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Analysis of changes in soil physical properties and CO_2 emissions under the influence of biopreparations of different composition

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Abstract. The introduction of innovative technologies in agriculture is key not only to improving the efficiency of agricultural production, crop yields, and quality but also to balancing energy use and preserving a cleaner environment. Biopreparations are environmentally friendly means of restoring the vitality of the soil on which plants can thrive. Biopreparations have an impact on soil health and alter greenhouse gas emissions. The aim of this work was to investigate the effects of different biopreparation formulations on soil physical properties and CO_2 emissions from the soil by establishing correlations. The experimental studies were carried out over three years, and each spring, after the resumption of winter crops, the soil surface was sprayed with biopreparations of different properties or mixtures of biopreparations, under 7 scenarios, with one scenario left as a control. Soil porosity, temperature, and CO_2 emissions from the soil were measured regularly every month from April to August. The application of the biopreparations showed a cumulative effect on the soil properties. In the third year of the study, the total porosity of the soil was higher in all scenarios compared to the control, ranging between 51% and 74%. The aeration porosity of the soil was also higher in all years of the study than in the control, although no significant differences were obtained. The results of the studies on CO_2 emissions from the soil showed that in the first year, the application of the biopreparations increases emissions compared to the control. However, when assessing the cumulative effect of the biopreparations on soil respiration intensity, it was found that in the third year, most of the biopreparations led to a reduction in CO_2 emissions compared to the control. The lowest emissions were achieved with the biopreparations consisting of essential oils of plants, 40 species of various herbs extracts, marine algae extracts, A*zospirillum sp.*, *Frateuria aurentia, Bacillus megaterium*, mineral oils, A*zotobacter vinelandi*, humic acid, gibberellic acid, sodium molybdate, azototbacter chroococcum, azospirillum brasilense, etc.

Keywords: GHG emissions, carbon dioxide, bioproducts, soil porosity, soil temperature

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Introduction

1.1 Importance of biopreparations

Decades of soil degradation have led to a search for ways to contribute to soil sustainability by preserving soil properties without harming the environment. Over the last decade, European agricultural policy has increasingly turned towards environmental sustainability, with the aim of reducing the use of chemicals and increasing the organic area (European Commission, 2020). An increasing number of agricultural operators and farmers have adopted environmentally friendly biotechnologies that use biopreparations, i.e., bioproducts designed to inhibit the growth of pathogenic fungi or bacteria, stimulate plant growth, improve plant nutrient uptake, and restore soil properties and fertility (Michalak et al., 2016; Trevisan et al., 2019; Szparaga et al., 2019). Consumers have started to increasingly value agricultural products with high nutritional and functional value and environmentally sustainable production (Caruso et al., 2019; Szparaga et al., 2018). Therefore, bioproducts used in agricultural practice aim to enhance the biological protection of plants by reducing the spread of pathogens and pests, increase crop productivity, improve soil microbiology, change the physical and chemical properties of soil, reduce environmental pollution, and weaken the properties of crop residues (Khattab et al., 2009; Vaitauskiene et al., 2015; Oskiera et al., 2017; Naujokienė et al., 2018). Blaszczys et al. (2014) stated that *Trichoderma harzianum* and *Trichoderma atroviride* are common components of biopreparations used in agriculture. Fungi of the genus *Trichoderma* can effectively reduce phytopathogens in agricultural soils through various mechanisms (Oskiera et al., 2017). A combination of edaphic and dynamic factors, including crop rotation, residue management, soil type, tillage, and climate, affect the microorganism community (Bünemann et al., 2008; Gil et al., 2011; Zhang et al., 2014). A growing body of research demonstrates that plant-derived phytochemicals affect the soil microbiota through interactions between plant roots and soil (Bais et al., 2006; Kong et at., 2008; Lorenzo et al., 2013). Biopreparations have multiple effects, but scientists are placing more emphasis on their positive effects on plants and soil (Tarantino et al. 2018). Biopreparations are also used as seed diluents to increase germination and reduce seed contamination with pathogenic microorganisms (Selby et al., 2016; Rouphael et al., 2018). Kocira et al. (2020) report that the mixtures of seeds and biopreparations obtained from *Archangelica officinalis L.* significantly inhibit fungal development on the seed surface. Biopreparations have antimicrobial activity because they contain biologically active substances that can inhibit the development of microorganisms. The appropriate composition of the biopreparations to be used depends mainly on the plant species (Nostro et al., 2000; Sen and Batra, 2012; Shihabudeen et al., 2010). The use of biopreparations can reduce the cost of crop production and increase the efficiency of soil nutrient use by reducing the incidence of diseases caused by nutrient deficiencies. However, this effect is not easy to achieve, as it requires a lot of knowledge on the proper selection of biopreparations, their application method, and the correct adjustment of the amount and concentration (Ertani et al., 2018; Szparaga et al., 2019; Michałek et al., 2018).

1.2. Effects of biopreparations on soil

Soil microorganisms are an essential link in the nutrient cycle in the soil and maintain soil fertility. Their activity determines the physical and chemical properties of the soil, and these properties in turn determine how the microorganisms feel in the soil. Soil physical properties such as porosity and temperature are constantly changing under the influence of the environment. A research team from Poland investigating the influence of microorganisms on soil density and porosity found no significant changes over 5 years (Pranagal et al., 2020). Other researchers (Montemurro et al., 2010; Peltre et al., 2015; Juknevičius et al., 2020) have suggested that biopreparations increased the organic carbon content of the soil, which presumably led to a decrease in soil density and an increase in overall porosity. Researchers have pointed out that soil water content influences soil density (Lu et al., 2018; Tian et al., 2018; Tong et al., 2020). Naujokienė et al. (2018) reported that the use of differently prepared biopreparations reduced soil hardness by up to 28% and increased total porosity by up to 25% in the second year of the study, which resulted in lower diesel fuel consumption and reduced GHG emissions to the environment.

1.3. CO_2 emissions from soil

The agricultural sector is one of the most important GHG polluters of the environment, and cleaner production processes in this sector are of particular interest (Hamzei and Seyyedi, 2016; Wu et al., 2017). CO_2 emissions from soil are the second largest component of the carbon cycle and contribute to climate change (Mohammed et al., 2022). Agricultural producers are encouraged to increase agricultural production by developing alternative technologies that address climate change, i.e., reducing the carbon footprint of agriculture (Dias et al., 2016; Foley et al., 2011; Tilman et al., 2002). Soil bioactivity is the set of biological processes that determine soil respiration, enzyme activity, humification, and mineralization processes. A group of researchers (Ma et al., 2021) has observed that microorganism structure (community structure) and soil properties change together depending on environmental conditions and determine the dynamics of GHG emissions. Stimulating soil microorganisms increases CO_2 release and improves nutrient mobilization (Klenz, 2015). Scientific results showed that the preparation of biocrusts biopreparation significantly improved soil physicochemical properties, respiration, and alkaline phosphatase, protease, and cellulose, and reduced CO_2 emissions in vegetation areas (Liu et al., 2017). A team of researchers (Juknevičius et al., 2018) found that biopreparations increased soil organic carbon by up to 0.2%, which can have an impact on soil respiration.

The dependence of soil respiration intensity, GHG emissions, and physical soil properties on tillage and other technological operations has already been studied quite extensively. However, the impact of environmentally friendly biopreparations on soil physical properties and the dynamics of CO_2 emissions during the growing season has not yet been sufficiently studied. The limited number of scientific papers on this topic shows that research on the effects of biopreparations on soil under different meteorological conditions is new and relevant. The aim of this study was to investigate the effects of different biopreparation formulations on soil porosity, temperature, and CO_2 emission from the soil by establishing correlations.

2. Material and methods

2.1. Site description and experimental design

Experimental field research was carried out in 2014–2017 at the Experimental Station of Vytautas Magnus University Agriculture Academy (54º534’N, 23º50’E) in ųgleyic satiated planosoil (*Endohypogleyic-Eutric Planosol – PLe-gln-w*) (Buivydaitė and Motuzas, 2001). The experimental station is in central Lithuania, on the left bank of river Nemunas, in Kaunas district.

