Dynamic responses of nitrous oxide emission and nitrogen use efficiency to nitrogen and biochar amendment in an intensified vegetable field in southeastern China

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Abstract

Intensive vegetable production exhibits contrasting characteristics of high nitrous oxide (N$_2$O) emissions and low nitrogen use efficiency (NUE). In an effort to mitigate N$_2$O emissions and improve NUE, a field experiment with nine consecutive vegetable crops was designed to study the combined effects of nitrogen (N) and biochar amendment and their interaction on soil properties, N$_2$O emission and NUE in an intensified vegetable field in southeastern China. We found that N application significantly increased N$_2$O emissions, N$_2$O–N emission factors and yield-scaled N$_2$O emissions by 51–159%, 9–125% and 14–131%, respectively. Moreover, high N input significantly decreased N partial factor productivity (PFPN) and even yield during the seventh to ninth vegetable crops along with obvious soil degradation and mineral N accumulation. To the contrary, biochar amendment resulted in significant decreases in cumulative N$_2$O emissions, N$_2$O–N emission factor and yield-scaled N$_2$O emissions by 5–39%, 16–67% and 14–53%, respectively. In addition, biochar significantly increased yield, PFPN and apparent recovery of N (ARN). Although without obvious influence during the first to fourth vegetable crops, biochar amendment mitigated N$_2$O emissions during the fifth to ninth vegetable crops. The relative effects of biochar amendments were reduced with increasing N application rate. Hence, while high N input produced adverse consequences such as mineral N accumulation and soil degradation in the vegetable field, biochar amendment can be a beneficial agricultural strategy to mitigate N$_2$O emissions and improve NUE and soil quality in vegetable field.

Keywords: biochar, intensified vegetable field, N$_2$O emissions, nitrogen use efficiency, soil quality

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Introduction

In China, vegetable crops are produced on an area of about 24.7 × 10$^6$ hectare (ha), equivalent to 12.4% of the total cropping area. The production represented 52% of the world vegetable production in 2012 (FAO, 2015). The Chinese vegetable production is characterized by multiple annual cropping sequences, high nitrogen (N) application rates and frequent management practices. As a result, the intensified vegetable fields usually had low nitrogen use efficiency (NUE; Ju et al., 2009) and high nitrous oxide (N$_2$O) emissions (Xiong et al., 2006; He et al., 2009; Jia et al., 2012; Zhang et al., 2015a) as compared to other croplands. Moreover, long-term high N input results in negative effects including soil acidification and soil nitrate accumulation on agriculture soil (Guo et al., 2010; Zhang et al., 2015b), which have a feedback on the N transformation processes in vegetable fields (Zhu et al., 2011; Cheng et al., 2015). Therefore, agricultural management strategies must be optimized to meet the joint challenges of high production and acceptable environmental consequences (Zhang et al., 2013) and thus to achieve the improvement in NUE and soil quality in intensified vegetable agriculture.

The estimated direct N$_2$O emissions from vegetable cultivation account for approximately 20% of the Chinese cropland emissions (Zheng et al., 2004; Wang et al., 2011). N$_2$O is a potent and long-lived atmospheric greenhouse gas (GHG), with a strong contribution to the annual increase in radiative forcing in the atmosphere, that is, 296 times stronger than CO$_2$ (IPCC, 2013). New practices are needed to balance GHG sources and sinks while not impairing the capacity of the ecosystems to ensure food security. Therefore, improvements of yield-scaled N$_2$O emissions approach should be pursued as a more comprehensive index to
assess N2O emissions in agricultural ecosystems (Van Groenigen et al., 2010).

There are a variety of new management practices and technologies that can promote crop productivity and reduce N losses and thus achieving a higher NUE and lower yield-scaled N2O emissions (Chien et al., 2009). Chen et al. (2014) demonstrated that substantially increased yields can be achieved with reduced inputs of N fertilizer and so lower environmental costs by a set of integrated soil-crop system management practices based on a modern understanding of crop ecophysiology and soil biogeochemistry. However, a nonlinear (accelerating) relationship between N2O emissions and increasing N inputs was observed for a wide range of N inputs (Zhang et al., 2015b), thus indicating a disproportionately high risk of N2O emissions in vegetable production systems, where up to 1217 kg N ha⁻¹ yr⁻¹ may be applied (Li et al., 2015b). Considering both environmental issues and economic profits, reasonable N application rate and reduction should be given high priority in intensified vegetable production.

