression model for bacilli was more similar to the experimental data between days 12 and 31 (Fig. 5). There the residuals are smaller for both composting treatments.

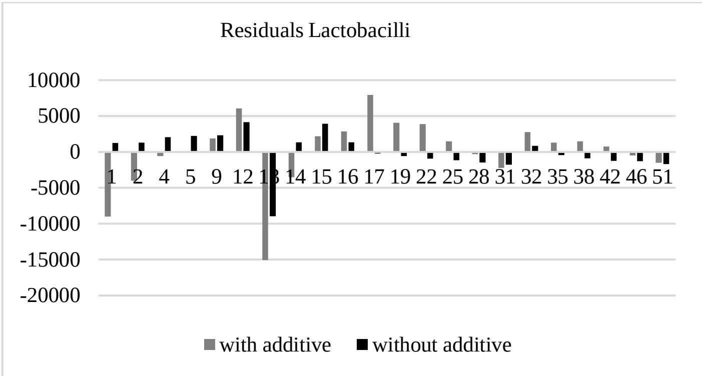


Figure 6 Comparison of residuals when creating a regression model of the quantity of lactobacilli

For lactobacilli, there was generally no significant difference between the different phases, only around day 13 the residues in both models had higher values (Fig. 6). Day 13 is the beginning of the thermophilic phase. Such an increase is also present in most groups of microorganisms, with the exception of bacilli. The reason for this is the relatively abrupt change in temperature during this phase, which also leads to an abrupt change in the abundances of individual species. In modelling, this change may not always be adequately reflected in the mathematical model.

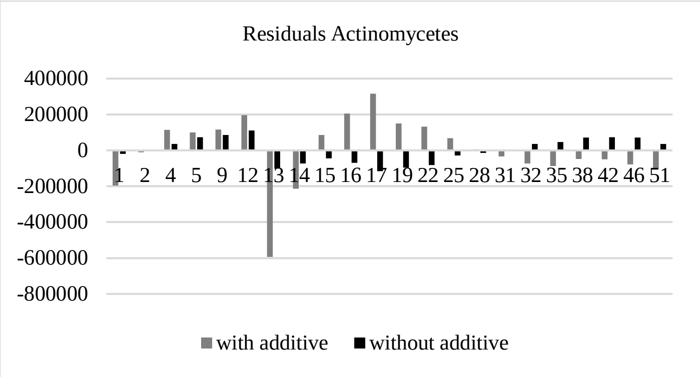


Figure 7 Comparison of residuals in creating a regression model of actinomycetes abundance

An interesting finding was that the actinomycete residuals had different sign in the two composting treatments, but were similar in absolute value for the period after day 14 to the end of observation (Fig. 7). There is also a similarity in values for micromycetes and mineral nitrogen uptake bacteria (Fig. 8 and Fig. 9). In general, the residue value was slightly lower in the case of composting without additive. These findings are consistent with the values of R^2 and Fisher's criterion calculated and presented above.

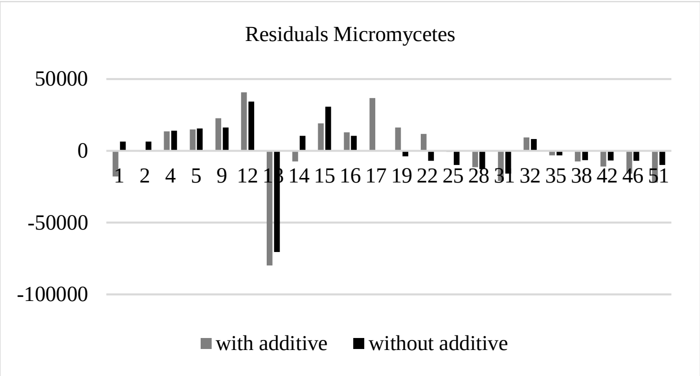


Figure 8 Comparison of residuals when creating a regression model of the quantity of micromycetes