In the first year of the study, winter wheat (variety “Ada”) was grown, in the second year – winter wheat (“Famulus”) was grown, and in the third year – winter oilseed rape (“Cult”) was grown. Eight scenarios (SC) were selected to determine the effect of biopreparations on soil properties and CO_2 emissions from the soil, of which SC1 was the control with no biopreparations used. In the other seven SCs, biopreparations or mixtures of biopreparations were used. The components of the bio-preparations are given in Table 1. The biopreparations were applied after the resumption of winter crops in the second half of April. The experimental plots were laid out in a linear pattern. The initial size of the plots was 600 m^2 and the reference size was 400 m^2. The layout of the experimental field scenarios is presented in Figure 1.

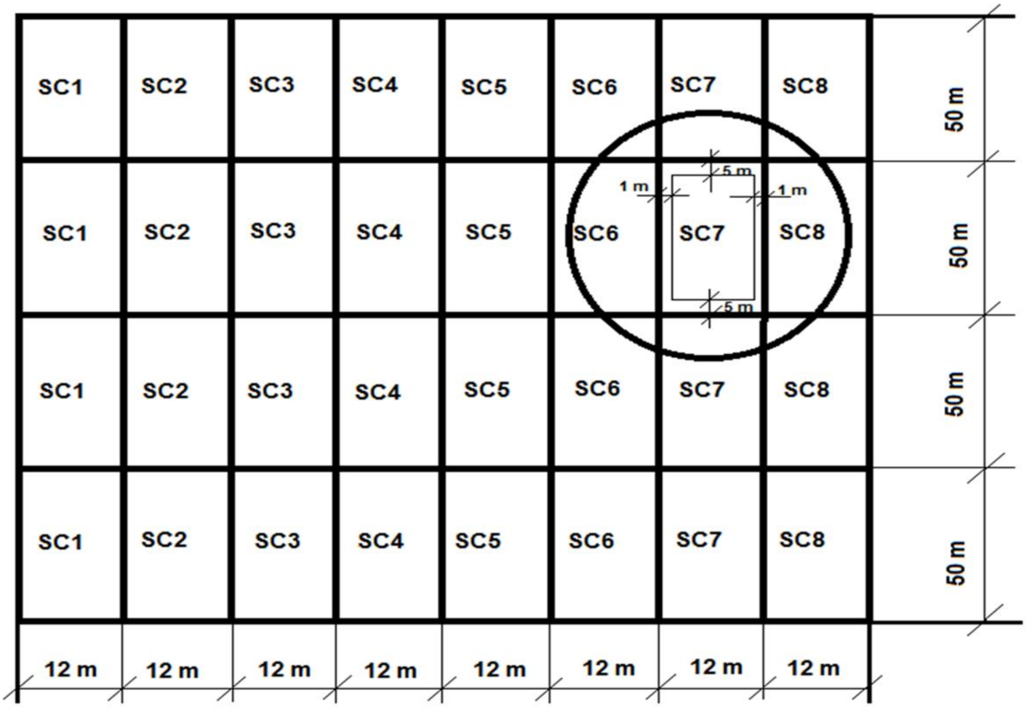


Fig. 1. Scheme of experimental field study scenarios

Table 1. Composition of the biopreparations used in different scenarios

The composition of biopreparations	Scenario							
	SC1	SC2	SC3	SC4	SC5	SC6	SC7	SC8
40 species of various herbs	-	+	+	-	+	-	+	-
Marine algae extracts	-	+	+	+	+	-	+	+
Essential oils of plants		+	+		+		+	
Mineral oils	-	+	+		+	-	+	-
Azospirilum spp.	-	-	+	+	-	-	-	-
Bacillus magetarium	-	-	+	+	-	-	-	-
Frateuria autentia	-	-	+	+	-	-	-	-
Azotobacter chroococcum	-	-	-	-	+	-	-	+
Azotospirilum brasilense	-	-	-	-	+	-	-	+
4.5% of humic acids	-	-	-	-	-	+	+	-
0.5% gibberellic acid	-	-	-	-	-	+	+	-
0.01% copper (Cu)	-	-	-	-	-	+	+	-
0.01% zinc (Zn)	-	-	-	-	-	+	+	-
0.01% manganese (Mn)	-	-	-	-	-	+	+	-
0.01% iron (Fe)	-	-	-	-	-	+	+	-
0.01% calcium (Ca)	-	-	-	-	-	+	+	-
0.005% sodium molybdate (Na_2MoO_4)	-	-	-	-	-	+	+	-
Phosphorus P (P_2O_5)	-	-	-	-	+	-	-	+
Potassium K(K_2O)	-	-	-	-	+	-	-	+
Azotobacter spp.	-	-	-	-	-	+	+	-
Water (H_2O)	+	+	+	+	+	+	+	+

"+" – a compound is used; "-" – a compound is not used.

2.2. Measurements of soil physical properties

Soil properties were measured in April, May, June, July, and August over the three-year period. Depending on the meteorological conditions, a total of 14 tests were carried out (Table 2).

Table 2. Dates of soil property tests in 2015–2017

2015	2016	2017
25 04 2015	29 04 2016	05 05 2017
11 05 2015	25 05 2016	30 05 2017
June was too dry	06 20 2016	28 06 2017
04 07 2015	20 07 2016 (after harvesting)	31 07 2017 (after harvesting)
07 08 2015 (after harvesting)	08 08 2016 (after soil tillage)	08 01 2017 (after soil tillage)

Soil porosity was sampled with a soil sampling drill from a depth of 0–10 cm. For each scenario, 5 measurements were taken. Soil porosity was determined with a vacuum air pycnometer after drying the samples to an air-dry mass. The total porosity P_b was calculated according to the formula (Maikštėniene et al., 2007):

$$P_b = \left(1 - \frac{\rho_d}{\rho_{k.f.}}\right) \cdot 100 \quad , \qquad (1)$$

where ρ_d – soil density, g cm^{-3};

$\rho_{k.f.}$ – soil solid phase density, g cm^{-3}.

Aeration porosity $P_{aer.}$ was calculated according to the formula (Maikštėniene et al., 2007):

$$P_{aer.} = P_b - (w \cdot \rho_d), \tag{2}$$

where w – soil water content, %.

Soil temperature at a depth of 0–5 cm in all variants was determined with a hand-held portable device "HH2 Moisture Meter", to which a "WET-2" type sensor was connected.

2.3. Measurement of CO_2 emissions from soil

CO_2 emissions from the soil were measured on the same dates as other physical soil properties. The measurements were carried out with the ADC BioScientific Lcpro+ System, a portable CO_2 gas analyzer consisting of a compact programming console, a soil respiration chamber, and a plastic ring to be inserted into the soil. Carbon dioxide emissions were measured 5 times in each scenario.

The programming console is connected to the soil breathing chamber at the selected measurement location. A metal ring was inserted into the selected measurement site and the chamber attached to it. The ring is inserted perpendicular to the soil and left in place. The measurement site must be free of grass or other elements that could damage the sensors. The telescopic probe shall deliver CO_2 from the atmosphere at a height of 3 meters. This height was chosen to prevent the measurement from being influenced by the person taking the measurement. The measurement is carried out for 10 minutes, observing fluctuations in carbon dioxide. The data is automatically recorded on a memory stick.

2.4. Meteorological conditions

April 2015 was unusually warm. The average temperature for the month was 1 ºC above the long-term average and precipitation was 7.6 mm above the long-term average (Fig. 2). May and June 2015 were 0.9 °C and 0.2 °C colder than the long-term average, with 10 mm of precipitation in May and 46.2 mm less than the long-term average in June. July 2015 was close to the long-term average, with 8.8 mm less precipitation than the long-term average. August was hot and dry, with an average air temperature of 20.3 °C and only 6.9 mm of precipitation. These data show that the 2015 growing season was very dry and deficient in moisture.