Biochar, made by pyrolysis or gasification of biomass residues, is a recently promoted soil amendment that may contribute to soil organic carbon (SOC) sequestration, improved soil quality and agronomic benefits (Lehmann et al., 2011). Recent reviews have highlighted the benefits of adding biochar to agricultural soils, including the improvement of soil water-holding capacity (Laird et al., 2010b), a pH amelioration of acidic soils (liming effect) and reduction in nutrient leaching losses (Laird et al., 2010a). Moreover, a recent meta-analysis of 30 studies (published from 2007 to 2013) revealed a statistically significant reduction of 54% in N2O emissions when soils were amended with biochar (Cayuela et al., 2014). Moreover, the effects of biochar amendment on N2O emissions from acidic vegetable field (Wang et al., 2015a) seem to be distinct as compared to other croplands (Cayuela et al., 2014) since the surplus N inputs probably mask the beneficial effect of biochar addition on N transformation, such as inorganic N absorption and microbial NH₄ immobilization (Clough et al., 2010). So far, however, as limited by trial temporal extent and crop species, N2O emissions and NUE from biochar-amended agricultural ecosystems were typically estimated on a whole-trial scale (Zhang et al., 2010, 2015b; Li et al., 2015a), complicating mechanistic insight into the dynamic effects of N and biochar amendment in agricultural ecosystems.

Therefore, the objectives of the present study were to quantify the time course (dynamic) effects of N fertilization and biochar amendment on soil properties, N2O emissions and NUE in intensive vegetable production systems. The study was done during nine consecutive vegetable cropping sequences in an intensified vegetable field in southeastern China.

Materials and methods

Study site and biochar properties

The field experiment was conducted at a suburban site (31°59′N, 118°51′E) in Nanjing, Jiangsu Province, China, from April 12, 2012, to October 29, 2014. This area has a subtropical monsoon climate with a mean annual rainfall of 1107 mm and an annual mean air temperature of 15.3 °C according to Nanjing weather station. The selected site had been conventionally cultivated with vegetables for approximately 10 years and is a typical vegetable field. The studied soil is classified as Fimi-Orthic Anthrosols (CRGCST, 2001), with a bulk density of 1.2 g cm⁻³, a total porosity of 51%, a clay (<0.002 mm diameter) fraction of 30.1%, a silt (0.002-0.02 mm diameter) fraction of 64.7%, and a sand (0.02-2 mm diameter) fraction of 5.2%. The main properties of this soil are as follows: pH, 5.5; total N, 1.9 g kg⁻¹; SOC, 15.6 g C kg⁻¹; and cation exchange capacity (CEC), 31.2 cmol kg⁻¹.

For the field experiment, biochar was produced from wheat straw at the Sanli New Energy Company in Henan, China, by pyrolysis and thermal decomposition at 400 °C. The biochar had a carbon content of 467 g C kg⁻¹ and a N content of 5.9 g N kg⁻¹. The initial values of pH, CEC and ash content were 9.4, 24.1 cmol kg⁻¹ and 20.8%, respectively.

Treatments and vegetable management

A 3² factorial design was conducted in triplicate for nine consecutive vegetable crops from April 2012 to October 2014. The experimental plot area of each treatment was 7.5 m² (3 × 2.5 m). The biochar mass, which was originally in a particulate form, was plowed at a depth of 20 cm topsoil to the vegetable fields before sowing on April 8, 2012, and only one biochar application was made in this study. Biochar was applied at rates of 0 t (C0), 20 t (C1) and 40 t (C2) ha⁻¹. N fertilizer was applied at zero (N0), at the conventional application rate of 1233 kg N ha⁻¹ yr⁻¹ (N1) and at 4/3 of the conventional application rate of 1644 kg N ha⁻¹ yr⁻¹ (N2), the typical overdose of fertilization in vegetable production. For conventional N fertilization, compound fertilizer with a m(N) : m (P₂O₅) : m(K₂O) ratio of 15 : 15 : 15 was applied at a rate of 313 kg N ha⁻¹ for Amaranth (Amaranthus mangostanus L.) and Coriander (Coriandrum sativum L.), 600 kg N ha⁻¹ for Tung choy (Ipomoea aquatic Forssk.) and 250 kg N ha⁻¹ for Baby bok choy (Brassica chinensis L.) and Spinach (Spinacia oleracea L.), according to local farmers’ practice (Table 1). All fertilization with the form of ammonium-based fertilizer was broadcasted before transplanting and as the base fertilizer for each vegetable crop except for the Tung choy crop, which had 313 kg N ha⁻¹ as basal fertilization and 287 kg N ha⁻¹ as top dressing for the N1 rate. The treatments without N fertilization (the N0 group treatments) were applied with corresponding P and K fertilizers, which were broadcast in the form of calcium phosphate and potassium chloride, respectively, and all
treatments were amended with equal amounts of P and K fertilizers.