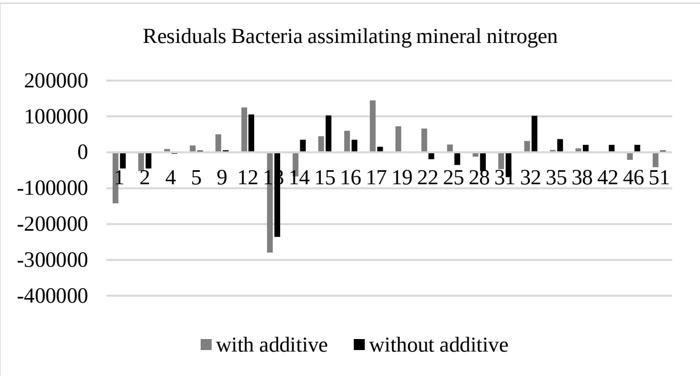


Figure 9 Comparison of residuals when creating a regression model of the amount of bacteria assimilating mineral nitrogen

Often when composting we are interested in the total amount of microflora. It is obtained as the sum of the amounts of the respective species for which regression models were created. Figure 10 shows a graphical comparison between the amount of microflora in composting with and without additive and the values obtained by regression analysis.

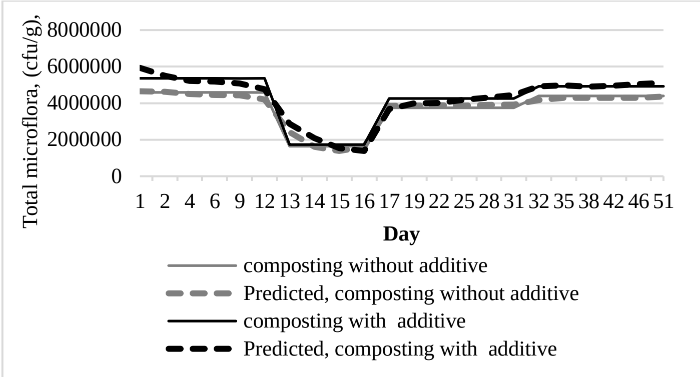


Figure 10 Total microflora during composting with and without additive - comparison of experimental data and those obtained by regression model

When composting with additive, the amount of microflora is slightly higher than when composting without additive. In the thermophilic phase the difference is not significant. In terms of the data obtained by regression model, both cases are similar to the experimental data.

Mineralization coefficient during composting

The mineralization coefficient is also an important indicator when examining the microflora in composts. It depends on the amount of non-spore-forming bacteria, bacilli, lactobacilli and mineral nitrogen-fixing bacteria as described above.
The values of the mineralization coefficients calculated from the experiments and those obtained from the regression models are presented graphically in Figure 11. Here in the thermophilic phase there is a significant discrepancy in model and experimental data. In the other composting phases there is a complete agreement between the model and experimental values for both composting types.

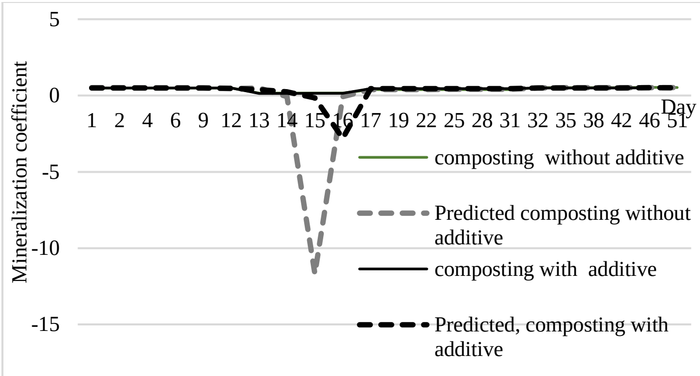


Figure 11. Mineralization coefficients for composting with and without additive

CONCLUSION

The biogenicity of the compost variants is enhanced by the addition of a microbial additive. There was a regrouping in the composition of the total microflora in the composting process compared to previous studies, with actinomycetes occupying the major proportion, followed by non-spore-forming bacteria, except in the thermophilic phase, where the proportion of spore-forming bacteria was higher and mould fungi completely disappeared. The rate of mineralisation of organic matter in compost mixtures is higher in the maturation and mesophilic phases and significantly lower in the thermophilic phase.
Using classical statistical methods, mathematical models describing the amount of different groups of microorganisms in the compost were created. The resulting regression equations make it possible to relatively predict the corresponding quantities from the values of factors such as temperature, humidity and pH, with the same composition of compost variants and method of composting. The values of the factors can be easily measured with standard measuring equipment. The regression relationships thus obtained can be used in future experiments to establish approximately the quantity of different groups of microorganisms from indirect measurements, in compost variants with the presented recipes and method of composting.
Statistical analysis showed varying degrees of dependence of different groups of microorganisms on the selected factors. The influence of temperature is strongest. It is included in all regression equations. Humidity has the weakest influence. It is interesting to note that in the case of composting with additives the influence of the pH of the medium is slightly higher than in the case of composting without additives.
Despite the good coincidence between the microflora quantities calculated by the regression models and those measured experimentally in the calculation of the

