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LONG-TERM EFFECT OF BIOCHAR APPLICATION ON CO_2 EMISSIONS IN A FLUVISOL

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Abstract

The use of biochar (BC) improves soil properties and crop yields, as well as reduces greenhouse gas emissions. The aim of this study was to evaluate the long-term effect of BC added to soil on CO_2 emissions in a Fluvisol. The study was conducted in the experimental field of Tsalapitsa village, where BC and nitrogen fertilization were applied 4, 5, and 6 years ago. The experiment started in 2019 with two doses of BC (5 and 10 t. ha^{-1}) and two rates of nitrogen fertilizers (130 and 260 kg. ha^{-1}). In 2023 another experiment was set up on the same field, with BC applied at 10 and 15 t. ha^{-1}. In 2023 and 2024, greenhouse gas CO_2 emissions were measured during the wheat and barley growing seasons. Following the 1–6 years of biochar application during the growing seasons significant differences in CO_2 emissions at different rates of BC and time of application were noted. The higher CO_2 emissions were recorded in the variants with high rate of nitrogen (N_{260}) in combination with the two doses of BC applied 4–6 years ago ($p < 0.007$, $p < 0.0038$, $p < 0.001$). A positive correlation was found between CO_2 emissions, total organic carbon (TOC), soil reaction, mineral nitrogen (N_{min}) content, and soil moisture for 2024, also supported by results from principal component analysis (PCA).

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In conclusion, application rates, time of addition of BC and nitrogen fertilizers have their impact on soil CO_2 emissions, but other controlling factors, such as temperature, precipitation, and soil characteristics, also played an important role.

Key words: biochar, CO_2 emissions, long-term experiment, soil properties

Introduction. The policy of European agriculture is to develop practices for reducing emissions and mitigating the effects of climate change in agricultural practices that do not only limit emissions, but also help soils absorb and retain excess carbon from the atmosphere [1]. In its plans for climate neutrality by 2050, Europe aims to bring 25% of agricultural land under organic farming as part of its Mission for Soil Health. In this context, the use of biochar (BC) as a soil amendment, which reduces the mineralization of soil organic carbon and makes it more resistant, is very relevant [2,3]. The research shows that the use of BC improves the physicochemical, hydro-physical, and biological properties and increases yields, as well as helps for carbon sequestration in soil [4,5]. Compared to other organic amendments, BC has greater stability, thereby increasing soil fertility, water retention capacity, crop growth and development [6].

Many studies have shown that the application of BC can have a positive, negative, or no effect on greenhouse gas exchange in soil [7–9]. These effects depend mainly on the types of biochar feedstocks, the conditions of pyrolysis, the type and properties of soil, climatic conditions, as well as the duration of biochar application [10,11]. Some studies indicate that BC increases CO_2 emissions from soil and reduces N_2O emissions [12,13]. Spokas [12] reports that ageing of BC for two years increases CO_2 emissions compared to the fresh BC changes. In a meta-analysis, Zhang et al. [13] reported that BC significantly increased CO_2 emissions in soil under wheat and vegetables (13% and 26%), at application rate of < 10 t. ha^{-1} or over 80 t. ha^{-1}. In the same analysis, it was found that BC produced at 400–600 °C also increased CO_2 emissions from soil by 14% and 45%, respectively, as well as in neutral and alkaline soil (24% and 17%).

The studies of the long-term effects of BC in mitigating greenhouse gas emissions in agriculture are an important issue. Ageing of BC in field conditions changes the properties of BC particles, increases the number of oxygen-containing functional groups, such as carboxylic and phenolic on their surfaces, increases their cation exchange capacity [14,15] and affects nitrogen retention and microbial activity in soil, and therefore greenhouse gas emissions [5,6].

The aim of this study was to assess the long-term effect of BC application on CO_2 emissions in a Fluvisol.