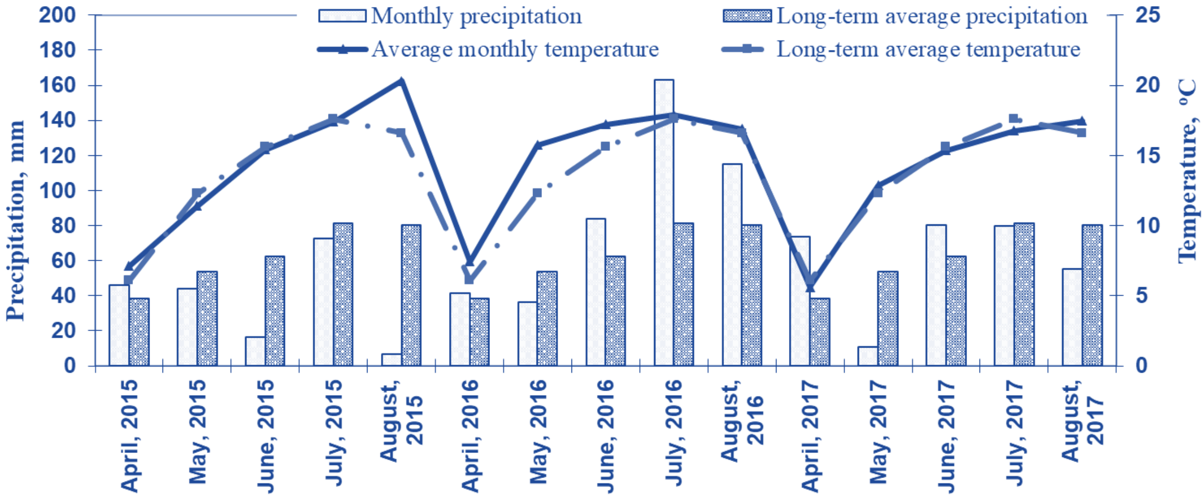


Fig. 2. Meteorological conditions during the study in 2015–2017

In April 2016, the average air temperature was 1 °C above the long-term average and in May it was 3.43 °C above the long-term average. April received 41.2 mm of precipitation, while May was a low-precipitation month, with only 36.4 mm, 17.4 mm below the long-term average. Warm and humid weather prevailed in summer. June was particularly warm, with an average air temperature of 17.21 °C, 1.61 °C above the long-term average. July and August were about 0.3 °C warmer than the long-term average. Compared to the long-term average, precipitation was 21.1 mm higher in June, 81.7 mm higher in July, and 34.6 mm higher in August. The summer period of 2016 was humid.

Although the average temperature in April 2017 (5.61 °C) was close to the long-term average (6.1 °C), precipitation was 1.9 times higher than the long-term average. The weather in May was moderately warm and dry. The air temperature was 12.87 °C, 0.57 °C above the long-term average. Precipitation was very low, at just 10.5 mm, compared with the long-term average of 53.8 mm for May. The summer weather in Lithuania was humid and cool. The average temperature in June was no different from the long-term average, but precipitation was 1.28 times the long-term average. Meteorological conditions in July were close to the long-term average, with an air temperature of 16.77 °C and 79.6 mm of precipitation. The weather warmed up to 17.47 °C in August, with a long-term average of 16.6 °C. Precipitation in August was 25.3 mm lower than the long-term average. Precipitation in the summer of 2017 was in line with the long-term average.

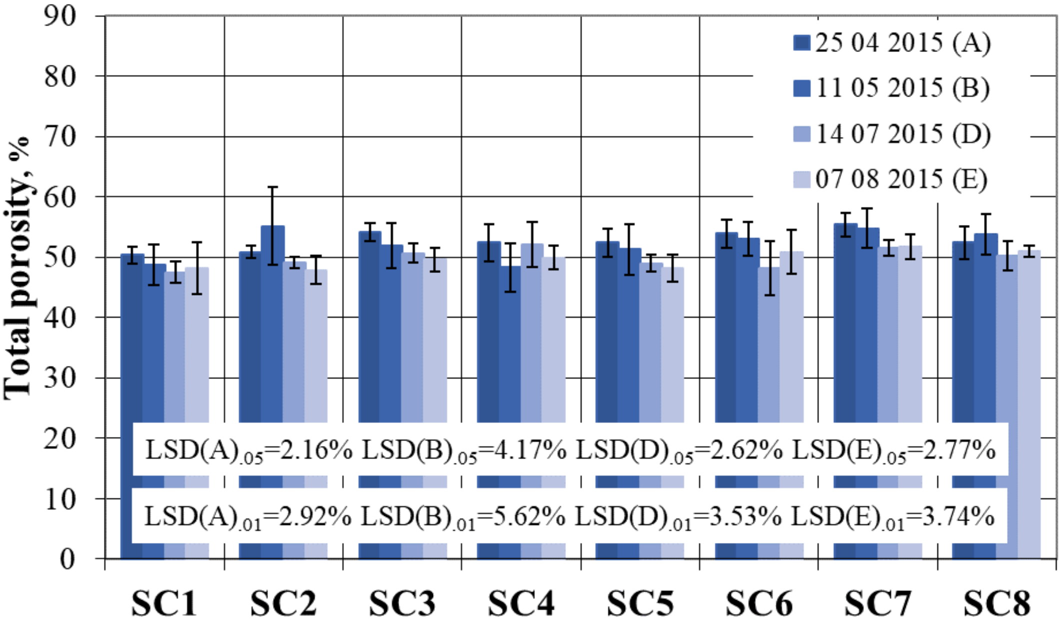
2.5 Statistical analysis

To assess the reliability of the results obtained, the data were evaluated by analysis of variance, ANOVA program. Arithmetic averages, standard deviations, and confidence intervals at 0.95 and 0.99 probability levels were determined. Significant differences between the averages of the variant data were determined by calculating the minimum threshold for the significant difference at $\text{LSD}_{.05}$ and $\text{LSD}_{.01}$ (Raudonius, 2017; Olsson et al., 2007). The research data were evaluated using correlation-regression methods of analysis. Correlation-regression analysis of data was performed by using STAT and SIGMA PLOT software. The probability level was indicated as follows: * – differences are significant at the 95% probability level; ** – differences are significant at the 99% probability level.

