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Increasing the Environmental Sustainability of Greenhouse Vegetable Production by Combining Biochar Application and Drip Fertigation—Effects on Soil N₂O Emissions and Carbon Sequestrations

Yiming Zhao ¹, Shan Lin ^{1,2,*}, Haofeng Lv ¹, Yafang Wang ¹, Jing Hu ¹, Guoyuan Li ²
and Klaus Butterbach-Bahl ^{3,4,5} 

- ¹ College of Resources and Environmental Sciences, China Agricultural University, Beijing 100193, China; yimingzhao111@gmail.com (Y.Z.); lvhaofeng@cau.edu.cn (H.L.); yafangw65@gmail.com (Y.W.); bs20203030281@cau.edu.cn (J.H.)
- ² College of Life Science and Technology, Hubei Engineering University, Xiaogan 432000, China; lgyuan9@gmail.com
- ³ Institute for Meteorology and Climate Research (IMK-IFU), Karlsruhe Institute of Technology, 82467 Garmisch-Partenkirchen, Germany; klaus.butterbach-bahl@kit.edu
- ⁴ State Key Laboratory of Atmospheric Boundary Layer Physics and Atmospheric Chemistry, Institute of Atmospheric Physics, Chinese Academy of Sciences, Beijing 100029, China
- ⁵ Pioneer Center Land-CRAFT, Department of Agroecology, University of Aarhus, 8000 Aarhus, Denmark
- * Correspondence: linshan@cau.edu.cn; Tel.: +86-10-62733636



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Abstract: Drip fertigation with reduced fertilizer and water inputs has been widely used in greenhouse vegetable production in China. However, farmers usually do not apply additional organic material with a high carbon content, although soil organic carbon (SOC) concentrations are mostly below the optimum level for vegetable production. Returning straw or biochar to fields is an effective strategy for sustainability and environmental friendliness. We tested whether drip fertigation, (DIF) combined with maize straw (DIF+S) or biochar (DIF+BC), is a suitable option to improve SOC sequestration over eight growing seasons, and how these options affect soil N₂O emissions and yields or partial factor productivity of applied N (PFP_N) of crops over three growing seasons. During the winter–spring growing season, DIF+BC significantly reduced soil N₂O emission by 61.2% and yield-scaled N₂O emission by 62.4%, while increasing the tomato yield and PFP_N compared with DIF. Straw incorporation had similar trends but without significant effects. Conversely, straw and biochar incorporation increased N₂O emission during the autumn–winter season. The structural equation model indicated N₂O emission was dominantly driven by soil NH₄⁺-N concentration, temperature and moisture. The N₂O emission factor decreased significantly with increased PFP_N. Moreover, the contribution of biochar to the increased SOC was approximately 78%, which was four times higher than that of straw incorporation. Overall, the results highlighted the potential of drip fertigation with biochar incorporation to mitigate N₂O emissions, improve PFP_N and significantly increase SOC storage, which could all contribute to maintaining environmental sustainability and soil quality of greenhouse vegetable production.

Keywords: drip fertigation; incorporation of straw or biochar; N₂O emissions; soil organic carbon; greenhouse vegetable production

1. Introduction

Vegetables are an essential part of the human diet and a remarkable source of vitamins and minerals. Vegetable fields account for approximately 7% of the total croplands globally, and China has become the largest vegetable-producing country in the world [1]. The greenhouses in China cover an area of approximately 4.7 million hectares [2].

However, greenhouse vegetable production, specifically in China, is generally characterized by the use of large inputs of fertilizer and irrigation water compared with other arable cropping systems [3–5]. In China, a common practice amongst farmers is the overuse of N fertilizer ($>2000 \text{ kg N ha}^{-1} \text{ yr}^{-1}$) and irrigation water ($>1200 \text{ mm yr}^{-1}$) [6–8]. Such overuse results in low nitrogen and water use efficiencies, and it has a serious effect on the environment, leading to soil acidification [7,9], leaching of nitrate and dissolved organic nitrogen [10–12] and emission of the powerful greenhouse gas N_2O [13–15]. The total annual N_2O emissions from greenhouse vegetable production systems have been estimated to reach $50 \text{ kg N ha}^{-1} \text{ yr}^{-1}$ [8,14,15], with a global mean of approximately $12.4 \text{ kg N ha}^{-1} \text{ yr}^{-1}$ [5], which is 5–20 times higher than the mean soil N_2O emissions from global arable cropping systems [16].

To improve the resource utilization efficiency and reduce the N_2O emission and N leaching, an increasing number of farmers are using drip fertigation to produce vegetables in greenhouses in China [5,8,15]. However, this drip fertigation practice with a reduced rate of fertilization and irrigation may jeopardize fruit yields, as N or water may become limited for rapid crop growth, especially in sunken greenhouses with low soil organic carbon (SOC). The reason for the low SOC content is related to the construction of sunken greenhouses and the management of greenhouse vegetable cultivation. For construction, 0.5–2 m of autochthonous fertile topsoil is removed to build a north-facing back wall. This procedure exposes the subsoil, which originally shows SOC concentrations of $<4.9 \text{ g kg}^{-1}$ [6]. Although the SOC content could increase with decades of vegetable cultivation, it is still below the recommended optimum SOC value (25 g kg^{-1}) [6,17,18]. In addition, the over use of fertilizer and irrigation water combined with high soil temperature, intensive cropping and vegetable residue removal resulted in more soil C decomposition than sequestration [19–21]. Moreover, as a result of low heterotrophic soil respiration rates that are intrinsically linked to SOC content, the CO_2 concentrations in greenhouses during midday are often below atmospheric concentrations; such a phenomenon negatively affects crop productivity [22]. Therefore, improving SOC content is needed for the breakdown of the limited soil water and nutrient retention, as well as to release more CO_2 for plant photosynthesis.

The application of straw is the preferred option, as it may not only improve SOC content and promote soil respiration but also alleviate soil acidification and promote high crop productivity [23,24]. However, given the relative high soil moisture and temperature values in greenhouses, the added straw is quickly mineralized and significant increases in SOC may only occur after decades of high rates of straw application [25–27]. In addition, straw application may stimulate soil N_2O emissions through microbial nitrification and denitrification [28,29] and offset its advantages of increasing soil carbon sequestration [30].

Meanwhile, considering the risk of spreading soilborne fungi and pathogens to the next crop, the residues of vegetables grown in greenhouses could not be applied to soil and are usually completely removed at final harvest [31,32]. However, vegetable residues may be reapplied to vegetable soils if converted to biochar by pyrolysis at high temperatures. Biochar is a more recalcitrant organic material with higher organic carbon content than maize straw [33–36] and it has been found to have the potential to reduce greenhouse gas emissions when used as a soil amendment in agroecosystems [37–39]. Furthermore, biochar incorporation improves soil physical properties, such as the water-holding capacity [40] and aeration of soils [41,42], and provides an additional absorption capacity for mineral nutrients [33,43,44], which would be beneficial for plant growth under the drip fertigation practice. However, whether the application of biochar to greenhouse soils could significantly reduce soil N_2O emission fails to reach an agreement [45–47], as they depend on soil temperature and residual mineral N content [37,38,48].