There were nine consecutive vegetable crops grown successively during the entire observation period. Each type of vegetable was seeded by hand and all management procedures, including crop species, irrigation and harvest time, followed the local farmers’ practices (Table 1). Furthermore, a short fallow period was imposed after the fresh biomass was harvested of each vegetable crop season. Soon after harvesting each vegetable crop, the field was tilled to a depth of 12–15 cm. A protective plastic film, 2.3 m in height, was used to cover the crops, these are, from April 12, 2012, to May 25, 2012, and March 15, 2014, to May 12, 2014, for Amaranth and from November 20, 2012, to February 24, 2013, and November 5, 2013, to March 14, 2014, for Baby bok choy, as Amaranth and Baby bok choy require relatively warm weather conditions for growth.

Measurements of N$_2$O fluxes, soil samples and environmental factors

A static opaque chamber method was used to collect air samples from three replicates for each treatment. Each chamber was made of PVC and consisted of a chamber body (50 × 50 × 50 cm$^3$). The outside of the chamber was coated with sponge and aluminum foil to prevent solar radiation heating the chamber. During each measurement of the GHGs, the chamber was placed on the prefixed frame in each plot, covered both the soil and the vegetable crops, and the frame was inserted 0.1 m into the soil. Sampling was conducted between 8:30 and 10:30 in the morning every other day for 1 week after fertilizer application and then once per week thereafter. The gas fluxes were measured on 121 occasions over the whole observation period. On each sampling occasion, the air samples were taken 0, 10, 20 and 30 min after chamber closure. The soil temperature at a depth of 10 cm beneath the collection point was measured by a manual soil thermometer with a sensor of silicon semiconductor when the gas samples were collected. The samples, collected in 20-mL syringes, were returned to the laboratory, and N$_2$O was determined on the same day with a gas chromatograph (Agilent 7890A; Agilent Ltd, Shanghai, China) equipped with an electron capture detector (ECD). The carrier gas was argon–methane (5%) at a flow rate of 40 mL min$^{-1}$. The column (SS-2 m × 2 mm Porapak Q [80/100]) and ECD temperatures were maintained at 40 and 300 °C, respectively. The concentrations of N$_2$O were quantified by comparing the peak area with those of reference gases (Nanjing special gas factory, Nanjing, China). N$_2$O fluxes were calculated by using the linear increases in gas concentration over time. The measured N$_2$O fluxes were weighted by the interval between two measurements (Xiong et al., 2006). The seasonal mean N$_2$O flux for each vegetable crop was calculated as the time-weighted average flux. Cumulative seasonal N$_2$O was calculated as the product of the mean flux and the seasonal duration.

In addition to the soil that was analyzed immediately before the experiment in April 2012, another batch of soil samples for each treatment was collected shortly after the last
crop harvest on October 30, 2014, and stored at −20 °C for laboratory analysis. In accordance with Lu (2000), soil texture was measured using pipette analysis, SOC was analyzed by wet digestion with H2SO4-K2Cr2O7, and TN was determined by semi-micro Kjeldahl digestion using Se, CuSO4 and K2SO4 as catalysts. Soil pH was measured at a volume ratio of 1 : 2.5 (soil to water ratio), while the biochar pH was measured for a 1 : 5 char/water suspension using a PHS-3C mv/pH detector (Shanghai, China). Electric conductivity (EC) was measured using a Mettler-Toledo instrument (FE30-K; Shanghai, China) at a 1 : 5 (w : v) soil to water ratio. CEC was determined by the BaCl2 (0.1 M, 20 mL for 2 g soil) compulsive exchange method.

Simultaneously with the determination of the trace gas fluxes, soil sampling at 0–10 cm depth was conducted for the determination of soil mineral N and soil water content. The soil NH4+ – N and NO3− – N contents were extracted by shaking for one hour on a rotary shaker with 2 mol L−1 KCl solution. According to Lu (2000), soil NH4+ – N and NO3− – N contents were measured following the two-wavelength ultraviolet spectrometry and indophenol blue methods, respectively, using an ultraviolet spectrophotometer (U-2900; HITACHI, Tokyo, Japan). The water-filled pore space (WFPS) was obtained by converting soil moisture content from the oven-drying method and soil bulk density from the cutting ring method to 0–10 cm depth by assuming soil particle density of 2.7 g cm−3.

Vegetable yield and aboveground N uptake
After each vegetable crop reached physiological maturity, vegetable yields were measured by weighing all of the aboveground vegetable parts that were grown in each plot as fresh weight. Then, N uptake was calculated from the sum of the N masses harvested in the biomass from each plot. The vegetable biomasses were air-dried, then further dried for 3 days at 65 °C and weighed to obtain the dry matter yields. Subsamples were ground with a ball mill and analyzed for N concentration with an elemental N analyzer (Foss KT260, Suzhou, China).