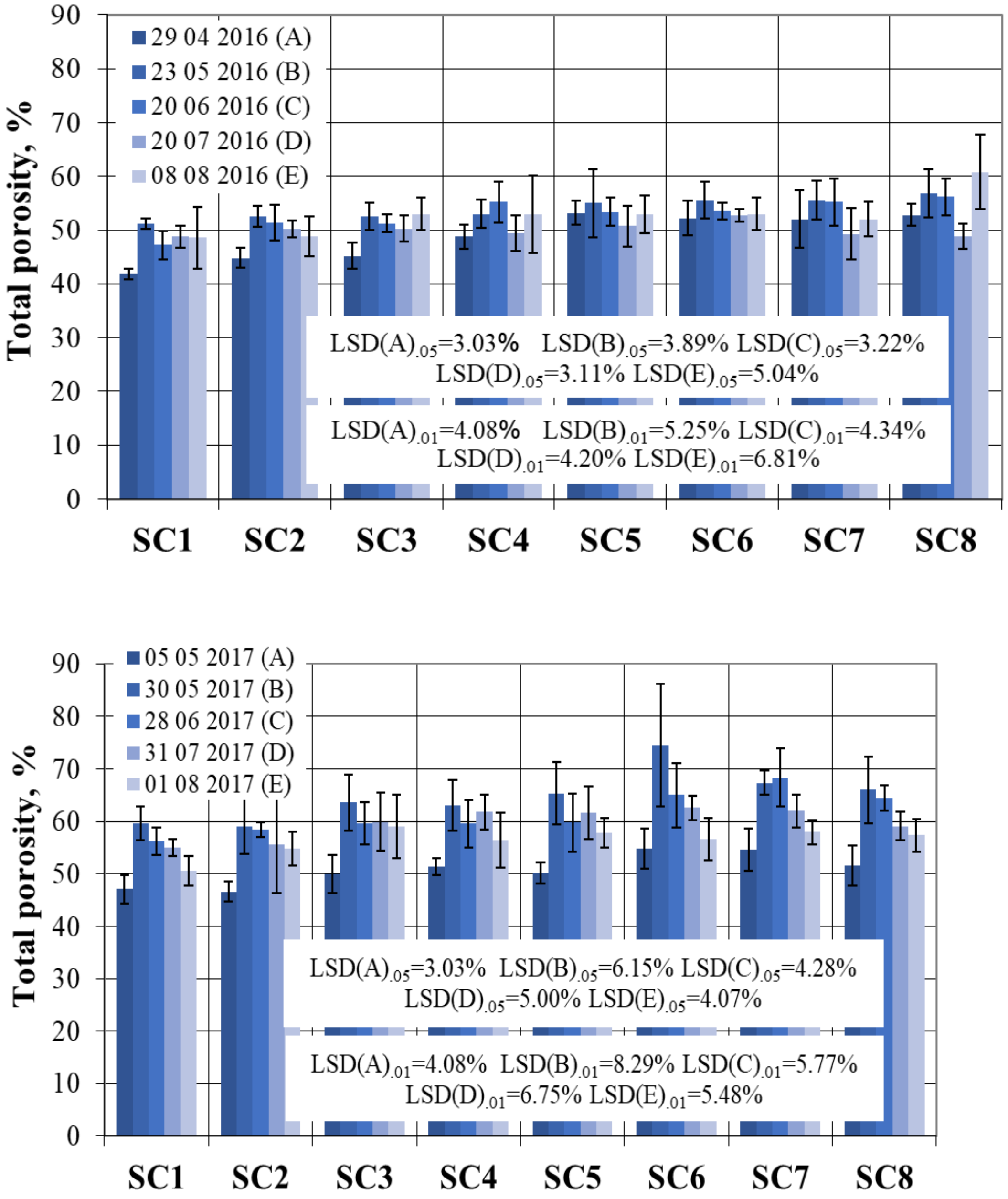
3. Results and discussion

3.1. Soil total porosity

In the first year of the study (2015), the total porosity ranged from 50.4% to 55.4% before the application of the biopreparations (Fig. 3a). Two weeks after the spraying of the biopreparations (11 May 2015), the total porosity was measured and it was found that all variants showed a decrease in total porosity ranging from 1.08% to 7.82%, except for variants SC2 and SC8, which showed an increase in total porosity of 8.4% and 2.6% respectively. No studies were carried out in June due to drought. In July, total porosity varied from 47.5% to 52.2% for all variants tested. Only one scenario, SC4, showed an increase in total porosity up to 8.1% compared to the total porosity found in May. In August, the range of variation in total porosity was between 47.9% and 51.7%. Significant differences were obtained in scenarios SC5 and SC6. Already in the first year of the study, a strong correlation between temperature and total porosity was found (r = –0.909 **).



a)



b)

c)

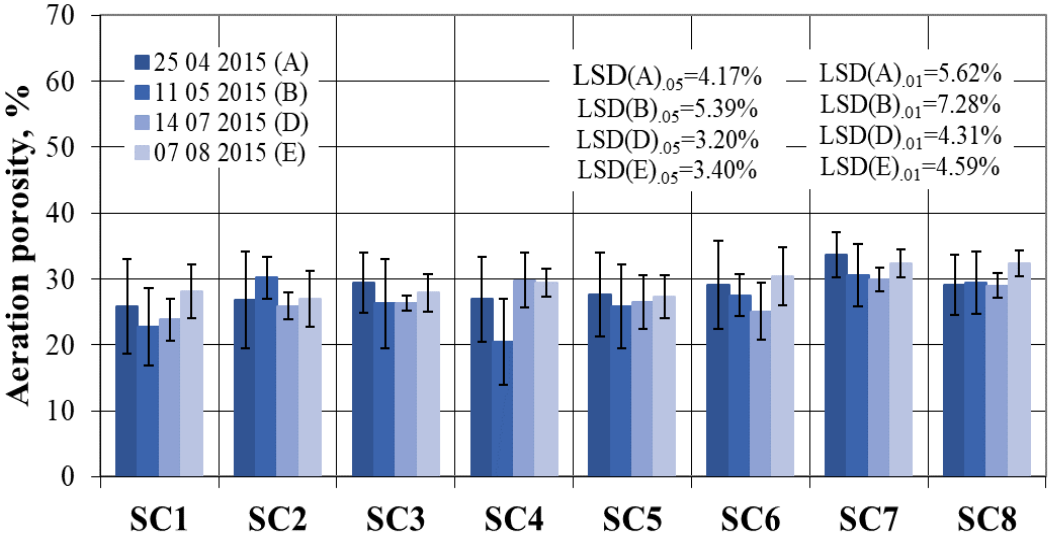
Fig. 3. Dynamics of soil total porosity: a) 2015, b) 2016, c) 2017

In 2016, the post-winter soil total porosity ranged from 41.8% to 53.2% (Fig. 3b). Total porosity was measured on 23 May 2016 after the application of the biopreparations and showed an increase in total porosity in all SCs. In the control variant SC1, an increase in total porosity was also found due to the meteorological conditions, as the warm and dry month of May prevailed. Carson et al. (2010) found that bacterial diversity increases with water potential ≤2.5 kPa in the sand and ≤4.0 kPa in silt + clay, which corresponds to a pore space filled with ≤56% water. The higher precipitation in June resulted in soil compaction, which reduced the total porosity in all scenarios except SC4, due to the presence of higher levels of microorganisms (*Azospirillum sp., Frateuria aurentia, Bacillus megaterium*) that prevented soil compaction. July was a high-precipitation month, which resulted in a decrease in total porosity of between 1.5% and 13% compared to June in all scenarios except SC1. In the control scenario, an increase of 3.4% was observed in July due to the filling of soil pores with water, which slightly increased the total porosity. In August, all scenarios showed an increase in total porosity compared to July, with the exception of scenarios SC1 and SC2, which consisted of non-bacterial components. These scenarios showed a decrease but not a significant one. Comparing the results obtained in April (before the application of the biopreparations) and August, it was found that the application of the biopreparations which were dominated by microorganisms, resulted in a more porous soil. The increase in total porosity ranged from 1.53% to 17.26% in most scenarios.

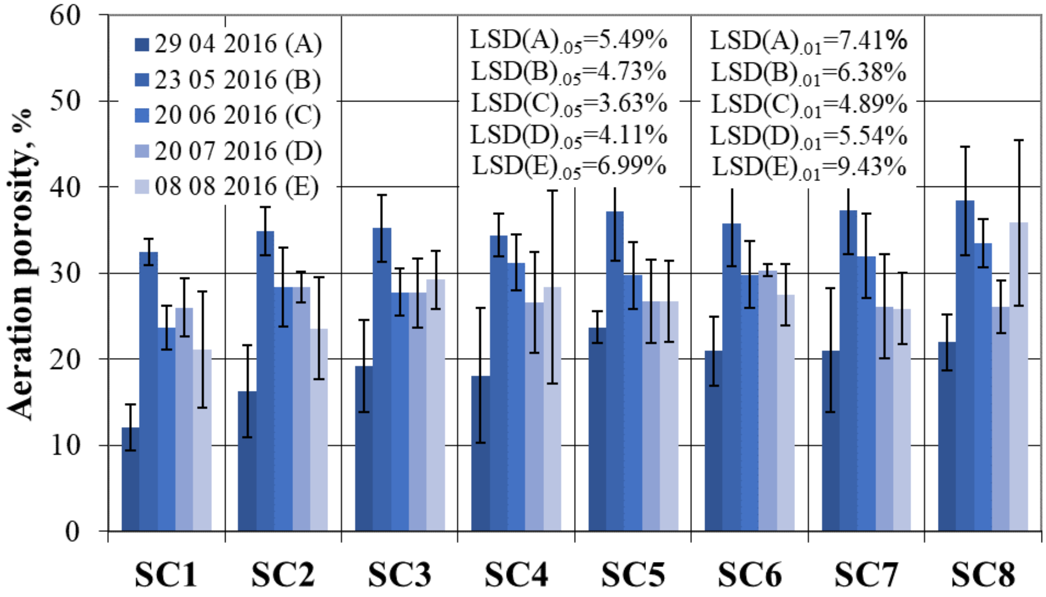
In 2017, total porosity at the beginning of May varied from 46.6% to 54.8% (Fig. 3c). Significant differences between the variants compared to the control variant were obtained in scenarios SC4, SC6, SC7, and SC8 at probability $P<0.01$ and in scenario SC5 at $P<0.05$. Biopreparations with higher bacterial content have a long-lasting effect, which is felt after overwintering with a higher total porosity index. The measurement of total porosity after spraying the biopreparations showed that in all SCs the total porosity increased from 18.54% to 26.54% because of the biopreparations and the environmental conditions. Scientists have found that biotreatments alter soil physicochemical properties (Banerjee, 2011; Cittenden et al., 2016). In June, when compared to the control, significant differences were obtained in scenarios SC6, SC7, and SC8 at $P<0.01$, although almost all SCs showed a decrease in total porosity, except for scenario SC7, which used *Azotobacter spp*. bacteria in combination with mineral oils, seaweed, and various grass extracts, which affected total porosity. A strong correlation was found between soil temperature and total porosity ($r = 0.932^{**}$), with increasing temperature having a positive effect on total porosity.