In North China, greenhouse vegetable production is usually across two different growing seasons namely, the winter–spring (WS) season (from the end of August to the end of January) and the autumn–winter (AW) season (from the end of February to the end of June). These two growing seasons involve large differences in soil temperature and residual soil mineral N content before transplanting [4,15] which would influence soil C

and N cycling when organic material is added. Whether biochar or straw additions in combination with reduced rates of N fertilizers and irrigation water in different growing seasons could significantly reduce soil N₂O emissions and improve vegetable yield and SOC content has not been widely focused on. Here, we report on an experiment comparing the effect of straw versus biochar application on soil N₂O emissions over three growing seasons and SOC contents over a four-year period of eight growing seasons for greenhouse vegetable production systems, based on drip irrigation with optimized fertilization (DIF). As a control, DIF (380 kg N ha⁻¹ season⁻¹) was used without any additional application of straw or biochar. The implementation of drip fertigation with biochar incorporation was hypothesized to result in a significant reduction in soil N₂O emissions, with dependence on environmental conditions related to different growing seasons. Moreover, biochar addition could result in a significantly higher SOC sequestration rate than straw addition, due to its low decomposability and the fact that its yields cannot be negatively affected.

2. Materials and Methods

2.1. Field Site and Experiment Design

The field experiment was conducted in a sunken solar greenhouse in Northern China (Tianjin, Wuqin District, 39°25'35" N, 116°57'18" E), which has been used since the beginning of 2016 (i.e., the start of the experiment) for the production of tomatoes. The greenhouse was built in 2011, and a former arable field was used for wheat and maize cropping. Since 2011, greenhouses have been used for tomato and cucumber cultivation using flood irrigation and fertilization [4]. The main climatic and soil properties of the study sites are presented in Table 1. In agreement with local farmers' practices, tomatoes were cultivated during the WS (from the end of February to the end of June) and AW (from the end of August to the end of January) seasons, with both seasons separated by an approximately 1.5 month-long summer fallow period of August. The experiments covered the period from the beginning of 2016 to February 2020, that is, eight growing seasons (Table S1).

Table 1. General climatic characteristics at the experimental site and main soil properties of greenhouse soil (0–30 cm depth).

| | |
|--|---|
| Characteristics of experimental site | |
| Site | Wuqin District, Tianjin |
| Coordinates | 39°25'35" N, 116°57'18" E |
| Climate | Warm temperate monsoon climate |
| Mean annual precipitation (mm) | 532 (1981–2020) |
| Mean annual air temperature (°C) | 13.5 (1981–2020) |
| Annual sunshine duration (h) | 2392 (Meteorological Observation Station) |
| Characteristics of experimental soil | |
| Soil type | Aquic cambisols (FAO classification) |
| Soil texture | Silty loam |
| Particle fraction (%) | |
| Sand (20–2000 µm) | 30 ± 1.40 |
| Silt (2–20 µm) | 62 ± 1.50 |
| Clay (<2 µm) | 8 ± 0.26 |
| Bulk density (g cm ⁻³) | 1.34 ± 0.02 |
| Soil organic carbon (g kg ⁻¹) | 17.3 ± 1.31 |
| pH | 8.6 ± 0.05 |
| Electric conductivity (µs cm ⁻¹) | 370 ± 21 |

Values are expressed as the mean ± standard error ($n \geq 3$).

In the experiment, three different treatments were investigated, which are as follows:

- DIF: drip irrigation with reduced fertilization;
- DIF+S: drip irrigation with reduced fertilization, plus the incorporation of maize straw;
- DIF+BC: drip irrigation with reduced fertilization, plus the incorporation of biochar.

A randomized block design was used with three replicates; thus, nine plots were investigated. Each plot covered an area of 24 m² (6.7 m × 3.6 m). Adjacent plots were separated by an impermeable plastic membrane, which was dug down to a soil depth of 0.6 m. Each plot consisted of three raised beds (0.7 m in width), plus walkways (0.5 m in width) in between. One raised bed was used for soil and plant sampling, whilst the other two were used for greenhouse gas flux measurements or determination of fruit yields. Two rows of tomato were planted in each raised bed, with a row spacing of 0.5 m and a plant spacing of 0.35 m. Two drip lines (2 cm in diameter) were laid on the soil surface close to the tomato plants, with drip emitters spaced every 0.4 m. Each emitter allowed for a mean flow rate of 3.19 ± 0.07 L h⁻¹.

All plots were managed in the same manner for fertilization and irrigation. Fertilization management consisted of basal fertilization and fertigation during the growth period. At the beginning of each growing season, 200 kg N ha⁻¹ commercial chicken manure (1.45% N and 13.2% C, δ¹³C = -21.83‰ ± 0.061‰) was incorporated into the top 20 cm of the soil as basal fertilization 2 days before transplanting. Immediately following transplantation, all plots received initial flood irrigation with 50–100 mm of water to ensure the survival of tomato seedlings. Drip fertigation started 20 days after transplanting, with rates of fertigation depending on weather conditions and the tomato growth stage. On sunny days, the drip fertigation rate was 3–5 mm of irrigation water with 2.0–4.0 kg N ha⁻¹ of water-soluble chemical fertilizer (N:P₂O₅:K₂O, 19:8:27) (Figure 1a and Table S1). In agreement with farmers' practices and following transplanting, all treatment plots were covered with a plastic film (10 µm thick) to reduce soil evaporation losses and increase soil temperatures. Details of the fertilization and irrigation schemes in the eight growing seasons are provided in the Supplementary Materials (Table S1).

DIF treatment did not receive any C input in addition to the C applied with the chicken manure (1821 kg C ha⁻¹ season⁻¹, Table 2), DIF+S treatment received an additional 3500 kg C ha⁻¹ season⁻¹ of maize straw while DIF+BC received 2100 kg C ha⁻¹ season⁻¹ of biochar (Table 2), which was based on 40% of the organic carbon that was assumed to be lost during the conversion of peanut husks to biochar. Therefore, from the perspective of carbon balance, the initial carbon inputs of DIF+S and DIF+BC remained essentially the same. As there is no biochar produced from vegetable residues on the market, peanut husks biochar commonly found in the market was used in this study. Maize straw and biochar were incorporated with chicken manure (basal fertilizer) into the top 20 cm of soil. The total C and N concentrations in maize straw were 41.6% and 0.96%, respectively. The δ¹³C signal of maize straw was -14.18‰ ± 0.055‰ (Table 2). The total C and N concentrations of the commercial biochar (Golden Future Agriculture Technology Co., Ltd., Anshan, China), which were derived from peanut husks, were 54.8% and 1.08%, respectively, with the C fraction showing δ¹³C of -24.37‰ ± 0.019‰. The pH of biochar was 9.2 ± 0.1, measured in distilled water at a w/w ratio of 1:5 (Table 2).

Table 2. Total C and N concentrations and δ¹³C signature of chicken manure, maize straw and biochar. In addition, seasonal rates of organic matter applications for the different treatments are provided.

| Organic Matter | Total C % | Total N % | δ ¹³ C ‰ | Incorporation Rate (kg C or N ha ⁻¹ Season ⁻¹) | | | | | |
|----------------|--------------|--------------|------------------------|---|-----|-------|------|--------|------|
| | | | | DIF | | DIF+S | | DIF+BC | |
| | | | | C | N | C | N | C | N |
| Chicken manure | 13.2 ± 0.06 | 1.45 ± 0.01 | -21.83 ± 0.061 | 1821 | 200 | 1821 | 200 | 1821 | 200 |
| Maize straw | 41.6 ± 0.10 | 0.96 ± 0.01 | -14.18 ± 0.055 | - | - | 3500 | 80.8 | - | - |
| Biochar | 54.8 ± 0.10 | 1.08 ± 0.01 | -24.37 ± 0.019 | - | - | - | - | 2100 | 41.4 |

Values are expressed as the mean ± standard error (n = 3). - means no addition.