Materials and methods. The study was conducted in the period 2023 and 2024, in the experimental field of the ISSAPP "N. Poushkarov" institute in Sofia, in the village of Tsaplitsa (Plovdiv) on Alluvial-Meadow soil, classified as Fluvisol (WRB 2015). In 2022–2023 winter wheat was grown, and in 2024 barley

was grown, both without fertilization on the area of a five year experiment. The variants are with two doses of BC (5 and 10 t. ha^{-1}) and two doses of nitrogen (N) fertilizer (130 and 260 kg. ha^{-1}) applied in previous years (in 2019 and 2020) and left for observation of the past effect of BC application. In 2023 BC was applied in two doses (10 and 15 t. ha^{-1}) and one background rate of N fertilizer in the autumn of 2022. We used the fertilized variant only as control (KN130). The scheme of the experiment includes the following treatments: KN130 – control, $B_{(1)}$ – one year after BC application in 2023 and $B_{(2)}$ – two years after BC application in 2023, $B_{(4)}$ – four years after BC application in 2020, $B_{(5)}$ – five years after applying BC in 2019 and 2020, and $B_{(6)}$ – six years after applying BC in 2019. Wheat and barley were grown without adding N fertilizers, and the effect of BC was observed over time.

Biochar properties (450° (`https://nikimol.eu/`)) were: pH (H_2O) 9.7, organic C content 49%, total N 0.59%, available N 72 mg/kg, K 499 mg. 100^{-1} g, available P 43.4 mg. 100^{-1} g. Wheat (*Triticum aestivum* L.) was sown in October 2022. Barley (*Hordeum vulgare*) was sown in February 2024. Seeding density ensured 600 germinated seeds per 1 m^2. The experimental plot area was a randomised plot design in three replicates, each variant was 12.5 m^2.

Greenhouse gas emissions of CO_2 were measured during the growing season of the crops by variants and by crop. Plastic chambers were used to record emissions [16]. The accumulating chamber method is based on measuring the increase of CO_2 inside a chamber with an open bottom and a known volume, turned over on the soil surface. The flow value is calculated by a theoretical equation, according to the values of chamber's volume, atmospheric pressure and temperature, and CO_2 emissions are measured every month during the growing season. Data on N_2O emissions were compiled but did not show consistent trends and relationships with other parameters, and are, therefore not presented. The measurement times were between 9:00 and 13:00 h during the experiment. The selected measurement time is the optimal sampling time to average daily CO_2 emissions. CO_2 fluxes were measured on site using the calibrated "ALMEMO" tester with a CO_2 sensor. Then, the CO_2 leakage (kg $\times$ 1^{-1} day $\pm$ 1) is calculated according to [17] as:

$$FCO_2 = (M \times P \times V \times (C_2 - C_1))/(R \times T \times A \times (t_2 - t_1)),$$

where FCO_2 is the release of CO_2 from the soil (kg. ha^{-1} day^{-1}); M is the the molar mass of CO_2 (kg. mol^{-1}); P is the air pressure (Pa); V is the chamber volume (m^3); C_1 is the initial concentration of CO_2 (μmol.mol^{-1}); C_2 is the concentration of CO_2 after the incubation time (µmol.mol^{-1}); R is the gas constant (J.mol^{-1}·K); T is the air temperature (K); A is the chamber surface (m^2); $t_2 - t_1$ is the incubation period in days.

Soil samples were collected from each plot at the end of the growing season from the 0–15 cm soil layer. The main agrochemical parameters of the soil were analysed using the following methods: nitrogen (N) by the method of Bremner,

available phosphorus and potassium by the oxalate-lactate method of Ivanov. The electrical conductivity was determined in soil:water (1:5), ISO 11265: 2002. Soil pH/Eh were measured in a 1:2.5 soil:water. Total organic carbon (TOC) was determined by oxidation with K_2Cr2O_7/H_2SO_4 according to the method of Tyurin.

The daily air temperatures and precipitation were measured by the Meteobot weather station at the institute's experimental field. Measurements were taken at 15-minute intervals, and the average daily temperatures and total precipitation were calculated. Statistical data were analyzed by making use of STATGRAPHICS Centurion XV, One-way Anova methods, principal component analysis (PCA).