3.2 Soil aeration porosity

In the first year of the study, the aeration porosity before the application of the biopreparations ranged from 25.84% to 33.67% (Fig. 4a). Two weeks after the application of the biopreparations, the aeration porosity was measured as well. It was found that aeration porosity increased in all treatments, except for SC4, which showed a decrease of 10.13%. Scientists suggest that the application of biopreparations increases the organic carbon content of the soil, which can lead to a decrease in soil density and an increase in porosity (Montemurro et al., 2010; Peltre et al., 2015; Juknevičius et al., 2020). In July, aeration porosity varied from 23.81 % in SC1 to 29.95% in SC7 for the variants studied. In August, the variation in aeration porosity ranged from 26.92% in SC2 to 32.38% in SC.


70
60
50
40
30
20
10
0
Aeration porosity, %
25 04 2015 (A)
11 05 2015 (B)
14 07 2015 (D)
07 08 2015 (E)
LSD(A)$_{.05}$=4.17%
LSD(B)$_{.05}$=5.39%
LSD(D)$_{.05}$=3.20%
LSD(E)$_{.05}$=3.40%
LSD(A)$_{.01}$=5.62%
LSD(B)$_{.01}$=7.28%
LSD(D)$_{.01}$=4.31%
LSD(E)$_{.01}$=4.59%
SC1
SC2
SC3
SC4
SC5
SC6
SC7
SC8

a)


60
50
40
30
20
10
0
Aeration porosity, %
29 04 2016 (A)
23 05 2016 (B)
20 06 2016 (C)
20 07 2016 (D)
08 08 2016 (E)
LSD(A)$_{.05}$=5.49%
LSD(B)$_{.05}$=4.73%
LSD(C)$_{.05}$=3.63%
LSD(D)$_{.05}$=4.11%
LSD(E)$_{.05}$=6.99%
LSD(A)$_{.01}$=7.41%
LSD(B)$_{.01}$=6.38%
LSD(C)$_{.01}$=4.89%
LSD(D)$_{.01}$=5.54%
LSD(E)$_{.01}$=9.43%
SC1
SC2
SC3
SC4
SC5
SC6
SC7
SC8

b)

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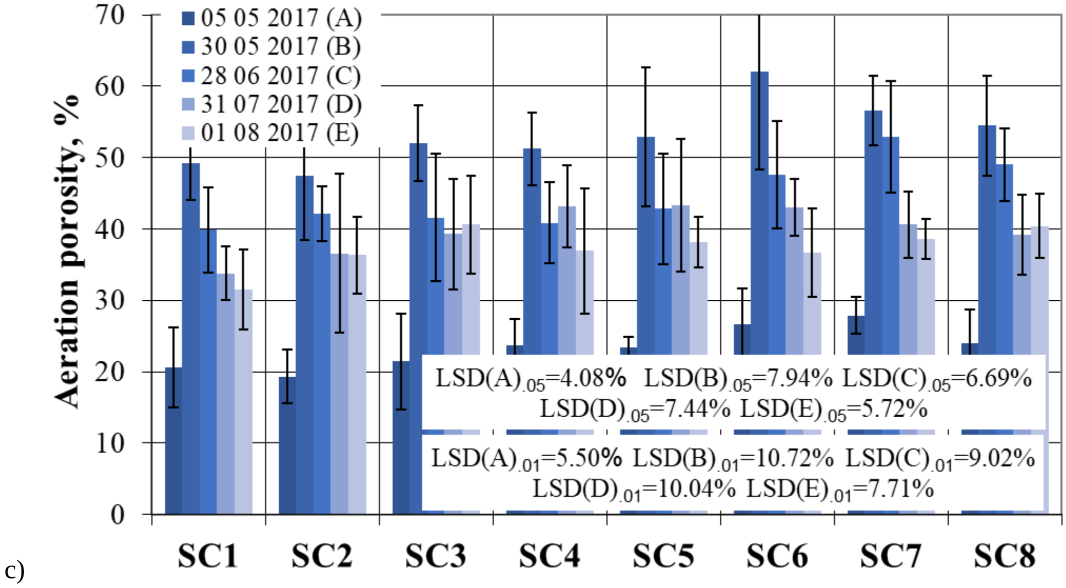


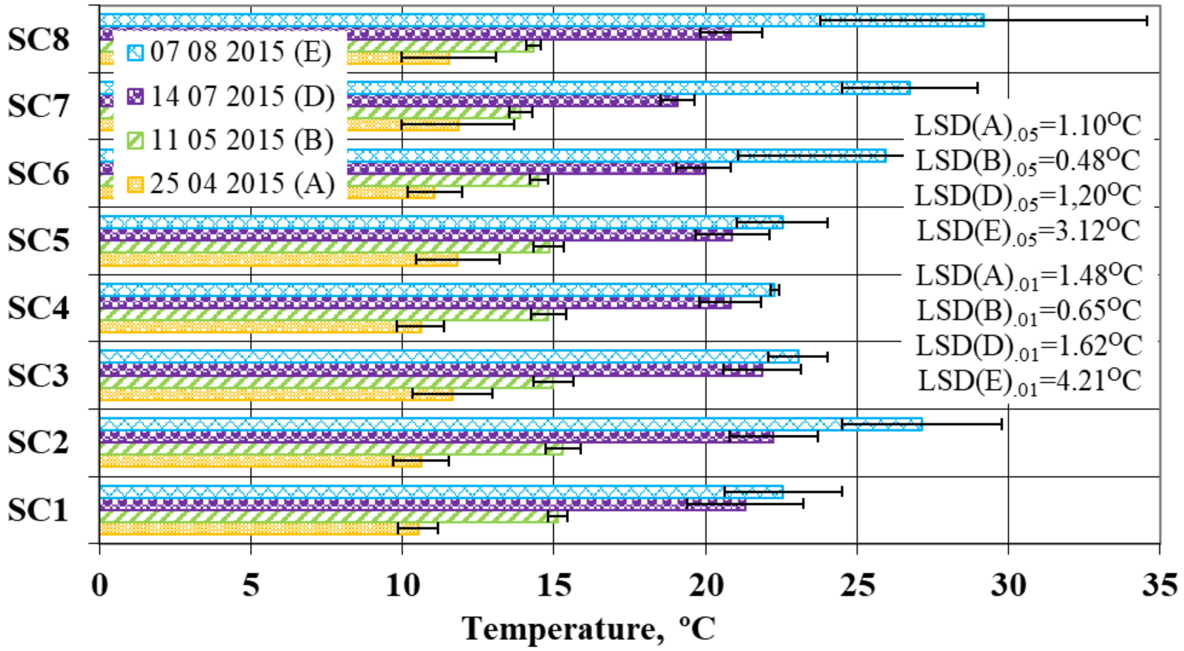
Fig. 4. Dynamics of soil aeration porosity: a) 2015, b) 2016, c) 2017