Estimation of N2O emission factors, yield-scaled N2O emissions, N partial factor productivity and apparent recovery of N
The N2O emission factors were calculated as the amount of fertilization N2O–N emissions minus the emissions without fertilization (background N2O emissions) as a percentage of the fertilizer N applied for a specific period.

The yield-scaled N2O emissions were related to yield as in Van Groenigen et al. (2010) and were calculated as the amount of cumulative N2O emissions per fresh vegetable yield.

Here, we calculated NUE in two different ways simultaneously as N partial factor productivity (PFPN) and apparent recovery of N (ARN). PFPN was calculated as the fresh crop yield per the amount of the fertilizer N applied (kg kg−1), and ARN was calculated by dividing the differences in the N amount in the aboveground biomass between the N-fertilized plots and the control plots under the same rate of biochar amendment.

Data processing and statistics
All figures in this study were plotted using SigmaPlot 11.0 (Systat Software Inc., San Jose, CA, USA). A two-way ANOVA was used to analyze the effects of N, biochar and their interactions on soil properties, soil mineral N, vegetable yield, N2O emissions, N2O emission factors, yield-scaled N2O emissions, PFPN and ARN. A Tukey’s multiple range test was used to determine whether significant differences occurred among the treatment means at a significance level of 0.05 using JMP v. 7.0 (SAS Institute, Cary, NC, USA, 2007). The effects of the main driving factors on N2O emissions were determined through the pairwise correlation and partial correlation analysis using SPSS 16.0 (SPSS China, Beijing, China).

Results

Soil properties

Compared with the treatments without N addition, N application resulted in significant increases in soil TN, soil EC and bulk density at the same biochar amendment rates by 2–25%, 17–241% and 1–9%, respectively (Table 2, P < 0.01). Biochar amendment significantly increased soil TN and SOC by 1–17% and 4–48%, respectively, while significantly decreased soil pH, EC and bulk density under equal N amounts in relation to the treatments without biochar (Table 2, P < 0.01). However, biochar amendment significantly increased the soil pH by 0.11–0.23 units without N application (Table 2, P < 0.01), while significantly decreasing the soil pH by 0.02–0.23 units in the N-fertilized treatments (Table 2, P < 0.01). This shows the significant interactions between the N and biochar amendment on soil pH (Table 2, P < 0.01). Moreover, N application decreased SOC by 8–14% in the C0 group treatments but significantly increased SOC by 7–23% in the C1 and C2 group treatments (Table 2, P < 0.05), indicating that biochar amendment can alleviate SOC decreasing effects caused by N application. Besides, significant interactions on soil TN, EC and bulk density were observed (Table 2, P < 0.05).

Soil conditions and mineral N content dynamics

Figure 1a shows temporal variations on soil WFPS and temperature at the 10 cm depth in the vegetable field. The soil temperature, ranging from 3 to 29 °C, was detected with seasonal changes of the outside temperature, although the plastic film was in place sometimes in low-temperature seasons. In addition, soil WFPS values varied from 32% to 76% due to heavy rainfall or irrigation events.

As shown in Figs 2 and 3, soil NH4+ – N and NO3− – N contents ranged from 36 to 414 mg N kg−1...
Table 2 Soil total nitrogen (TN), soil organic carbon (SOC), soil pH, electric conductivity (EC) and bulk density affected by different treatments over the entire experimental period

| Treatments† | TN (g N kg\(^{-1}\)) | SOC (g C kg\(^{-1}\)) | pH | EC (ds m\(^{-1}\)) | Bulk density (g cm\(^{-2}\)) |
|-------------|-----------------------|------------------------|-----|---------------------|----------------------------|
| N0C0        | 1.49 ± 0.04 e\(\dagger\) | 14.7 ± 0.3 d           | 5.05 ± 0.01 c | 0.09 ± 0.01 e       | 1.24 ± 0.01 c             |
| N0C1        | 1.51 ± 0.09 e           | 15.3 ± 0.1 d           | 5.16 ± 0.03 b | 0.07 ± 0.01 f       | 1.20 ± 0.01 d             |
| N0C2        | 1.52 ± 0.05 e           | 17.1 ± 0.8 bc          | 5.28 ± 0.03 a | 0.06 ± 0.01 g       | 1.18 ± 0.01 d             |
| N1C0        | 1.60 ± 0.04 d           | 12.7 ± 0.3 e           | 4.37 ± 0.01 d | 0.25 ± 0.01 a       | 1.31 ± 0.02 a             |
| N1C1        | 1.70 ± 0.02 c           | 18.1 ± 0.2 ab          | 4.14 ± 0.04 ef| 0.20 ± 0.01 d       | 1.29 ± 0.01 b             |
| N1C2        | 1.79 ± 0.04 b           | 18.8 ± 0.5 a           | 4.18 ± 0.01 e | 0.22 ± 0.01 c       | 1.29 ± 0.03 b             |
| N2C0        | 1.63 ± 0.01 cd          | 13.6 ± 0.4 e           | 4.11 ± 0.01 fg| 0.24 ± 0.01 b       | 1.30 ± 0.02 ab            |
| N2C1        | 1.66 ± 0.01 cd          | 16.5 ± 0.5 c           | 4.04 ± 0.01 h | 0.24 ± 0.01 b       | 1.30 ± 0.01 ab            |
| N2C2        | 1.90 ± 0.05 a           | 16.7 ± 1.3 c           | 4.09 ± 0.01 g | 0.22 ± 0.01 c       | 1.29 ± 0.01 ab            |