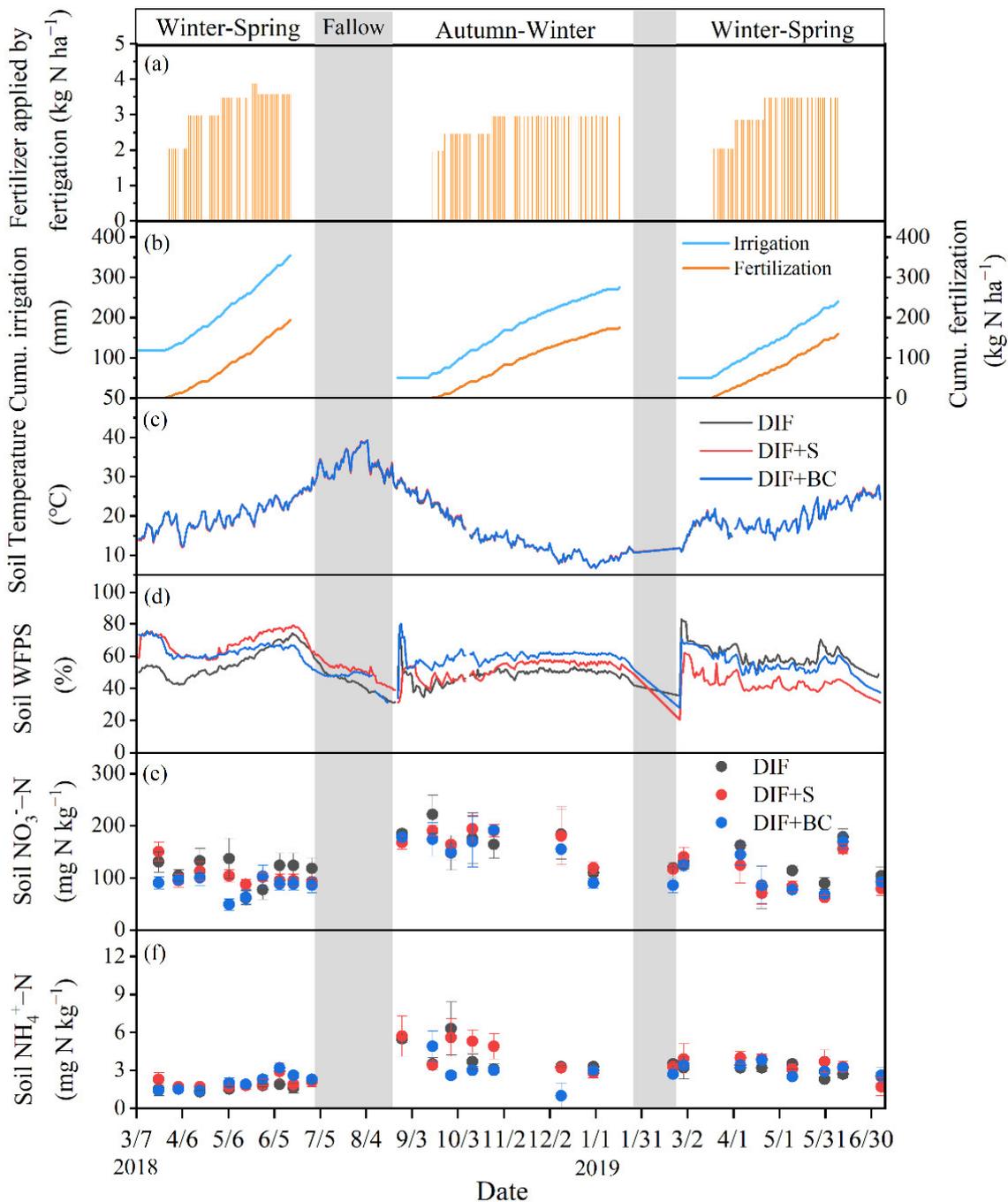


Figure 1. Temporal dynamics of daily fertigation (i.e., synthetic fertilizers applied with irrigation water) (a), cumulative amount of irrigation and fertilization (b), soil temperature (c) and soil water-filled pore space (WFPS, 0–12 cm soil depth) (d) and soil inorganic N concentrations in 0–30 cm soil depth ((e): NO_3^- -N concentration; (f): NH_4^+ -N concentration) for the three growing seasons, for which information on soil N_2O fluxes (Figure 3) is also available. Different colors in panels (c–f) refer to different treatments, including black: drip fertigation (DIF); red: drip fertigation with straw incorporation (DIF+S); blue: drip fertigation with biochar incorporation (DIF+BC). SDW represents soil dry weight. Error bars represent the standard error ($n = 3$).

2.2. Measurement of Soil N₂O Emission

Soil N₂O fluxes were measured in only three out of eight growing seasons, specifically from March 2018 to July 2019 (2018WS, 2018AW, and 2019WS). Moreover, soil N₂O fluxes were measured during the 6-week fallow period in the summer of 2018 by using the static vented chamber method described by Yao et al. (2019) [14]. A stainless-steel frame (50 × 50 cm) was inserted into the soil to a depth of 25 cm in each plot directly following field preparation and 2–3 days before vegetable transplanting. Each frame contained one tomato plant. For flux measurements, a chamber was fixed tightly on the metal frame, with the height of the chamber varying between 60 and 120 cm, depending on plant growth. Flux measurements were conducted daily or at intervals of every 2 days in the morning hours between 08:30 and 10:00. For each flux measurement, the headspace of the closed chamber was sampled using a 50 mL polypropylene syringe with a Luer lock at 0, 15, 30, 45 and 60 min after chamber closure. Headspace gas samples were analyzed for N₂O by gas chromatography (Agilent 7890A, Agilent Technologies, Santa Clara, CA, USA; equipped with an electron capture detector) within 7 h of sampling. N₂O fluxes were determined from the linear or nonlinear changes in gas concentrations over time after chamber closure, using the approach outlined by [49]. The fluxes were corrected for the actual values of air temperature and pressure, as described by Zhao et al. (2021) [15].

2.3. Measurements of SOC and δ¹³C

The first topsoil (0–30 cm) sampling for the analysis of SOC content and bulk δ¹³C signals was conducted in January 2016. In brief, before the start of the experiment, nine independent soil samples were randomly obtained within the greenhouse. The soil was resampled for SOC and δ¹³C in February 2020, and soil samples were removed from all treatment plots. Three soil samples down to a depth of 30 cm were obtained from the raised beds in each plot for soil sampling. All soil samples for SOC analysis were acquired using a gouge auger (3 cm diameter). For later analysis, the soil samples were air-dried and then ball-milled for 10 min (250 r min⁻¹). Then, soil samples (20–30 mg dry weight) were packed into tin capsules and analyzed for organic carbon and ¹³C/¹²C isotope ratios by using an elemental analyzer (Costech ECH 4024, Costech company, Picarro, Italy) and mass spectrometer (Elementar Isoprime 100, Elementar company, Hanau, Germany), respectively. Maize straw and BC were analyzed in the same manner for ¹³C/¹²C isotope ratios, with isotope ratios reported as δ¹³C values (‰) by referring to ¹³C_{VPDB} standards [50].

2.4. Auxiliary Measurements

In each plot, the soil temperature and volumetric water content at 0–12 cm were monitored hourly using frequency domain reflectometry devices and a temperature sensor (CS655, Campbell Scientific, Inc., Logan, UT, USA). Soil mineral N (NO₃⁻-N and NH₄⁺-N) concentrations at 0–30 cm soil depth were determined in soil samples from raised beds at weekly intervals (Figure 2) by extracting fresh soil with 100 mL of 0.01 mol/L CaCl₂ solution. The extracts were analyzed using a continuous flow analyzer (AA3, Nordstadt Hamburg, Germany). The tomato yield was calculated as the total weight of fresh fruit (36 plants per subplot), which were picked by hand at the time of commercial maturity, that is, when the tomato seeds were yellow and the fruit pulp appeared red. Fruit samples were obtained from raised beds that were used to determine vegetable yields.