In 2016, in the spring, at the resumption of vegetation, aeration porosity was very low, ranging from 12.08% in SC1 to 23.72% in SC5 (Fig. 4b). Other researchers (Yevtushenko et al., 2016) have found that aeration porosity was above 20% regardless of tillage technology. In our case, SC1 had the lowest aeration porosity of 12.08%, while in other SCs it was around 20%. Aeration porosity measurements taken two weeks after the application of the biopreparations showed an increase in aeration porosity in all the SCs compared to the April tests. The highest increase of 2.6 times in aeration porosity was found in the control scenario SC1. The increase in aeration porosity may have been influenced not only by the sprayed biopreparations but also by favorable meteorological conditions. Many researchers suggest that porosity is particularly sensitive to tillage and environmental conditions (Cassaro et al., 2011, Lipiec et al., 2012, da Costa et al., 2014). The month of June was particularly warm with an average air temperature of 17.21 °C. The highest aeration porosity in June was found in SC4, SC7, and SC8. The lowest aeration porosity of 23.67% was found in the control variant. In July, the aeration porosity was similar to that in June. In August, all scenarios showed an increase in total porosity compared to July, except for scenarios SC1, SC2, and SC3.

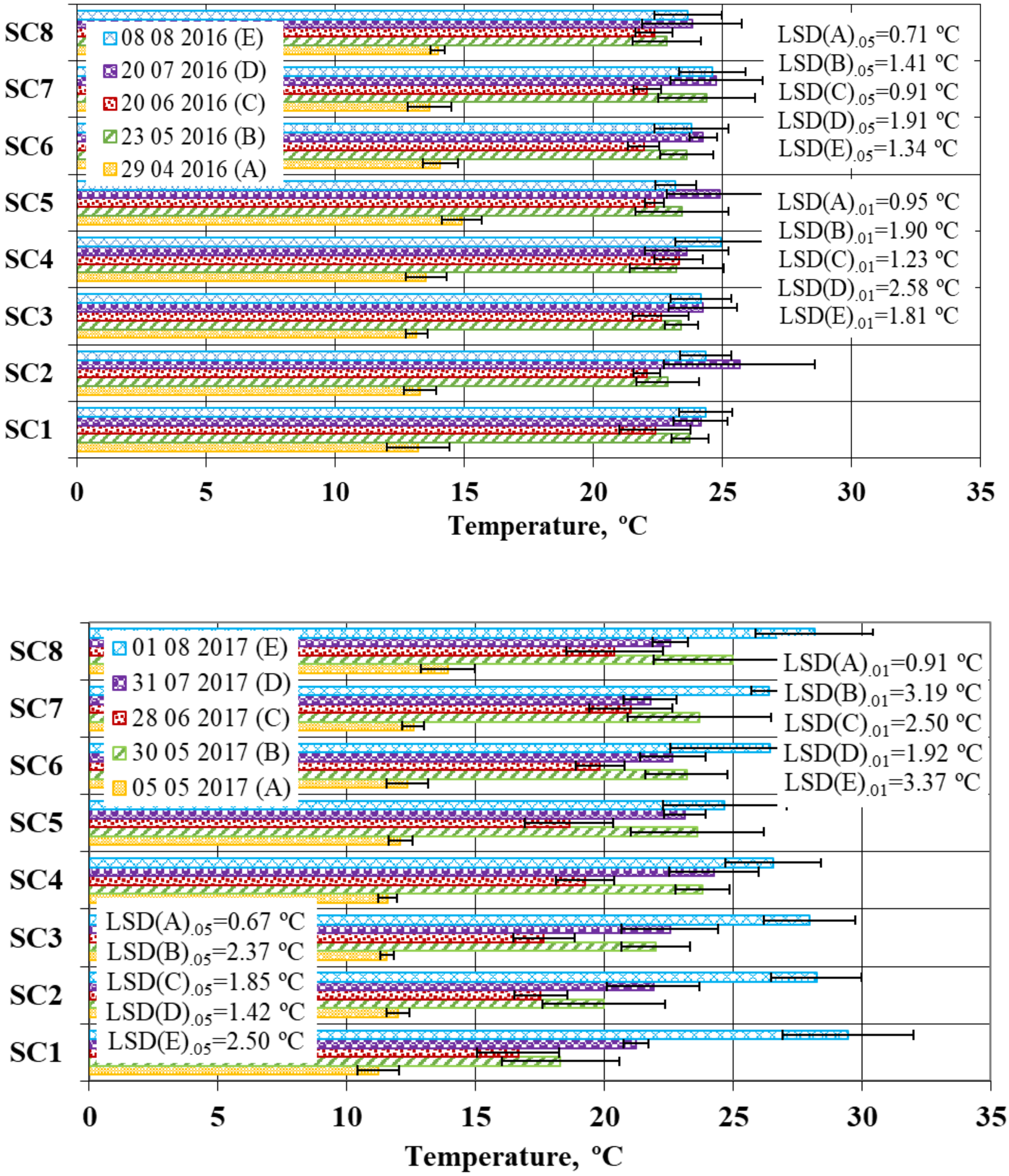
In 2017, the aeration porosity at the beginning of May varied from 19.33% to 27.88% (Fig. 4c). Aeration porosity measurements after spraying biopreparations showed that in all SCs, aeration porosity increased compared to the measurements taken in May because of biopreparations and environmental conditions. The measurements carried out in the third decade of June showed a decrease in aeration porosity in all variants compared to the measurements carried out in May. Researchers investigating effective microorganisms found no significant effect on porosity (Pranagal et al., 2020). At the end of July, aeration porosity ranged from 33.76% to 43.29%. After harvest, aeration porosity decreased in almost all scenarios except SC3 and SC8.

3.3 Soil temperature

Soil temperatures in April 2015 ranged from 10.52 °C to 11.81 °C (Fig. 5a). In April, compared to the control, soil temperature increased significantly in SC3, SC5, and SC7 scenarios ($P<0.05$). Soil temperature is one of the most important variables influencing soil respiration and depends on environmental conditions (Moyano et al., 2013, Sierra et al., 2015). As the soil gradually warmed in May, soil temperatures were found to be about 2–5 ºC higher than in May. Compared to the control scenario SC1, significantly lower soil temperatures were observed in SC7 and SC8 at $P<0.01$ and $P<0.05$ in SC6. In July, the soil temperature ranged from 19.08 ºC (SC7) to 22.04 ºC (SC2). In July, a significant decrease in soil temperature was found between control SC1 and SC6 ($P<0.05$) and between SC1 and SC7 ($P<0.01$) due to a denser crop. In August, the soil temperature was the highest recorded, as the absence of vegetative cover resulted in a significant warming of the soil. Researchers (Dai el al., 2021) found that soil temperature was lowest in the non-arable soil with straw mulch. It is likely that the uneven spread of crop residues after harvest increased or decreased soil temperature. Significant increases were found between scenarios SC1 and SC6, SC1 and SC7 at the 95% probability level, and between SC1 and SC2, SC1 and SC8 at the 99% probability level.



a)



b)

c)