ANOVA results

\(N\) *** ** *** *** ***
\(Bc\) *** *** *** ***
\(Bc\times N\) *** *** *** ***

†N0: without N fertilization, N1: conventional N fertilization rate, N2: 4/3 conventional fertilization rate. C0: without biochar amendment, C1: 20 t ha\(^{-1}\) biochar amendment, C2: 40 t ha\(^{-1}\) biochar amendment.
\(\dagger\) Means ± SD with different letters in the same column indicate significant differences according to the Tukey’s multiple range test (\(P < 0.05\)) among all treatments.

***Significant at \(P < 0.001\); **significant at \(P < 0.01\); *significant at \(P < 0.05\); n.s. not significant.

and 11 to 292 mg N kg\(^{-1}\), respectively. N fertilization events considerably increased both soil NH\(_4\)\(^+\) – N and NO\(_3\) – N contents (Fig. 2, 3, \(P < 0.01\)). However, biochar amendment had significant effect on neither soil NH\(_4\)\(^+\) – N nor NO\(_3\) – N content. Moreover, the partial correlation analysis showed that both soil NH\(_4\)\(^+\) – N (\(P < 0.05\)) and NO\(_3\) – N (\(P < 0.01\)) contents were correlated with soil WFPS (Table 3).

\(N_2O\) emission, \(N_2O\)-N emission factor and yield-scaled \(N_2O\) emissions

Nitrous oxide fluxes from all treatments were relatively consistent and followed a sporadic and pulse-like pattern (Fig. 1b–d). Following basal fertilization, temperature increase, tillage or irrigation events, peaks in \(N_2O\) emissions were observed, and the highest \(N_2O\) flux was 4566 μg N m\(^{-2}\) h\(^{-1}\), which occurred in the N1C0 treatment in the ninth vegetable cropping season (Fig. 1c). \(N_2O\) fluxes were mainly observed during the time when the soil temperature was higher (from May to October), and no significant increases in \(N_2O\) fluxes were observed following the supplemental fertilization events coupled with irrigation in the fourth and seventh vegetable crop seasons (Fig. 1), which may be due to the lower soil temperature (Fig. 1a). Moreover, the partial correlation analysis showed that the natural logarithms of \(N_2O\) fluxes were correlated with the soil temperature and WFPS in all treatments (Table 3, \(P < 0.01\)).

Total cumulative \(N_2O\) emissions show appreciable differences among all treatments (Fig. 4, \(P < 0.05\)). The highest total cumulative \(N_2O\) was 129 kg N ha\(^{-1}\) in the N2C0 treatment, which was 159% higher than that of the N0C0 treatment. Significant increases in cumulative \(N_2O\) emissions induced by N application were observed by 51–159% under the same rate of biochar amendment related to the treatments without N (Fig. 4, \(P < 0.01\)). To the contrary, biochar amendment significantly decreased cumulative \(N_2O\) emissions by 4–39% in the N-fertilized treatments related to the treatments without biochar (Fig. 4, \(P < 0.01\)), while increased \(N_2O\) emissions by 5–18% in the treatments without N were not statistically significant (Fig. 4). In addition, biochar amendment resulted in relative reduction in cumulative \(N_2O\) emissions by 28% and 22% in the N1 and N2 group treatments, respectively, showing that the relative reduction induced by biochar amendment decreased with increasing N application rate (Fig. 4). However, two-way ANOVA shows that the mitigation effects of biochar on \(N_2O\) emissions in individual vegetable crop seasons were different in our study (Table 4). Biochar amendments had no significant influences on \(N_2O\) emissions during the first to fourth crop seasons but significantly decreased cumulative \(N_2O\) emissions during the fifth to ninth crop seasons (Table 4, \(P < 0.05\)) and thus had an overall positive effect on \(N_2O\) mitigation (Fig. 4, \(P < 0.01\)).