mineralization coefficient, a significant difference is the thermophilic phase, especially in the case of composting without additive. The reason here is mainly the accumulation of errors in each of the mathematical models. Several types of microflora are involved in the calculation of the mineralization coefficient and the errors in the description of each are superimposed. Further experiments and accumulation of a larger amount of data could be conducted to solve this problem, from which more accurate mathematical modelling could be done and possibly additional factors incorporated. The goal of the study, however, is to create a good mathematical description by a minimum number of factors.

Acknowledgments: The analyzes were carried out thanks to the support of the national project KP-06-H66/10/15.12.2022 "Investigation of interrelationships in different variations of composting organic waste, by creating dynamic compost mixtures with the addition of ameliorants and tracking microbiological and enzyme activity – options for applying a quality compost-based organo-mineral improver in agricultural practice", financed by the Scientific Research Fund.

REFERENCES

Billington, F. H. (1956). *Compost for garden plot or thousand-acre farm: A Practical Guide to Modern Methods*. London, England: Faber and Faber, 140 p.

Ceglie, F.G., Abdelrahman, H.M. (2014). Ecological intensification through nutrients recycling and composting in organic farming. In D. K. Maheshwari (Ed), *Composting for Sustainable Agriculture* (1-22). Springer, Cham. https://doi.org/10.1007/978-3-319-08004-8_1

Chanal, A., Chapon, V., & Benzerara, K. (2006). The desert of Tataouine: an extreme environment that hosts a wide diversity of microorganisms and radiotolerant bacteria. *Environmental Microbiology*, *8*(3), 514-525. https://doi.org/10.1111/j.1462-2920.2005.00921.x

Chang, J. I., Tsai, J. J., & Wu, K. H. (2006). Composting of vegetable waste. *Waste management & research*, *24*(4), 354-362. https://doi.org/10.1177/0734242X06065727

Chikae, M., Ikeda, R., Kerman, K., Morita, Y., & Tamiya, E. (2006). Estimation of maturity of compost from food wastes and agro-residues by multiple regression analysis. *Bioresource Technology*, *97*(16), 1979-1985. https://doi.org/10.1016/j.biortech.2005.09.026

Cooperband, L. R. (2000). Composting: Art and science of organic waste conversion to a valuable soil resource. *Laboratory medicine*, *31*, 283-290. https://doi.org/10.1309/W286-LQF1-R2M2-1WNT

De Gannes, V., Eudoxie, G., & Hickey, W. J. (2013). Prokaryotic successions and diversity in composts as revealed by 454-pyrosequencing. *Bioresource Technology*, 573–580. https://doi.org/10.1016/j.biortech.2013.01.138

Diaz, L. F., De Bertoldi, M. (2007). History of composting. In L.F. Diaz, M. de Bertoldi, W. Bidlingmaier, E. Stentiford (Eds), *Waste Management* (vol. 8, 7-24). Elsevier. https://doi.org/10.1016/s1478-7482(07)80005-4

Draper, N. R., & Smith, H. (1998). *Applied regression analysis*, vol. 326 (3rd ed.). John Wiley & Sons, USA. https://doi.org/10.1002/9781118625590

Gogtay, N. J., & Thatte, U. M. (2017). Principles of correlation analysis. *Journal of the Association of Physicians of India*, *65*(3), 78-81. PMID: 28462548.

Gotaas, Harold Benedict & World Health Organization. (1956). Composting : sanitary disposal and reclamation of organic wastes / Harold B. Gotaas. World Health Organization. https://iris.who.int/handle/10665/41665

Graves, R. E., Hattemer, G. M. (2000). Chapter 2 Composting. Part 637 Environmental Engineering National Engineering Handbook. United States Department of Agriculture. Natural Resources Conservation Service. (210-VI-NEH). Available at (https://irrigationtoolbox.com/NEH/Part637_EnvironmentalEngineering/H_210_637_02.pdf) (Accessed on 24/11/19).