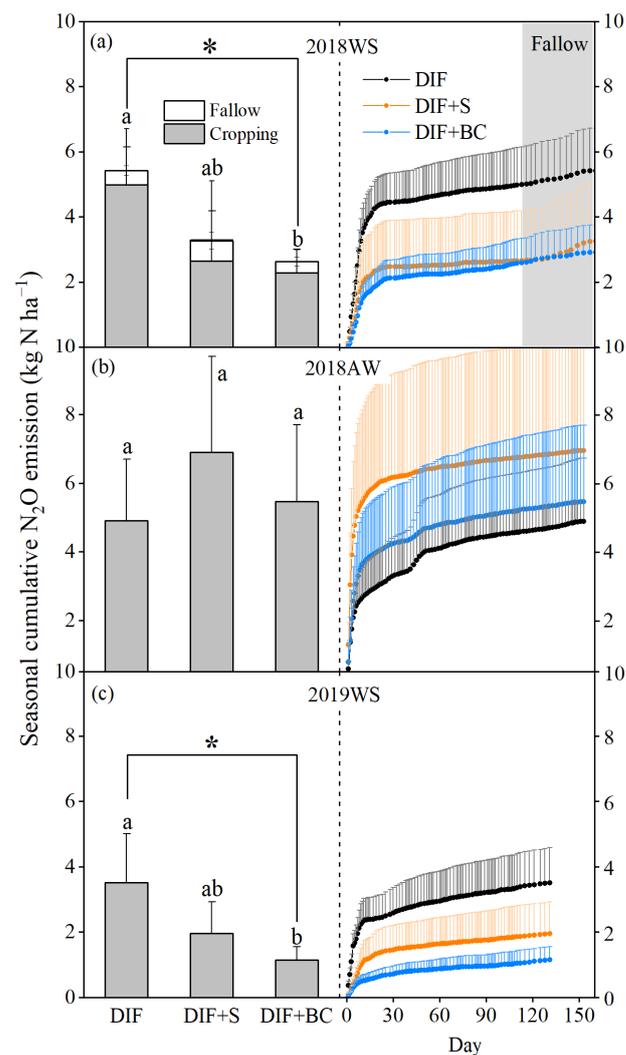


Figure 2. Cumulative seasonal N_2O emissions for 2018WS (a), 2018AW (b) and 2019WS (c) for the three different treatments. Following the 2018WS growing season, measurements of soil N_2O fluxes also continued during the fallow period (52 days). DIF: drip fertigation; DIF+S: drip fertigation with straw incorporation; DIF+BC: drip fertigation with biochar incorporation. * indicates significant difference between treatments ($p < 0.05$, $n = 3$). Different lowercase letters indicate significant difference amongst three treatments ($p < 0.05$, $n = 3$). Error bars represent the standard error ($n = 3$).

2.5. Calculation of Crop Performance and Environmental Parameters

Annual fruit yields, N fertilizer application rate, and annual N_2O emission were all calculated by the mean value of 2018WS and 2019WS plus 2018AW.

Seasonal or annual partial factor productivity of applied N (PFN_N) was calculated as follows:

$$PFN_N \text{ (kg yield kg}^{-1} \text{ N)} = \frac{\text{Fresh vegetable yield (kg ha}^{-1}\text{)}}{\text{N fertilizer application (kg N ha}^{-1}\text{)}} \quad (1)$$

Seasonal or annual yield-scaled N_2O emission ($Yield-N_2O$) was calculated as follows:

$$Yield - N_2O \text{ (g N Mg}^{-1} \text{ yield)} = \frac{\text{Cumulative } N_2O \text{ emissions (g N ha}^{-1}\text{)}}{\text{Fresh vegetable yield (Mg ha}^{-1}\text{)}} \quad (2)$$

In this study, the seasonal or annual apparent N_2O emission factor ($EF_{Apparent}$) was calculated without considering the background N_2O emissions as follows:

$$EF_{Apparent} (\%) = \frac{\text{Cumulative } N_2O \text{ emissions (kg N ha}^{-1}\text{)}}{\text{Total N fertilizer application (kg N ha}^{-1}\text{)}} \times 100 \quad (3)$$

The annual rate of soil C sequestration (ΔSOC) between the end (2020) and beginning (2016) of the experiment was calculated as follows:

$$\Delta SOC (\text{t C ha}^{-1} \text{ yr}^{-1}) = \frac{SOC_{2020} - SOC_{2016} (\text{t C ha}^{-1})}{4} \quad (4)$$

here SOC_{2020} and SOC_{2016} represent the measured topsoil (0–30 cm) SOC content in 2020 and 2016, respectively.

The percentage of increased SOC contents due to the additional application of straw or biochar was calculated as follows:

$$SOC (\%) = \frac{SOC_{DIF+S \text{ or } DIF+BC} - SOC_{DIF} (\text{g C kg}^{-1} \text{ SDW})}{C \text{ input (straw or biochar)} (\text{g C kg}^{-1} \text{ SDW})} \times 100 \quad (5)$$

where $SOC_{DIF+S \text{ or } DIF+BC}$ represents the SOC content in DIF+S and DIF+BC, SOC_{DIF} represents it in DIF, $C \text{ input}$ is the total amount of straw or biochar carbon addition rate for the eight growing seasons and SDW represents the soil dry weight.

2.6. Statistical Analyses

Statistical analyses were conducted using SAS 8.0 (SAS Institute Inc., Cary, NC, USA) and OriginPro 9 (OriginLab Corporation, Northampton, MA, USA). Treatment effects were assessed using one-way ANOVA and independent-sample t tests. Differences between treatments were determined by Duncan's test at a significance level of 0.05. Shapiro–Wilk normality and Leven's test were used to check the normality and homogeneity of variance of all the data. Pearson's correlation was used to analyze the correlation between PFP_N and $EF_{Apparent}$. The latent variable path analyses of structural equation modelling (Amos, 24, SPSS, IBM) were used to analyze the influence of soil water content, temperature and mineral N content on N_2O emissions.

3. Results

3.1. Soil Environmental Conditions

Except for the flood irrigation event on the day of transplanting, the average number of drip fertigation events was approximately 62 during for all the treatments in each growing season (Figure 1a). The seasonal rates of irrigation water and chemical N fertilizer were $343 \text{ mm season}^{-1}$ and $187 \text{ kg N ha}^{-1} \text{ season}^{-1}$, respectively (Figure 1b and Table S1). Soil temperatures showed a pronounced seasonal pattern, with the highest values of up to $40 \text{ }^\circ\text{C}$ in summer and lowest values during the winter periods ($>5 \text{ }^\circ\text{C}$, Figure 1c). Soil temperatures during transplanting for the WS season (February–March) were usually approximately $11 \text{ }^\circ\text{C}$ – $14 \text{ }^\circ\text{C}$. By contrast, soil temperatures during transplanting for the AW season (August) were always $>25 \text{ }^\circ\text{C}$ (Figure 1c). Across all treatments, the highest soil water-filled pore space (WFPS) values were observed immediately following transplantation, which was directly followed by a single flood irrigation event (50–100 mm). In the following event, soil moisture was maintained at approximately 60% WFPS by drip fertigation (Figure 1d). Soil mineral N concentration fluctuated in the range of 61–222 mg N kg^{-1} for $\text{NO}_3^- \text{-N}$ (mean: $128 \pm 5.0 \text{ mg N kg}^{-1}$) and 1.0–6.3 mg N kg^{-1} for $\text{NH}_4^+ \text{-N}$ (mean: $3.2 \pm 0.2 \text{ mg N kg}^{-1}$). Soil $\text{NO}_3^- \text{-N}$ and $\text{NH}_4^+ \text{-N}$ concentrations following transplanting tended to be higher in the AW season than in the WS season (Figure 1e,f).