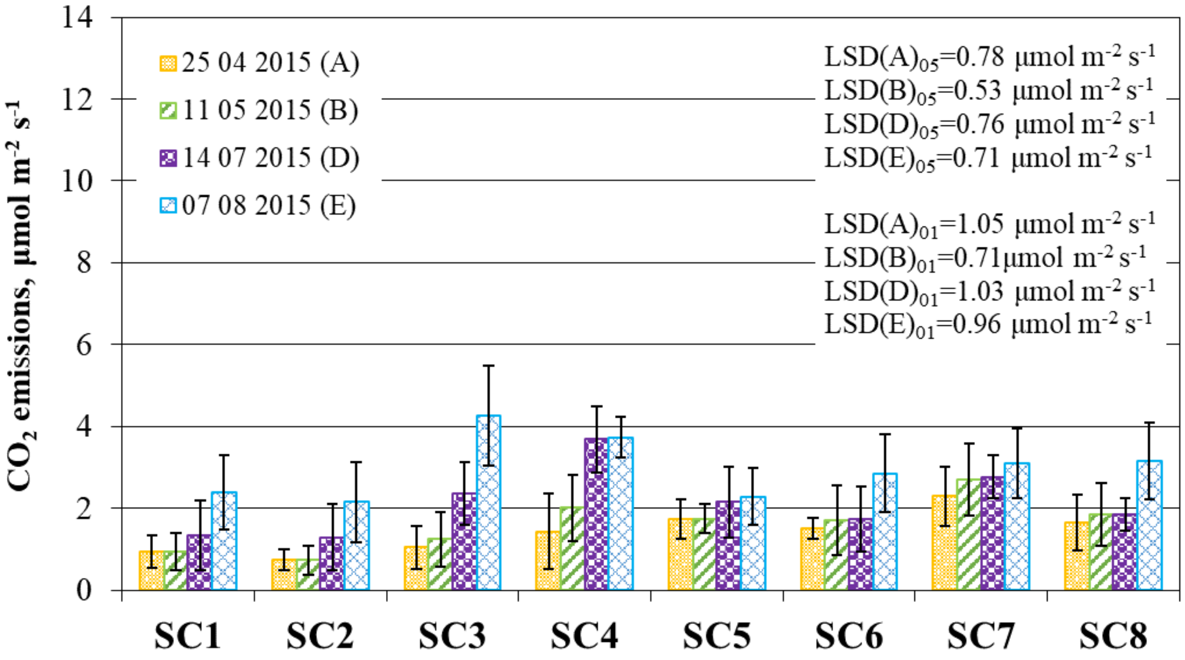
295 **Fig. 5.** Dynamics of soil temperature: a) 2015, b) 2016, c) 2017

In 2016, soil temperatures varied from 13.16 °C to 14.08 °C after the resumption of plant growth (Fig. 5b). A significant increase was found between scenarios SC1 and SC5 ($P<0.01$) and between SC1 and SC8 ($P<0.05$). At the end of May, soil temperature increased on average by about 10 ºC. In June, soil temperature ranged from 21.96 ºC (SC6) to 23.32 ºC (SC4). In the SC4 scenario, soil temperature was significantly higher than the control ($P<0.05$). In July and August, soil temperatures were found to be similar due to the settled weather, and no significant differences were found between the scenarios and the control.

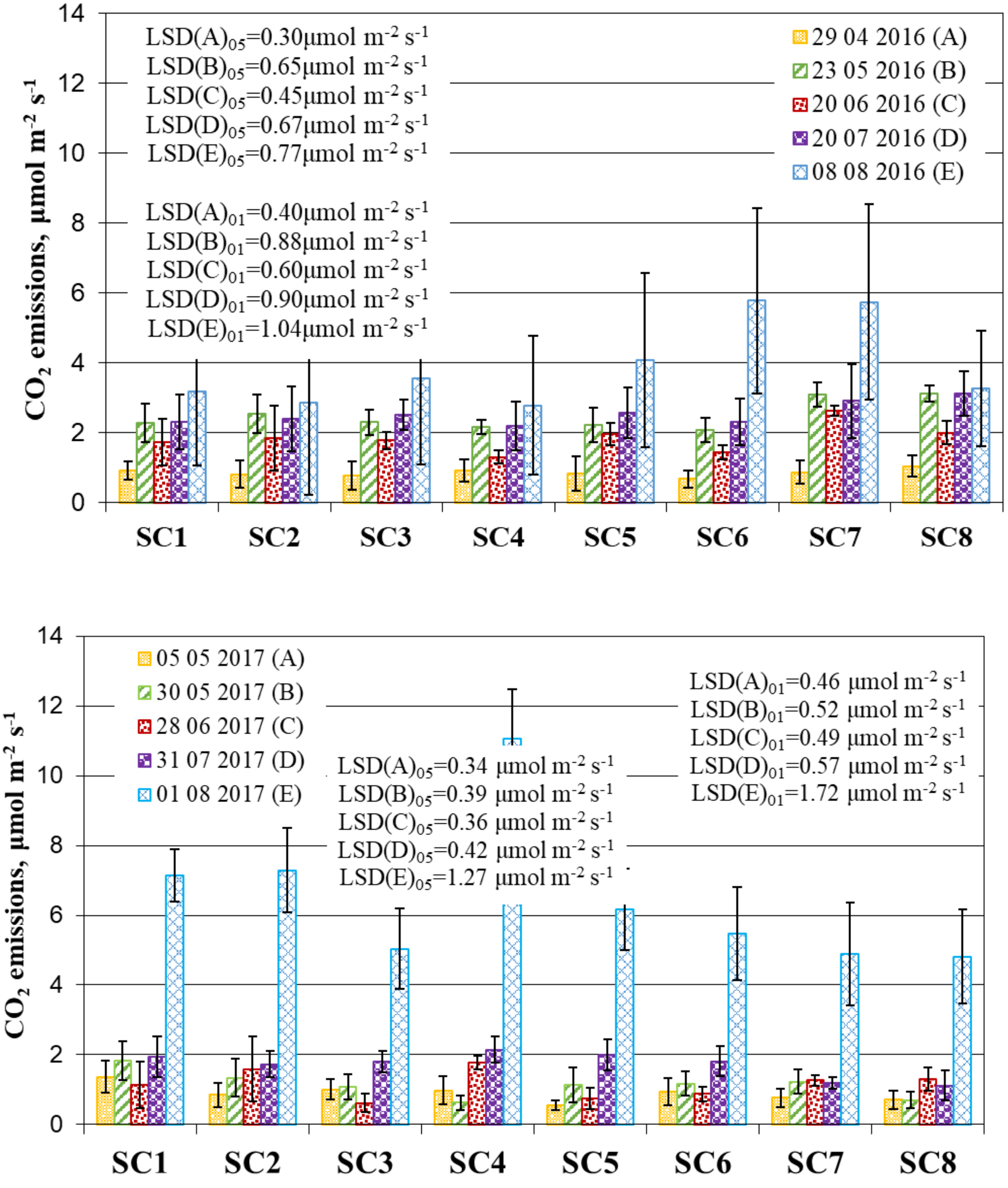
On 5 May 2017, the highest soil temperature was found in SC8 and the lowest in the control scenario (Fig. 5c). Soil temperatures were significantly higher in scenarios SC2 and SC5 ($P<0.05$), SC6, SC7, and SC8 ($P<0.01$) compared to the control scenario. Soil warming at the end of May resulted in a significant increase in soil temperature in all scenarios except SC2 at the 99% probability level. In SC8, a substantial increase was found at the 95% probability level compared to the control SC1. In June, the lowest soil temperature of 16.66 ºC was found in SC1 and the highest of 21.02 ºC in SC7. Significantly higher soil temperatures compared to the control were found in scenarios SC4, SC6, SC7, and SC8 at the 99% probability level. An increase in soil thermal conductivity increases temperature whereas an increase in soil heat capacity reduces temperature (Obia et al, 2020). At the end of July, soil temperatures ranged from 21.78 to 24.26 °C. A significant increase was found between control and SC4 ($P<0.01$) and between control and SC5 and SC6 scenarios ($P<0.05$). On 1 August, soil temperature increased in most of the scenarios studied compared to soil temperature at the end of July. However, a significant decrease in soil temperature ($P<0.01$) was obtained after the harvest between control and SC5, and at the 95% probability level soil temperature was significantly lower in the control scenario than in SC4, SC6, and SC7. It is likely that in all scenarios the higher soil surface temperatures were due to tillage, which allowed warm air to enter the soil.