Grigorova-Pesheva, B., Malcheva, B., Hristov, B. (2023). Microbial abundance in compost mixtures in the final phase of composting. Scientific conference with international participation "Ecology and Agrotechnologies - fundamental science and practical implementation", December 5, 2023, Sofia, Institute of Soil Science, Agrotechnologies and Plant Protection "Nikola Pushkarov", 213-217. https://www.issapp-pushkarov.org/sites/default/files/pictures/ecology_and_agrotechnologies_vol_5.pdf

Grigorova-Pesheva, B. (2019). Dynamics of Microflora in the Process of Composting Organic Waste and Bioplastics. 13th International Conference on Microbial Interactions & Microbial Ecologyp July 19-20, 2018, Rome, Italy, 986-993. https://doi.org/10.4172/2471-9315-C2-014

Grigorova-Pesheva, B., Malcheva, B. (2023). Composting of biodegradable plastic waste - changes in the microbial community. SGEM VIENNA GREEN CONFERENCE, December 2023, 21-28. https://doi.org/10.5593/sgem2023V/4.2/s18.03

Kango, N., Uttam, J., & Ritumbhara, C. (2019) Fungal Enzymes: Sources and Biotechnological Applications. In T. Satyanarayana, S. K. Deshmurkh, M. V. Deshpande (Eds), *Advancing Frontiers in Mycology & Mycotechnology*, 515–538. Springer. https://doi.org/10.1007/978-981-13-9349-5_21

Koleva, R., Malcheva, B., Grigorova-Pesheva, B. (2024). Influence of microflora in irrigation water on the soil and soil-compost microbiome. The International Conference of the University of Agronomic Science and Veterinary Medicine of Bucharest, 2024, Bucharest, Romania. Scientific papers–series A–Agronomy, Vol. LXVII, No. 1, 121-128.

Konsulova, M., Naskova, P., Plamenov, D., Malcheva, B. (2017). Recognition and probability of migrant microbiological activity by indirect signature. *Soil Science Agrochemisty and Ecology*, *51*, 3-4, 12-20 (Bg).

Malcheva, B., Naskova, P. (2018). *Guide for laboratory exercises of microbiology*. Varna, Bulgaria: University Publishing House at TU-Varna, 70 p. (in Bulgarian).

Malcheva, B., Naskova, P., Plamenov, D., Iliev, Y. (2018a). Study on impact of mineral fertilizers on biogenity and enzymatic activity of soils with common wheat. *Int. J. Adv. Res.*, *6*(12), 137-144. http://dx.doi.org/10.21474/IJAR01/8124

Malcheva, B., Yordanova, M., Borisov, R., Vicheva, T., Nustorova, M. (2018b). Dynamics of microbiological indicators for comparative study of compost variants. The International Conference of the University of Agronomic Science and Veterinary Medicine of Bucharest, June 7-9 2018, Bucharest, Romania. Scientific papers-series B-Horticulture, vol. 62, 649-654. https://horticulturejournal.usamv.ro/pdf/2018/vol2018.pdf

Malcheva, B., Naskova, P., Plamenov, D. (2019a). Investigation of the influence of mineral nitrogen fertilizers on the microbiological and enzymic activity of soils with rapeseed. *New Knowledge Journal of Science*, *8-4*, 80-90 (Bg). https://www.science.uard.bg/index.php/newknowledge/article/download/570/pdf_184

Malcheva, B., Yordanova, M., Nustorova, M. (2019b). Influence of composting on the microbiological activity of the soil. The International Conference of the University of Agronomic Science and Veterinary Medicine of Bucharest, June 6-8 2019, Bucharest, Romania. Scientific papers-series B-Horticulture. Vol. LXIII, No. 1, 621-625.

Malcheva, B., Yordanova, M., Petrova, V., Naskova, P., Plamenov, D. (2020). Microbial and enzymatic degradation of carbohydrates: a comparative investigation of compost variants. International Conference of the University of Agronomic Science and Veterinary Medicine of Bucharest „Agriculture for life, life for agriculture", June 4-6 2020, Bucharest, Romania. Scientific papers-series B-Horticulture, vol. LXIV, №1, 425-433.

Malcheva, B. (2021). Influence of microbial fertilizers on microbiological and enzymatic decomposition of carbohydrates in agrogenic soil. *Bulgarian Journal of Crop Science*, *58*(3) 95-103 (Bg). https://cropscience-bg.org/page/bg/details.php?article_id=925

Mishustin, E., & Runov, E. (1957). The success of the development of the principles of microbiological diagnosis of soil condition Advances in Modern Biology, 44, 256-268 (in Russian).