3.2. Seasonal Cumulative N_2O Emission and Fluxes

The seasonal cumulative N_2O emissions in 2018WS, 2018AW, and 2019WS varied in the range of 3.5–5.4 kg N ha⁻¹ yr⁻¹ for DIF and 2.0–6.9 and 1.2–5.5 kg N ha⁻¹ yr⁻¹ for DIF+S and DIF+BC, respectively (Figure 2). Seasonal N_2O emissions for DIF+BC were significantly reduced compared with those for DIF in WS seasons 2018WS (−52%) and 2019WS (−69%), with opposing trends in the AW season. No significant difference was detected between DIF+S and DIF+BC in the WS and AW seasons (Figure 2). The peak N_2O emission fluxes occurred in the first 10 days following transplanting and initial flood irrigation, accounting for up to 70–80% of total seasonal emissions (Figures 2 and 3). Within 3–4 days after basal fertilization and irrigation in the WS seasons, the daily N_2O fluxes were up to the highest (DIF: 447–480, DIF+S: 142–236, DIF+BC: 105–197 g N ha⁻¹ d⁻¹) and then declined sharply by at least one order of magnitude, until the harvest time in the WS seasons (Figure 3a). Meanwhile, in the 2018AW season, the highest N_2O emission peaks appeared 1 day after basal fertilization in the order of DIF+S (1739) > DIF+BC (794) ≥ DIF (785 g N ha⁻¹ d⁻¹). The differences in daily N_2O emission fluxes between with and without straw or biochar incorporation showed an opposite trend in dependence on growing seasons (Figure 3b). In 2018WS and 2019WS, straw or biochar incorporation significantly reduced daily N_2O emissions compared with DIF during the basal fertilization period. Meanwhile, in 2018AW, the incorporation of straw or biochar stimulated daily N_2O emissions after basal fertilization, and the increased N_2O emissions induced by straw incorporation was the highest, up to 109 times higher than that induced by biochar (Figure 3b). However, in the WS and AW seasons, the different effects of straw or biochar incorporation on daily N_2O emissions was negligible after 1 month of basal fertilization (Figure 3b).

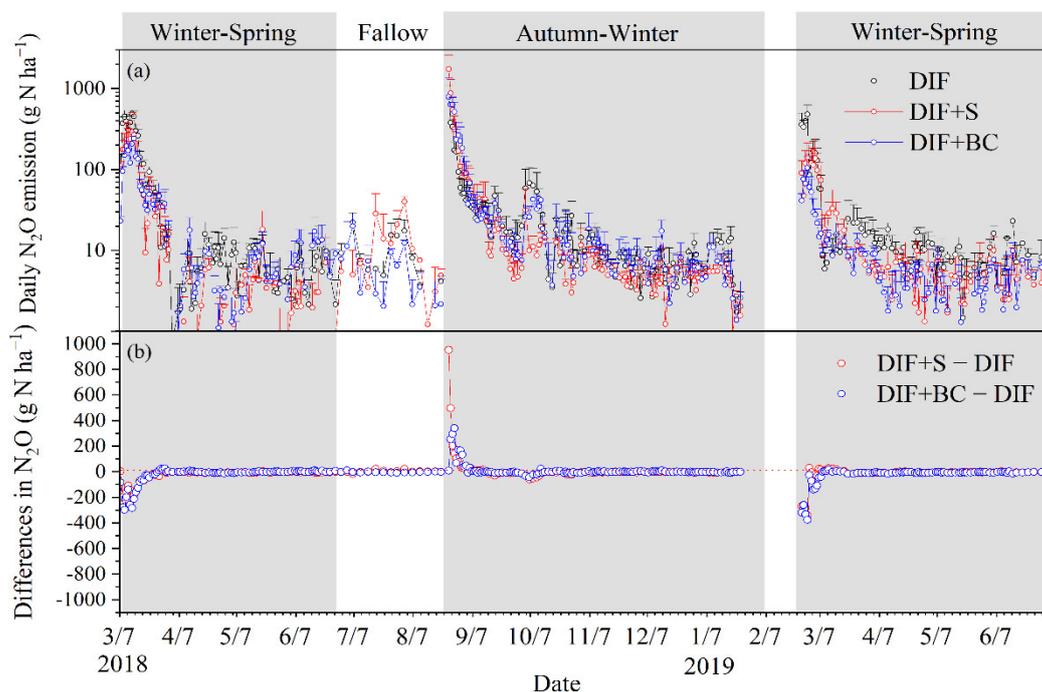
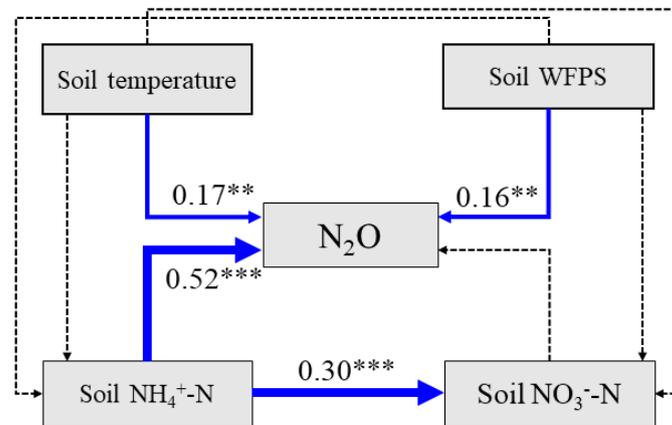


Figure 3. Daily fluxes of N_2O emissions during three growing seasons for the three different treatments (a) and differences in daily N_2O emissions between with and without straw or biochar incorporation (b). DIF: drip fertigation; DIF+S: drip fertigation with straw incorporation; DIF+BC: drip fertigation with biochar incorporation. DIF+S – DIF and DIF+BC – DIF means the differences between with and without S or BC incorporation. Error bars represent the standard error ($n = 3$).

Notably, daily N_2O emissions were significantly affected by soil temperature, moisture, and mineral N concentration (Figure S1). Soil NH_4^+ -N concentration had the largest positive direct effect (0.52) on soil N_2O emissions, followed by soil temperature (0.17) and

soil WFPS (0.16, Figure 4). Moreover, soil $\text{NH}_4^+\text{-N}$ concentration had a significant positive direct effect (0.30) on soil $\text{NO}_3^-\text{-N}$ concentration (Figure 4).



$$\text{CMIN/DF} = 0.824, \text{RMSEA} = 0.000, \text{AIC} = 38.82$$

Figure 4. Structure equation model of daily N_2O emissions and soil environment parameters (soil temperature, soil WFPS, soil $\text{NO}_3^-\text{-N}$, and $\text{NH}_4^+\text{-N}$). The solid blue lines indicate a significant effect, whilst the dashed lines indicate no significant effect. Larger path coefficients are reflected in the width of blue lines. The numbers are standardized path coefficients. CMIN/DF, RMSEA, and AIC are goodness of fit statistics. ** $p < 0.001$, *** $p < 0.001$. ($n = 208$).