Direct \(N_2O\)-N emission factors ranged from 0.6% to 1.9% and showed obvious variations (Fig. 4, \(P < 0.05\)). Compared with the treatments without N addition, N application significantly increased \(N_2O\)-N emission factor by 9–125% under the same rate of biochar...
amendment (Fig. 4, $P < 0.01$). When biochar was applied, N$_2$O–N emission factor decreased significantly by 16–67% under equal N application amount (Fig. 4, $P < 0.05$). In addition, relative decreases in N$_2$O–N emission factor by biochar amendment decreased with the increasing N application rate (Fig. 4).

Yield-scaled N$_2$O emissions ranged from 0.15 to 0.36 kg N$_2$O–N t$^{-1}$ over the entire period (Fig. 4). Compared with the treatments without N, significant increases in yield-scaled N$_2$O emissions induced by N application were detected by 14–131% with the same biochar rates (Fig. 4, $P < 0.01$). To the contrary, biochar amendment significantly decreased yield-scaled N$_2$O emissions by 14–53% in the N-fertilized treatments (Fig. 4, $P < 0.01$) while had no significant influence in the treatments without N, indicating a mitigation effect of biochar amendment when combined with N application. However, relative reduction in yield-scaled N$_2$O emissions induced by biochar amendment decreased with the increasing N rate (Fig. 4).

Fig. 1 Dynamics of WFPS, soil temperature (a) and soil N$_2$O emissions fluxes (b–d) under different treatments in intensified vegetable field. The horizontal dashed line in (a) indicates the field capacity (52%), and the vertical dashed lines in (b–d) separated different vegetable crop seasons. The solid and dashed arrows in (b) indicate basal fertilization and top dressing, respectively, in N-fertilized plots. The bars indicate the standard error of the mean (+SE) for the three replicates of each treatment. See Table 2 for treatment codes. The data for WFPS, soil temperature and N$_2$O fluxes during the first to fourth vegetable crops from April 12, 2012, to May 25, 2013, were redrawn from Li et al. (2015a).
Vegetable yield and NUE (PFPN and ARN)

The total fresh vegetable yields were obviously different among the treatments (Fig. 4, \( P < 0.05 \)). The highest total yield was 476 t ha\(^{-1}\) for the N1C2 treatment, which was 50% higher than that of the N0C0 treatment (Fig. 4). Although appreciable differences were observed in total vegetable yield (Fig. 4, \( P < 0.05 \)), that of individual vegetable crop seasons showed different results (Table 5). N application significantly increased the vegetable yield during the first to sixth crop seasons but significantly decreased during the seventh to ninth crop seasons in relation to the treatments without N. In addition, biochar amendments significantly increased vegetable yield during the first to fifth and the eighth to ninth crop seasons (Table 5, \( P < 0.05 \)) but had no significant effects during the sixth and seventh crop seasons. Moreover, the relative increases induced by biochar amendment on vegetable yields were 31% and 18% for N1 and N2 group treatments, respectively. It shows that the relative increases induced by biochar amendment in vegetable yield decreased with the increasing application N rate (Fig. 4).

Along with the increases in total vegetable yield, biochar amendment significantly increased PFPN by 9–44% under equal N amounts as compared with the treatments without biochar (Fig. 5a, \( P < 0.01 \)). The relative increases in PFPN induced by biochar amendment decreased by 33% and 18% in the N1 and N2 group treatments (Fig. 5), respectively, which attributed to the significant decreases in vegetable yield induced by increasing N application rate (Fig. 4, \( P < 0.01 \)). The ARN of each treatment was low and ranged from 0.4% to 12.6% in our study (Fig. 5b). Biochar amendment
significantly increased ARN (Fig. 5b, \(P < 0.01\)), while N application decreased ARN when biochar was applied (not statistically significant, \(P > 0.05\)), showing that biochar was less effective on the improvement of NUE (PFPN and ARN) when more N was applied. In addition, significant interactions between N application and biochar amendment were observed to affect total cumulative \(\text{N}_2\text{O}\) emissions (\(P < 0.01\)), direct \(\text{N}_2\text{O}\)-N emission factor (\(P < 0.01\)), yield-scaled \(\text{N}_2\text{O}\) emissions (\(P < 0.01\)), vegetable yield (\(P < 0.01\)) and PFPN (Fig. 5a, \(P < 0.05\)), while no significant effect on ARN (Fig. 5b) was observed in our study.