Mishustin, F., Emtsev, N. (1989). *Microbiology*. Moscow: Kolos, 367 p. (in Russian).

Naskova, P., Malcheva, B., Yankova, P., Plamenov, D. (2015). Some chemical and microbiological indexes at soils after a flood in the region of Varna, Bulgaria. *International Journal of Research Studies in Science, Engineering and Technology*, *2*(10), 62-71. https://ijrsset.org/pdfs/v2-i10/10.pdf

Naskova, P., Malcheva, B., Yankova, P., Plamenov, D. (2016). Impact of the biological fertilizers on chemical indexes and enzyme activities of soils at cucumbers. *International Research Journal of Natural and Applied Sciences*, *3*(11), 120-131. https://www.researchgate.net/publication/311793643_IMPACT_OF_THE_BIOLOGICAL_FERTILIZERS_ON_CHEMICAL_INDEXES_AND_ENZYME_ACTIVITIES_OF_SOILS_AT_CUCUMBERS

Naskova, P., Konsulova, M., Plamenov, D., & Malcheva, B. (2017). Mathematical model for determining the influence of different physico-chemical factors on the number of microflora in the anthropogenic soils. *New Knowledge Journal of Science*, 6-4, 88-102 (Bg). https://science.uard.bg/index.php/newknowledge/article/download/309/pdf_38

Naskova, P., Malcheva, B., Plamenov, D., Ivanova, A. (2024). Brief overview of microbiological aspects of composting. *Bulgarian Journal of Crop Science*, *61*(1), 68-74. (Bg). https://doi.org/10.61308/QHWT7218

Nozhevnikova, A. N., Mironov, V. V., Botchkova, E. A., Litti, Y. V., & Russkova, Y. I. (2019). Composition of a microbial community at different stages of composting and the prospects for compost production from municipal organic waste. *Applied Biochemistry and Microbiology*, *55*, 199-208. https://doi.org/10.1134/S0003683819030104

Nustorova, M., Malcheva, B. (2020). *Guide for laboratory exercises of microbiology*. Varna, Bulgaria: University Publishing House at TU-Varna, 118 p. (in Bulgarian).

Plamenov, D., Naskova, P., Malcheva, B., Iliev, Y. (2016). Chemical and microbiological studies for determination the influence of fertilizers produced by "Agropolychim" AD on winter common wheat and oilseed rape. *International Journal of Science and Research*, *5*(5), 1481-1486. https://www.researchgate.net/publication/325443155_Chemical_and_Microbiological_Studies_for_Determination_the_Influence_of_Fertilizers_Produced_by_Agropolychim_Ad_on_Winter_Common_Wheat_and_Oilseed_Rape

Sánchez, Ó. J., Ospina, D. A., & Montoya, S. (2017). Compost supplementation with nutrients and microorganisms in composting process. *Waste Management*, *69*, 136–153. https://doi.org/10.1016/j.wasman.2017.08.012

Steel, H., & Wim, B. (2012). Biodiversity of compost mesofauna and its potential as an indicator of the composting process status. *Dynamic soil dynamic plant.*, *5*(spec. iss. 2), 45-50. https://www.researchgate.net/publication/294291420_Biodiversity_of_compost_mesofauna_and_its_potential_as_an_indicator_of_the_composting_process_status

Tuomela, M., Vikman, M., Hatakka, A., & Itävaara, M. (2000). Biodegradation of lignin in a compost environment: a review. *Bioresource Technology*, 72(2), 169–183. https://doi.org/10.1016/S0960-8524(99)00104-2

Yankova, P., Naskova, P., Malcheva, B., Plamenov, D. (2016). Impact of the biological fertilizers on the microorganisms and the nutrient elements in the soil. *International Journal of Current Research*, *5*(5), 39681-39686. https://www.journalcra.com/article/impact-biological-fertilizers-microorganisms-and-nutrient-elements-soil

Yildiz, S., & Degirmenci, M. (2015). Estimation of oxygen exchange during treatment sludge composting through multiple regression and artificial neural networks (estimation of oxygen exchange during composting). *International Journal of Environmental Research*, *9*(4), 1173-1182. https://doi.org/10.22059/ijer.2015.1007

Yordanova, M. (2020). *Composting of vegetable waste*. Sofia, Bulgaria: Intel Entrans, 142 p. (in Bulgarian).