3.3. Yield-Scaled N_2O Emission and EF_{Apparent}

The seasonal yield-scaled N_2O emissions in DIF+S and DIF+BC were significantly 53% and 72% lower than that in DIF in the WS seasons (Table 3), whereas straw or biochar incorporation had an opposite effect in the AW season. On an annual scale, the yield-scaled N_2O emissions ranged from $40.5 \text{ g N Mg}^{-1} \text{ yr}^{-1}$ (DIF+BC) to $56.6 \text{ (DIF) g N Mg}^{-1} \text{ yr}^{-1}$, without significant differences between treatments. Similar trends were found for the apparent N_2O emission factor, i.e., % of fertilizer N emitted in form of N_2O without deducting background emissions. The annual EF_{Apparent} were 1.22% (DIF), 1.00% (DIF+S), and 0.82% (DIF+BC) (Table 3).

Table 3. Seasonal and annual values of yield-scaled N_2O emissions, apparent N_2O emission factor, tomato yield and partial factor productivity of applied nitrogen fertilizer (PPF_N) during winter–spring 2018, autumn–winter 2019 (2019AW) and winter–spring 2019 (2019WS).

| | 2018WS | 2018AW | 2019WS | 2018–2019 |
|---|----------------------------|---------------------------|----------------------------|----------------------------|
| Yield-scaled N_2O emission ($\text{g N}_2\text{O-N Mg}^{-1}$) | | | | |
| DIF | $51.8 \pm 8.2 \text{ a}$ | $63.6 \pm 14 \text{ a}$ | $46.7 \pm 15 \text{ a}$ | $56.6 \pm 9.4 \text{ a}$ |
| DIF+S | $33.0 \pm 7.7 \text{ ab}$ | $75.3 \pm 26 \text{ a}$ | $25.5 \pm 12 \text{ ab}$ | $52.4 \pm 17 \text{ a}$ |
| DIF+BC | $24.6 \pm 1.1 \text{ b}$ | $64.8 \pm 13 \text{ a}$ | $13.0 \pm 0.91 \text{ b}$ | $40.5 \pm 5.4 \text{ a}$ |
| Apparent N_2O emission factor (%) | | | | |
| DIF | $1.38 \pm 0.22 \text{ a}$ | $1.30 \pm 0.28 \text{ a}$ | $1.46 \pm 0.48 \text{ a}$ | $1.22 \pm 0.16 \text{ a}$ |
| DIF+S | $0.83 \pm 0.19 \text{ ab}$ | $1.61 \pm 0.55 \text{ a}$ | $0.81 \pm 0.38 \text{ ab}$ | $1.00 \pm 0.29 \text{ a}$ |
| DIF+BC | $0.67 \pm 0.03 \text{ b}$ | $1.46 \pm 0.30 \text{ a}$ | $0.48 \pm 0.032 \text{ b}$ | $0.82 \pm 0.041 \text{ a}$ |
| Tomato yield ($\text{Mg fresh fruit ha}^{-1}$) | | | | |
| DIF | $105 \pm 2.0 \text{ a}$ | $76.7 \pm 3.9 \text{ a}$ | $75.3 \pm 3.3 \text{ b}$ | $167 \pm 4.5 \text{ b}$ |
| DIF+S | $99.4 \pm 13 \text{ a}$ | $80.3 \pm 5.2 \text{ a}$ | $76.8 \pm 5.6 \text{ ab}$ | $176 \pm 2.3 \text{ a}$ |
| DIF+BC | $107 \pm 5.6 \text{ a}$ | $84.5 \pm 12 \text{ a}$ | $88.3 \pm 2.8 \text{ a}$ | $182 \pm 15 \text{ a}$ |
| PPF_N ($\text{kg fresh fruit kg}^{-1} \text{ N}$) | | | | |
| DIF | $266 \pm 5.0 \text{ a}$ | $204 \pm 10 \text{ a}$ | $210 \pm 9.2 \text{ b}$ | $222 \pm 3.3 \text{ a}$ |
| DIF+S | $252 \pm 34 \text{ a}$ | $213 \pm 14 \text{ a}$ | $214 \pm 16 \text{ ab}$ | $224 \pm 19 \text{ a}$ |
| DIF+BC | $271 \pm 14 \text{ a}$ | $225 \pm 33 \text{ a}$ | $246 \pm 7.8 \text{ a}$ | $242 \pm 19 \text{ a}$ |

Data in figure are mean \pm standard error ($n = 3$). Different lowercase letters indicate significant difference amongst drip fertigation (DIF), DIF with straw incorporation (DIF+S) and DIF with biochar incorporation (DIF+BC). $n = 3$, $p < 0.05$.

The annual tomato yields were, on average, $175 \text{ Mg ha}^{-1} \text{ yr}^{-1}$, without significant difference between treatments (Table 3). However, compared with DIF in 2019WS, DIF+BC significantly increased tomato yields and PFP_N by 17.2%. No significant effects were found in the yields or PFP_N of straw incorporation in WS and AW seasons (Table 3). A significant linear relationship was found between PFP_N and $\text{EF}_{\text{Apparent}}$ (Figure 5). $\text{EF}_{\text{Apparent}}$ decreased significantly with increased PFP_N across all three treatments and growing seasons ($p < 0.01$, Figure 5).

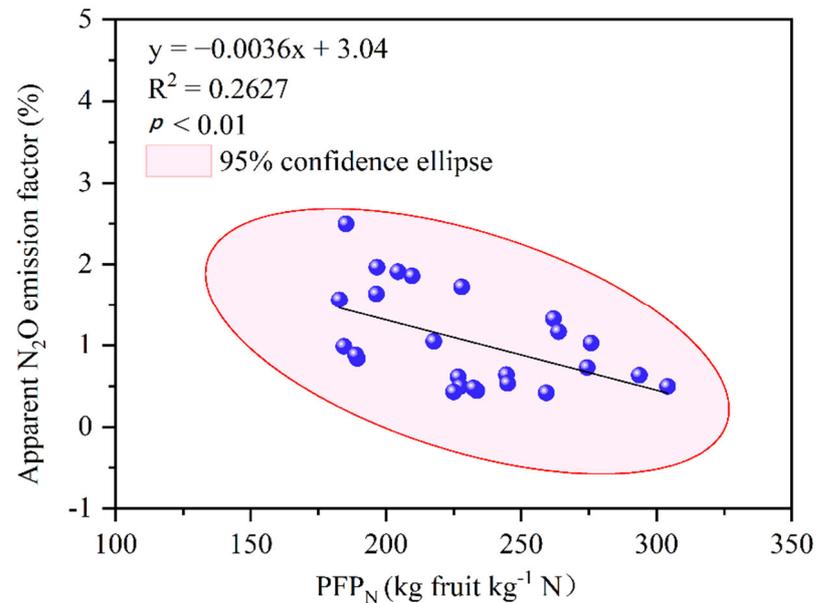


Figure 5. Pearson's correlation between partial factor productivity of applied N fertilizer (PFP_N) and apparent N_2O emission factor ($n = 25$).

3.4. Changes in SOC Content

In January 2016 (i.e., at the start of our experiment), the mean SOC (0–30 cm) concentration was $17.3 \pm 0.45 \text{ g C kg}^{-1} \text{ SDW}$. Resampling of topsoil after 4 years in February 2020 revealed that the SOC contents of all treatments significantly increased, and the differences in SOC amongst different treatments were significant. The highest SOC concentration was observed for DIF+BC ($23.6 \pm 0.40 \text{ g C kg}^{-1} \text{ SDW}$), followed by DIF+S ($21.8 \pm 0.17 \text{ g C kg}^{-1} \text{ SDW}$) and DIF ($20.4 \pm 0.28 \text{ g C kg}^{-1} \text{ SDW}$) in February 2020 (Figure 6a). The calculated annual rates of soil C sequestration were 6.31 ± 0.40 (DIF+BC), 4.49 ± 0.17 (DIF+S) and $3.03 \pm 0.28 \text{ t C ha}^{-1} \text{ yr}^{-1}$ (DIF) (Figure 7a).