**Discussion**

*Dynamic effects of N application on \(\text{N}_2\text{O}\) emissions, crop yields and NUE in vegetable fields*

In our study, N application significantly increased total cumulative \(\text{N}_2\text{O}\) emissions in nine consecutive vegetable growing periods in relation to the treatments without N
(Fig. 4, P < 0.01), which agreed well with previous studies (IPCC, 2013; Zhang et al., 2015b). This was supported by the high soil NH\textsubscript{4}\textsuperscript{+} – N and NO\textsubscript{3}\textsuperscript{−} – N concentrations under high N inputs in the vegetable soils (Figs 2 and 3). In our study, N\textsubscript{2}O–N emission factors ranged from 0.6% to 1.9% with an average value of 1.2%, which is higher than most previous results (Wang et al., 2011) and also the current IPCC default value (IPCC, 2013). We expect that this is because the N\textsubscript{2}O/N\textsubscript{2} ratio increased as a consequence of N\textsubscript{2}O reductase inhibition due to acidification, resulting in the increasing N\textsubscript{2}O emissions from denitrification (Saggar et al., 2013; Qu et al., 2014). In addition, the natural logarithms of N\textsubscript{2}O fluxes were correlated with soil temperature and WFPS in all treatments (Table 3, P < 0.01). Higher temperature caused frequent changes in soil WFPS (Fig. 1a) which lead to the variations in N\textsubscript{2}O emission between different crop seasons (Dobbie & Smith, 2001).

Vegetable yields were strongly affected by N application in this field trial (Fig. 4 and Table 5). N fertilization significantly increased total vegetable yield as compared to the treatments without N application (Fig. 4, P < 0.01). However, the effects on individual vegetable crops were different (Table 5). Increases in the EC and soil mineral N content induced by high N application rates may explain the lack of response to N addition in crop yield during the late experimental period (Table 2, Figs 2 and 3). Moreover, N application resulted in soil acidification even in the treatments amended with biochar (Table 2) also explained the decreases in vegetable yield.

N application significantly decreased PFPN (P < 0.01) while had no significant effect on ARN in the vegetable field (Fig. 5). However, ARN ranged from 0.4% to 12.6% in our study (Fig. 5b), and the lowest one was 0.4% in the N1C0 treatment, which was much lower than NUE values in other croplands (Zhang et al., 2008). This may be explained by several reasons. First, intensified vegetable cropping system always has a large amount of N input to obtain the maximum economic profits (Li et al., 2015a), which resulted in soil
Table 4  Cumulative N\textsubscript{2}O emissions of individual vegetable crop season under different treatments in intensified vegetable field

| Treatments | 1st  | 2nd  | 3rd  | 4th  | 5th  | 6th  | 7th  | 8th  | 9th  |
|------------|------|------|------|------|------|------|------|------|------|
|            | 2012/4/12- | 2012/7/11 | 2012/11/20 | 2013/3/28 | 2013/7/1 | 2013/11/5 | 2014/3/16 | 2014/6/23 | 2014/8/12 |
|            | – | – | – | – | – | – | – | – | – |
|            | 2012/7/10 – | 2012/11/19 – | 2013/3/27 – | 2013/6/30 – | 2013/11/4 – | 2014/3/15 – | 2014/6/22 – | 2014/8/11 – | 2014/10/29 – |
| (41 days/ | (100 days/ | (98 days/ | (62 days/ | (106 days/ | (67 days/ | (58 days/ | (21 days/ | (39 days/ |
| 90 days)† | 131 days) | 127 days) | 94 days) | 126 days) | 129 days) | 99 days) | 49 days) | 78 days) |