Results and discussion. ***Weather conditions.*** The data on temperature and precipitation during the study period were compared to a thirty-year period (1961–1990) accepted as the climatic norm (Fig. 1).

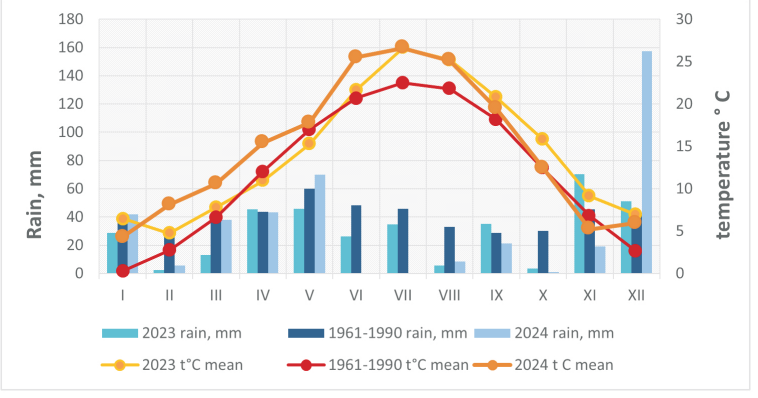


Fig. 1. Average temperature and precipitation for investigation period (2023–2024) compared to the climatic norm (1960–1990)

The summer months of 2023 and 2024 were very hot and dry. The average temperatures in July for the two years were higher than the average temperature for the climatic period. The study period of 2023 is characterized by reduced amount of precipitation, especially in the summer from June to August, but also in February and March. During the spring months (March to May) of 2024, the sum rainfall was 151 mm, and in terms of quantity they are over the average for the climatic period. The rainy period lasts until the end of May, after which a drought period follows (from June to August until the beginning of November), with the least amount of rainfall falling in June and July, around 10 mm (Fig. 1).

Carbon dioxide emissions. The impact of BC and fertilizers on CO_2 emissions over time is complex and depends on various factors, including the type of BC, the application rate, the soil type, and the specific environmental conditions. The results showed a high variability in the two growing seasons, 2023

and 2024. Applying BC in combination with N fertilizer, as well as the control (N130), resulted in significant differences ($p \leq 0.05$) in the measured emissions (Table 1). CO_2 emissions were in the range of 9.53 to 50 kg. $ha^{-1}day^{-1}$ during the three months of the first growing period in 2023. The higher CO_2 emissions were recorded in the variants with high rate of nitrogen (N260) in combination with the two doses of BC applied 4–6 years ago. Statistical analysis (Table 1) showed that the effect of adding BC on CO_2 emissions was significant ($p < 0.007$, $p < 0.0038$, $p < 0.001$) within three months in 2023. While the application time (year) and N fertilizer were not significant for all the measurements in 2023.