On the basis of the calculation of differences between increases in SOC contents for DIF compared with those for DIF+S and DIF+BC, approximately 5.6 or 12.8 t C ha^{-1} remained in the topsoil (0–30 cm), due to straw or BC being incorporated over the eight growing seasons into the soil (DIF+S: $3.5 \text{ t C ha}^{-1} \text{ season}^{-1} \times 8 = 28 \text{ t C ha}^{-1}$ over 4 years; DIF+BC: $2.1 \text{ t ha}^{-1} \text{ season}^{-1} \times 8 = 16.8 \text{ t C ha}^{-1}$ over 4 years). In other words, assuming that the subsoil C content did not change, the C sequestration rates due to additional straw and BC were 20% (DIF+S) and 78% (DIF+BC, Figure 7b), respectively. Besides SOC content, soil $\delta^{13}\text{C}$ changed significantly over the 4 observation years (Figure 6b). DIF+S soil $\delta^{13}\text{C}$ was higher in 2020 than in 2016, but lower soil $\delta^{13}\text{C}$ values were observed for DIF and DIF+BC (Figure 6b).

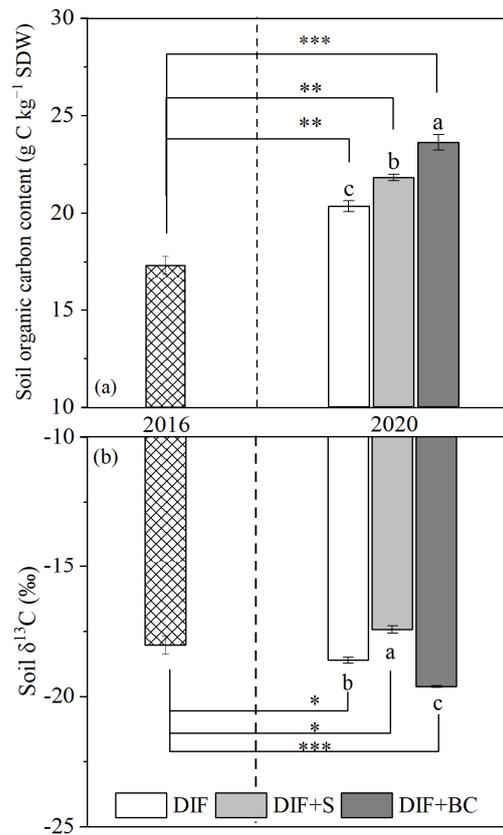


Figure 6. Observed changes in topsoil (0–30 cm) SOC contents (a) and soil organic matter $\delta^{13}\text{C}$ values (b) for different treatments between January 2016 (start of the experiment) and February 2020. DIF: drip fertigation; DIF+S: drip fertigation with straw incorporation; DIF+BC: drip fertigation with biochar incorporation. Different lowercase letters indicate significant difference amongst three treatments at the end of experiment ($p < 0.05$, $n = 3$). *, ** and *** mean significant difference between 2016 and 2020 ($p < 0.05$, $p < 0.01$, $p < 0.001$, $n = 3$).

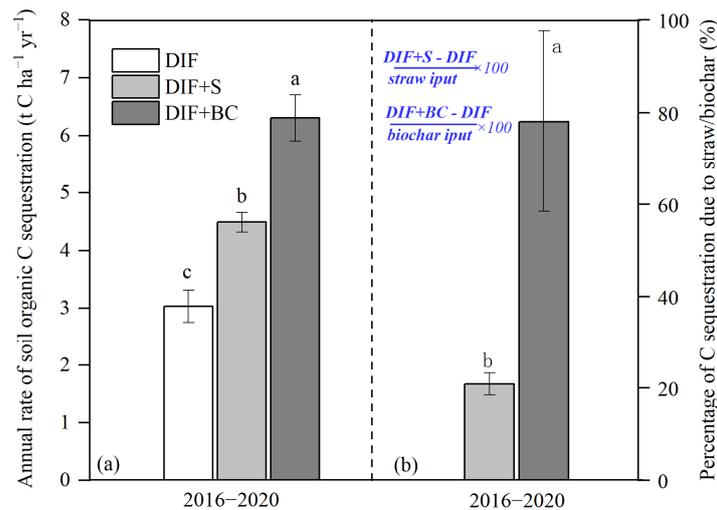


Figure 7. Calculated rates of annual soil C sequestration (a) and contribution of straw and biochar additions to total C sequestration between 2016 to 2020 (b). Different lowercase letters indicate significant difference amongst treatments ($p < 0.05$, $n = 3$). DIF: drip fertigation (DIF); DIF+S: drip fertigation with straw incorporation; DIF+BC: drip fertigation with biochar incorporation. Error bars represent the standard error ($n = 3$).

4. Discussion

Vegetable production in greenhouses is a hotspot of soil N₂O emissions and degradation of soil health is a major concern [5,13]. The negative environmental connotations are mainly due to the way greenhouses are usually managed by farmers, as flood irrigation schemes in combination with high rates of organic and mineral fertilizer applications are common practices [7,8]. In recent years, the use of drip fertigation (DIF) schemes for vegetable production in greenhouses, which allow for reduced rates of irrigation and fertilization, has been strongly promoted. However, the common drip fertigation scheme does not consider the additional application of organic matter, either in the form of straw (DIF+S) or biochar (DIF+BC), even though this may help to significantly increase the soil carbon content of greenhouse soils [18]. The present paper showed that the additional application of straw or biochar was advantageous, as it allowed a further increase in SOC content and decrease in seasonal N₂O emissions but maintained crop yields.

4.1. Magnitude and Interrelationships of Soil N₂O Emissions and N Productivity

The annual soil N₂O emissions ranged from 7.5 to 9.4 kg N ha⁻¹ yr⁻¹ from the investigated greenhouse soils (Figure 2). Although the annual N₂O emissions were much higher than the mean soil N₂O emissions from global arable growing systems (2.4 kg N ha⁻¹ yr⁻¹) [16], they were still significantly lower than the global mean value from greenhouse vegetable systems (12 kg N ha⁻¹ yr⁻¹) [5]. Moreover, the soil N₂O emissions in the present study were nearly one magnitude lower than those in several reports on annual soil N₂O emissions (40–238 kg N ha⁻¹ yr⁻¹) from intensively managed greenhouse vegetable systems with high fertilization (>2000 kg N ha⁻¹ yr⁻¹) and flood irrigation (>1300 mm yr⁻¹) [14,15,51].

Reduced N fertilizer application and lowered soil moisture during the growing season explain the differences in soil N₂O emissions between greenhouse vegetable systems managed by drip fertigation compared with those managed by over fertilization and flood irrigation [8,15,52]. Based on the structure equation modelling analysis, the significant positive direct effect (standardized path coefficients 0.52, $p < 0.001$, Figure 4) of soil NH₄⁺-N concentration on N₂O emissions indicated that the N₂O emissions in this drip fertigation systems were primed by the availability of inorganic N, and by nitrification rather than by coupled nitrification-denitrification [52–54], as soil moisture levels stayed lower. In contrast, N₂O emissions were driven by both nitrification and denitrification in the conventional systems managed with over fertilization and flood irrigation [55]. In addition, the high frequency of drip fertigation practice maintained stable soil WFPS (Figure 1d), and thus reduced N₂O emission pulses caused by soil drying and wetting [54,56], which usually existed in conventional flood irrigation systems with low irrigation frequency. Furthermore, the relative low soil WFPS (60–70%) of drip fertigation reduced N₂O emissions produced by the denitrification process in comparison to conventional flood irrigation [57], although coupled nitrification-denitrification may be favored under such conditions [58–60].