3.4 CO_2 emissions from soil

In April 2015, the highest CO_2 emissions were observed in scenario SC7 with 2.29 µmol m^{-2} s^{-1} (Fig. 6a) and the lowest in SC2 with 0.73 µmol m^{-2} s^{-1}. Soil moisture, temperature, and biopreparation composition were the main influences on soil respiration. Research groups suggest that soil moisture influences CO_2 emission, with continuous moisture conditions increasing the bacterial content of the soil, resulting in higher CO_2 emissions from the soil compared to reirrigation (Jiao et al. 2023; Gultekin et al., 2023; Barnard et al., 2015). Tillage technology also has an impact, as tilled soil emits up to 21% more CO_2, but this depends on the soil type, organic carbon, and microorganism content of the soil (Abdalla et al., 2016; Chaplot et al., 2015; Huang et al., 2013). Canarini et al. (2017) found that in soils with more than 2% organic carbon, CO_2 emissions increase after drought, in contrast to soils with low carbon content. In our case, substantial increases between the control SC1 and SC5 scenarios were found at the 95% probability level, and between SC1 and SC7 at the 99% probability level. In May, soil respiration increased or remained the same compared to April. A substantial increase was found between the control and scenarios SC4, SC5, SC6, SC7, and SC8 at the 99% probability level. It is likely that the bacteria present in the bioassay (*Azospirillum sp., Frateuria aurentia, Bacillus megaterium, Azotobacter chroococum, Azospirillum brasilense, Azobacter* *vinelandii*) contributed to the substantial increases. Scientists suggest that soil microorganisms can increase CO_2 release (Klenz, 2015) under certain environmental conditions. According to the results of the May study, soil temperature had a significant influence on CO_2 emissions. The May results showed a strong correlation between soil temperature and CO_2 emissions (r = -0.903**), with rising temperature reducing emissions. Although in the following months of July and August, 335 CO_2 emissions were increasing due to higher ambient temperatures in all scenarios. A group of researchers (Tóth et al., 2018) investigated that soil emissions may be higher during the growing season. In August, the highest CO_2 emissions were found in SC3 at 4.26 µmol m^{-2} s^{-1} and the lowest in SC2 at 2.15 µmol m^{-2} s^{-1}. Significant increases between control and SC8 were found at the 95% probability level, and at the 99% probability level – between scenarios SC1 and SC4.



340 a)



b)

c)

Fig. 6. Dynamics of CO_2 emissions from soil a) 2015, b) 2016, c) 2017

In 2016, CO_2 emissions from the resumption of vegetation ranged from 0.69 µmol m^{-2} s^{-1} to 1.06 µmol m^{-2} s^{-1} (Fig. 6b). Other researchers (Forte et al., 2017) who studied conventional tillage reported that it leads to higher CO_2 emissions due to higher decomposition rates of soil organic matter and higher temperature fluctuations. At the end of May, soil respiration was more intense in all scenarios, and a substantial increase compared to the control was found in SC7 and SC8 at $P<0.05$. In June, with a decrease in air temperature of 1-2 ºC, CO_2 release slowed down and varied between 1.31 µmol m^{-2} s^{-1} and 2.64 µmol m^{-2} s^{-1}. A significant increase at $P<0.01$ was found between SC1 and SC7. In July, CO_2 emissions increased, but at the 95% confidence level, a significant increase was found between SC1 and SC7. Soil respiration increased further after harvest in August, ranging from 2.79 to 5.78 µmol m^{-2} s^{-1}. CO_2 emissions increased significantly compared to the control in SC6 and SC7 at the 99% probability level and in SC5 at the 95% probability level.

At the beginning of May 2017, CO_2 emissions were in the range of 0.55-1.37 µmol m^{-2} s^{-1} (Fig. 6c). In the SC1 scenario, soil respiration was found to peak at 1.37 µmol m^{-2} s^{-1}. At the end of May, all scenarios had higher CO_2 emissions due to ambient conditions and were significantly different from the control, with all SCs at the 99% probability level except SC2. Many field experiments have shown that CO_2 is significantly and positively correlated with soil organic carbon (Liu et al., 2014) and soil temperature (Cartwright and Hui, 2014), but in our case, it is the opposite. In June, CO_2 emissions increased significantly in SC1 compared to SC2 ($P<0.05$) and to the SC4 scenario ($P<0.01$). Significant reductions were obtained in scenarios SC5 ($P<0.05$) and SC3 ($P<0.01$). The settled temperature in July, which was close to the long-term average (around 10 °C), resulted in more intense soil respiration in all scenarios except SC7 and SC8. A significant decrease ($P<0.01$) was found between the control SC1 and SC7, SC8 scenarios, which could be influenced by the different compositions of the biopreparations. Drulis et al. (2022) state that bioproducts are substances that can improve crop productivity and quality, increase nutrient availability in the soil, improve plant nutrient use efficiency, and promote organic matter decomposition and humification in the soil. In all scenarios, CO_2 emissions increased by a factor of 3 to 5 in the range of 4.89-11.07 µmol m^{-2} s^{-1} after the harvest and tillage in August. These changes are likely due to the fact that the study was carried out immediately after tillage. The process of tillage greatly intensifies CO_2 emissions to the environment (Buragienė et., 2015). However, the differences between the scenarios were influenced by the different compositions of the biopreparations. Emissions were significantly higher only in scenario SC4 compared to the control ($P<0.01$) and significantly lower in scenarios SC3, SC7, and SC8 compared to the control ($P<0.01$).

Conclusions

In the first and second years of the study, the total porosity of the soil varied between 41% and 62%, while in the third year, the total porosity of the soil increased in all scenarios and over the whole study period compared to the control, ranging between 51% and 74%. This increase was due to the interaction between the long-term use of biopreparations and meteorological conditions.

Soil temperature was dependent on environmental conditions, crop density, and plant height. In the first year, soil temperature in August showed a significant increase compared to the control ($P<0.05$) in scenarios SC6, which used a biopreparation consisting of *Azotobacter vinelandi*, humic acid, gibberellic acid, Cu, Zn, Mn, Fe, Ca, Sodium molybdate; SC7, which used a biopreparation consisting of essential oils of plants, 40 species of various herbs extracts, marine algae extracts, and mineral oils, *Azotobacter vinelandi*, humic acid, gibberellic acid, Cu, Zn, Mn, Fe, Ca, sodium molybdate; SC2, which consisted of essential oils of plants, 40 species of various herbs extracts, marine algae extracts; and SC8, which consisted of *Azototobacter chroococcum, Azospirillum brasilense*, P, K, marine algae extracts. Similar trends were confirmed in the second and third years.

The use of biopreparations has had an impact on CO_2 emissions from soil to the environment. In the first year, it was found that, when looking at the total emissions from all measurements, all biopreparations and their mixtures, except for scenario SC2, increased CO_2 emissions from soil. In the second year of the biopreparation approach, the soil respiration results showed that only two scenarios, SC6 and SC7, resulted in higher cumulative CO_2 emissions, while in the other scenarios, the emissions from soil were very similar to the control. The cumulative effect of biopreparation application was most pronounced in the third year. In all scenarios, cumulative CO_2 emissions from the soil until the end of July were similar to or lower than in the control SC1. The measurements taken in August of the third year, immediately after tillage, gave very important results. In all scenarios, CO_2 emissions were found to have increased by a factor of 3–5 compared to the previous measurements, in the range of 4.89–11.07 $\mu mol\ m^{-2}\ s^{-1}$. Knowing that tillage intensifies CO_2 emissions from the soil, these studies showed that some biopreparations and their mixtures can reduce the emission intensity after tillage. This is confirmed by the results of this study, wherein scenario SC3 (biopreparation composition: essential oils of plants, 40 species of various herbs extracts, marine algae extracts, *Azospirillum sp.*, *Frateuria aurentia*, *Bacillus megaterium*, marine algae extracts), SC7 (essential oils of plants, 40 species of various herbs extracts, marine algae extracts, mineral oils, *Azotobacter vinelandi*, humic acid, gibberellic acid, Cu, Zn, Mn, Fe, Ca, Sodium molybdate), and SC8 (*Azototbacter chroococcum*, *Azospirillum brasilense*, P, K, marine algae extracts), significantly lower CO_2 emissions from the soil were obtained after tillage compared to the control scenario where no biopreparations were used.

Future research on the use of bacteria-based and environmentally friendly bioproducts should focus on increasing CO_2 storage in soil, simplifying agricultural operations, reducing inputs, and increasing the efficiency of crop production.

Competing interests

The contact author has declared that none of the authors has any competing interests.

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