Table 1

Analysis of variance by controlling factors on CO_2 emissions in 2023 and 2024

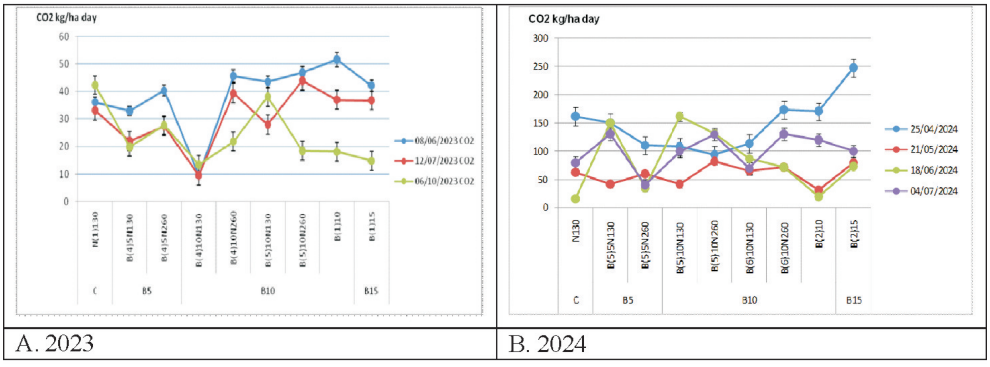
CO_2-emissions 2023				CO_2-emissions 2024				
Treatment	June	July	October	Treatment	April	May	June	July
$B_{(4)}$5N130	b	b	c	$B_{(5)}$5N130	e	h	g	b
$B_{(4)}$5N260	a	a	a	$B_{(5)}$5N260	b	i	d	b
$B_{(4)}$10N130	c	c	e	$B_{(5)}$10N130	c	c	a	c
$B_{(4)}$10N260	e	f	d	$B_{(5)}$10N260	a	g	f	f
$B_{(5)}$10N130	de	c	f	$B_{(6)}$10N130	d	f	b	d
$B_{(5)}$10N260	e	g	bc	$B_{(6)}$10N260	h	d	fg	e
$B_{(1)}$10	f	e	b	$B_{(2)}$10	g	b	e	a
$B_{(1)}$15	cd	de	a	$B_{(2)}$15	i	e	d	e
Control	b	d	g	Control	f	a	c	c
ANOVA factors	Significance, p							
N	*0.046*	*0*	*0.0014*	N	0.8253	*0.0362*	0.005	0.4438
BC	*0.007*	*0.0038*	*0.001*	BC	0.2025	0.0878	0.3645	*0.0095*
Year	0.653	0.9272	0.0531	Year	*0.0006*	0.6125	0.329	0.7676

Some studies have shown that BC amendment, especially when combined with high N fertilization, can lead to increased CO_2 emissions, an effect that can persist for years. According to Wang et al. [18], this is often attributed to the labile fraction of BC stimulating microbial activity and decomposition, leading to higher rates of respiration and C release. The highest emissions were measured at the variants in the new experiment, in which BC was applied alone in doses of 10 and 15 $t.ha^{-1}$ one year ago. Studies show that at the beginning of the BC application, emissions increased because of increased microbial activity.

In April 2024 CO_2 emissions were higher in the variants with added BC alone because the labile carbon of BC can become a food source for soil microorganisms, causing a temporary increase in their activity and respiration. In the same month, a decrease in emissions was observed at the higher dose of BC (10 t. ha^{-1}) applied 5–6 years ago. Very low emissions were measured in May, probably related to above-normal rainfall (70 mm), which may stimulate plant growth and

photosynthesis, that lead to absorption of CO_2 from the atmosphere. Analysis of variance of the controlling factors on CO_2 emissions in 2024 showed a significant difference ($p \leq 0.05$). Since p-values are less than 0.05, the factors analysed (BC, N fertiliser and time of application) have a statistically significant effect on CO_2 (Table 1).

In the variants with a low rate (130 mg $N.kg^{-1}$) of N fertilization and a low dose of BC (5 t. ha^{-1}), an increase of CO_2 emission from the soil was observed (Fig. 2A). These results are consistent with the study of Wang et al. [18], who used maize BC (produced at 500 °C) in combination with a low dose of N fertilization (50 mg $N.kg^{-1}$) and explained the effect of BC on promoting emissions with its labile fraction and the promotion of nitrification. According to other studies the CO_2 emissions increase with soil temperature [20]. The temperature increases lead to higher mineralization and organic matter losses as CO_2 is mainly produced because of soil microbial and root respiration.