The annual yield-scaled N₂O emissions were in the range of 40.5–56.6 g N₂O-N Mg⁻¹ yr⁻¹, with the lowest values observed for the DIF+BC treatment in the WS growing seasons (Table 3), in line with the mean yield-scaled N₂O emissions for global greenhouse vegetable production systems of 50 ± 10 g N₂O-N Mg⁻¹ yr⁻¹ [5]. No significant differences amongst the treatments were found not only for annual yield-scaled N₂O emissions and EF_{Apparent} but also for tomato yield and PFP_N (Table 3). Meanwhile, EF_{Apparent} significantly decreased with PFP_N (Figure 5), which is in accordance with previous reports [61]. However, the seasonal yield-scaled N₂O emissions and EF_{Apparent} were significantly lower in DIF+BC than in DIF during WS seasons (Table 3). This finding could be partly explained by opposite results (higher yield and PFP_N in DIF+BC than in DIF) and the different pattern of N₂O emission flux related to the change in seasonal environment factors (Figures 1 and 3) [47,62].

4.2. Different Seasonal Responses of N₂O Emissions to Biochar or Straw Incorporation

Compared with DIF, the additional application of biochar to greenhouse soils significantly reduced soil N₂O emissions by approximately 61.2% during the WS growing seasons (2018WS and 2019WS), whereas an opposite effect was observed during the AW season in 2018 (Figures 2 and 3). This result is consistent with previous reports that biochar incorporation could either mitigate N₂O emissions [30,38,62,63] or stimulate N₂O emissions [45,64,65] in agricultural systems. Straw incorporation had less pronounced effects on the mitigation of N₂O emissions than biochar during WS seasons, whereas it induced larger N₂O emissions during the AW season (Figures 2 and 3). These different responses of biochar or straw incorporation were dependent on the properties of organic carbon used, availability of nitrogen and carbon sources and soil temperature and moisture [30,63,66].

During the WS growing seasons, the mitigation effects on N₂O emissions of biochar or straw mainly occurred within 10 days after basal fertigation (Figure 3b), when the soil environment condition was conducive to nitrifier and denitrifier activity [28,52]. Firstly, the significant reduction in N₂O emission fluxes with biochar incorporation compared with DIF is related to its large specific surface and porosity, meaning lower availability of NH₄⁺-N and dissolved organic carbon for microbial nitrification and denitrification [38,39]. Secondly, a high C/N ratio of biochar and straw stimulates microbial N immobilization and reduces ammonium and nitrate concentration for N₂O production [48,67,68]. Thirdly, nitrification and denitrification were mitigated by limited biological available carbon [58] with biochar incorporation versus straw incorporation, due to its recalcitrant structure (Figure 7) [32,66]. Moreover, the emission of volatile organic compounds from biochar might show a toxic effect on soil microorganisms, and thus decrease the nitrification rate and N₂O emissions [69,70].

Oppositely, several days after basal fertigation in the AW growing season, the increase in daily N₂O flux induced by straw incorporation was significantly higher than that with biochar (Figure 3b; Δ (DIF+S–DIF): 953–498, Δ (DIF+BC–DIF): 8.7–255 g N ha⁻¹ d⁻¹), resulting in higher seasonal N₂O emissions in DIF+S than in DIF+BC (Figure 2b). This finding was closely related to the soil carbon and nitrogen cycling caused by the high soil temperature and moisture at the beginning of the AW growing season (Figures 1c and 4) [64,71]. On the one hand, the higher soil temperature (>28 °C) and moisture stimulated the decomposition of chicken manure and soil organic nitrogen, and thus increased soil NH₄⁺-N substrates for microbial nitrification (Figure 1f) [71,72]. This finding could be confirmed by the high pathway coefficient between N₂O emissions and soil NH₄⁺-N concentration (Figure 4). On the other hand, the soil denitrification process was mainly controlled by soil anaerobic conditions, sufficient nitrogen substrates, and organic carbon sources [58]. The high soil temperature during this period stimulated soil microbe activity and resulted in the increased consumption of soil oxygen when straw was incorporated, leading to a rapid reduction in soil redox potential and increased N₂O emissions produced by the denitrification process [55,73,74]. Moreover, the intensity of the denitrification process was influenced by the fraction of C and C/N ratios of organic matters [48,61,75]. Compared with biochar with condensed aromatic C [76,77], straw contained easily decomposable C fractions, which could strongly promote N₂O emissions by soil denitrification [55,75].

4.3. Changes in Rates of Soil C Sequestration Due to Additional C Inputs

Over the 4-year observation period, annual increases were found in topsoil SOC content in the range of 3.03 (DIF)–6.31 (DIF+BC) t C ha⁻¹ yr⁻¹ for all treatments (Figure 7a). These can be confirmed by the change in SOC and by the soil $\delta^{13}\text{C}$ signature (Figure 6). By contrast, no significant increase in SOC concentration was found in a parallel experiment, where tomatoes were grown by applying farmers' conventional management of flood irrigation with over-fertilization [18,78]. Thus, drip fertigation may generally improve soil C sequestration even for DIF treatment solely originating from the added chicken manure, which was used as a basal fertilizer. Moreover, the input of additional exogenous organic material, maize straw or biochar, in the present study increased soil C sequestration,

similar to the results of other studies (Figure 6) [27,79–81]. With regard to increasing soil C content, the most effective was the addition of biochar, as 78% of the added biochar carbon was finally found in the topsoil. By contrast, only 20% of the added straw-C remained in the topsoil (Figure 7b), whilst the remaining 80% was likely mineralized and respired. These observations were consistent with the results of recent meta-analyses on the effect of long-term straw return on SOC storage and sequestration in arable soils in China and India [82,83]. The much higher C sequestration rates for the DIF+BC treatment in the present study may be explained by the high stability of biochar, which strongly hampers its decomposition (Figure 6) [33,84,85].

5. Conclusions

This study showed that drip fertigation with the additional application of organic amendments in the form of straw or biochar is a suitable management option for greenhouse vegetable production. This approach significantly reduced irrigation water and fertilizer inputs, compared with farmers' current conventional practice of flood irrigation with over-fertilization. The tested method resulted in high rates of soil carbon sequestration and reduced seasonal N₂O emissions. This effect was most pronounced for biochar addition, whereas the effects of added straw on C sequestration rates and soil N₂O emissions were lower. The different seasonal response of N₂O emission induced by straw or biochar incorporation should be given further attention to optimize the carbon and nitrogen management of greenhouse vegetable soils. As many greenhouse vegetable production systems experience low indoor atmospheric CO₂ concentrations during midday in the AW season, the combined application of straw and biochar is likely to be the best option for decreasing soil N₂O emissions and increasing SOC stocks and soil respiration to favor crop growth.

Supplementary Materials: The following supporting information can be downloaded at: <https://www.mdpi.com/article/10.3390/agronomy12071661/s1>, Figure S1: Pearson's correlation between daily N₂O emissions and daily mean values of soil temperature, soil water-filled pore space (WFPS) and soil mineral N concentration (NO₃⁻-N and NH₄⁺-N); Table S1: Growth duration; transplanting and final harvest dates of tomato cultivation; amount of mineral nitrogen, phosphate, potassium and organic fertiliser application; number of irrigation events and total amounts of irrigation water for different seasons in drip fertigation systems.

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