A. 2023 B. 2024

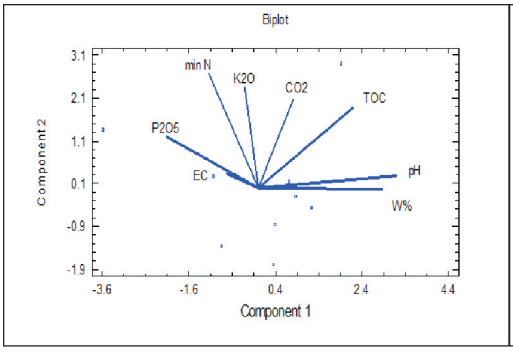
Fig. 2. Mean values of CO_2 emission (kg/ha day) from soil by variants, (A) year 2023 and (B) year 2024

The monthly variations in CO_2 emissions were influenced by climate parameters such as temperature, rainfall and soil moisture [19]. Correlation analysis of the data revealed that in 2023, CO_2 emissions measured in June and July had a positive correlation with air temperature ($R = 0.6332$, $p < 0.0004$ and $R = 0.5883$, $p < 0.0012$), while CO_2 emissions in October had a positive correlation with soil moisture ($R = 0.5222$, $p < 0.0052$). For the 2024 growing season (data not shown) there were no significant correlations between rainfall, soil temperature and CO_2 emissions.

Relationship between greenhouse gas emissions and other soil parameters. The principal component analysis (PCA) generated four components which had eigenvalues ≥ 1.0. Together they account for 92.07% (33%, 25%, 20%, and 13%) of the variance based on treatments in 2024 (Fig. 3). The first component was loaded by pH, TOC, soil moisture and CO_2 emissions, showing that

CO_2 emissions are related with organic carbon, soil moisture and pH. The second component was loaded only by CO_2 and N_{min}, because the impact of BC on soil CO_2 emissions is manifested with the availability of N. The third component is loaded by min P and K, whose behaviour is not related with CO_2 emissions. At low and moderate values of N_{min} in soil, BC can increase CO_2 emissions by stimulating microbial activity and decomposition of labile organic matter [19].

In our study, no consistent trends were observed for the studied CO_2 emissions in response to BC applied for 4–6 years on soil in the long term, similarly to other studies [18]. One reason might be the drought conditions during the growing seasons of both years. On the other hand, as BC aged in the field, its properties change, impacting soil processes and gas emissions.



Factor Loading Matrix After Varimax Rotation

	Component	Component	Component	*Component*
	1	*2*	*3*	*4*
pH	**0.857**	0.139	–0.400	–0.126
TOC	**0.844**	0.202	0.183	0.417
Min N	–0.191	**0.870**	0.357	0.102
P_2O_5	–0.368	0.033	**0.824**	–0.149
K_2O	0.271	0.162	**0.811**	0.294
CO_2	**0.399**	**0.920**	–0.111	–0.246
W%	**0.885**	–0.275	0.023	–0.304
EC	–0.08	–0.135	0.055	0.958

Fig. 3. Principal Component Analysis (PCA) based biplot, the correlation between investigation parameters (2024): CO_2 emission and soil parameters: pH, electrical productivity (EC), total organic carbon (TOC), soil moisture content (W%), min N ($mg.kg^{-1}$), available P_2O_5 (mg. 100 g^{-1}), K_2O (mg. 100 g^{-1}) and Factor Loading Matrix after Varimax Rotation

Conclusions. The study evaluated the long-term effects of BC application on soil CO_2 emissions in a Fluvisol. Following the 1 to 6 years from BC application during the growing seasons, significant differences in CO_2 emissions at different rates of BC and time of application were noted. The higher CO_2 emissions were recorded in the variants with high rate of nitrogen (N_{260}) in combination with the two doses of BC applied 4–6 years ago. A positive correlation was found between CO_2 emissions, total organic carbon, soil pH, N_{min}, and soil moisture for the year 2024. Seasonal variations, especially temperature and moisture, are major drivers of CO_2 flux, and may mask or influence the effects of BC. Complex factors such as time of application and rates of BC, and N fertilizers were identified as main triggers on soil CO_2, emissions, rather than just the temperature and soil moisture, which correlate only for some periods of the study. This highlights the complexity of CO_2 emissions dynamics in agricultural practices.

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