N0C0      | 5.7 ± 1.2 c† | 12.6 ± 3.5 e | 0.6 ± 0.2 c | 3.1 ± 1.9 b | 37 ± 0.5 e | 0.4 ± 0.1 c | 4.9 ± 2.4 c | 12.4 ± 3.3 c | 38 ± 0.4 c |
N0C1      | 8.3 ± 1.0 b  | 12.7 ± 2.0 e | 0.7 ± 0.1 c | 2.6 ± 0.2 b | 6.8 ± 1.3 de | 0.6 ± 0.1 b | 4.9 ± 0.7 c | 10.9 ± 1.2 cd | 97 ± 2.6 b |
N0C2      | 9.9 ± 0.6 ab | 12.7 ± 1.4 e | 1.0 ± 0.2 bc | 2.8 ± 1.1 b | 49 ± 0.5 de | 0.4 ± 0.1 c | 4.9 ± 1.7 c | 8.5 ± 1.8 d  | 57 ± 1.1 c |
N1C0      | 9.3 ± 1.0 ab | 16.3 ± 0.9 d | 1.4 ± 0.7 bc | 4.1 ± 1.2 b | 16.5 ± 2.8 b | 0.7 ± 0.1 b | 7.6 ± 0.4 b | 17.2 ± 1.9 b | 24.6 ± 0.7 a |
N1C1      | 9.6 ± 0.1 ab | 20.4 ± 4.0 c | 1.1 ± 0.1 bc | 4.4 ± 0.7 b | 11.0 ± 3.8 c | 0.8 ± 0.2 b | 7.3 ± 1.1 c | 13.2 ± 1.1 c | 100 ± 3.0 b |
N1C2      | 8.1 ± 0.7 b  | 20.9 ± 1.7 bc | 0.9 ± 0.2 bc | 3.6 ± 0.7 b | 8.8 ± 2.2 cd | 0.7 ± 0.1 b | 5.5 ± 0.7 bc | 8.6 ± 1.6 d  | 100 ± 2.5 b |
N2C0      | 10.7 ± 1.5 a | 26.3 ± 1.5 a | 1.6 ± 0.2 b | 7.1 ± 3.0 a | 18.0 ± 2.4 ab | 0.8 ± 0.2 b | 13.0 ± 0.8 a | 24.8 ± 0.6 a | 242 ± 1.8 a |
N2C1      | 9.6 ± 0.8 ab | 25.9 ± 0.5 ab | 2.6 ± 1.1 a | 6.9 ± 0.4 a | 20.8 ± 3.1 a | 12 ± 1.1 a | 12.2 ± 0.5 a | 18.5 ± 1.8 b | 253 ± 3.9 a |
N2C2      | 9.6 ± 1.2 ab | 18.2 ± 0.3 d | 1.5 ± 0.6 b | 4.2 ± 1.1 b | 12.6 ± 1.2 c | 0.9 ± 0.2 b | 6.0 ± 1.6 bc | 11.6 ± 2.3 cd | 133 ± 0.9 b |
N × Bc    | ** | *** | *** | *** | *** | *** | *** | *** | *** |

The data for cumulative N\textsubscript{2}O emissions in the 1st to 4th vegetable cropping seasons from April 12, 2012, to May 25, 2013, were redrawn from Li et al. (2015a).
†Data in the brackets indicate the vegetable growing days/total days including the following fallow period for each vegetable crop season.
‡Means ± SD with different letters in the same column indicate significant differences according to the Tukey’s multiple range test (\(P < 0.05\)) among all treatments.
***Significant at \(P < 0.001\); **significant at \(P < 0.01\); n.s. not significant.
Table 5  Vegetable yield of individual vegetable crop season under different treatments in intensified vegetable field

| Treatments | 1st 2012/4/12 | 2nd 2012/7/11 | 3rd 2012/11/19 | 4th 2012/3/27 | 5th 2013/3/28 | 6th 2013/7/1 | 7th 2013/11/4 | 8th 2014/3/28 | 9th 2014/7/1 |
|------------|---------------|---------------|---------------|---------------|---------------|--------------|--------------|--------------|--------------|
| N0C0       | 17.1 ± 2.0 cde† | 92.5 ± 3.1 cd | 18.9 ± 5.7 cd | 17.7 ± 5.0 ef | 97.0 ± 13.5 d | 16.6 ± 1.5 c | 17.2 ± 3.6 bc | 246 ± 0.6 ab | 155 ± 1.7 b |
| N0C1       | 21.5 ± 4.1 bc  | 96.5 ± 7.3 bc | 18.0 ± 4.3 cd | 27.6 ± 1.4 ab | 112.6 ± 1.7 cd| 17.7 ± 6.5 c | 24.8 ± 3.1 ab | 255 ± 3.1 a  | 179 ± 0.5 a |
| N0C2       | 15.3 ± 2.7 def | 77.3 ± 9.9 e  | 41.4 ± 11.6 b | 27.8 ± 1.8 ab | 95.8 ± 3.8 d  | 16.3 ± 2.5 c | 26.1 ± 4.2 ab | 234 ± 1.9 ab | 179 ± 0.4 a |
| N1C0       | 11.1 ± 1.8 f   | 83.1 ± 12.4 de| 10.2 ± 3.6 d  | 18.6 ± 1.0 de | 132.5 ± 6.0 b | 49.9 ± 2.5 ab| 11.5 ± 4.4 c  | 147 ± 3.3 e  | 41 ± 1.3 d  |
| N1C1       | 19.3 ± 1.4 bcd | 95.2 ± 3.9 cd | 39.0 ± 8.1 b  | 25.8 ± 5.2 abc| 131.9 ± 14.1 b| 49.9 ± 6.1 a | 16.1 ± 4.4 bc | 210 ± 0.4 bc | 45 ± 1.0 d  |
| N1C2       | 14.6 ± 5.7 def | 128.9 ± 6.8 a | 57.8 ± 11.4 a | 12.6 ± 2.6 f  | 172.2 ± 4.9 a | 43.1 ± 17.3 ab| 17.0 ± 9.3 bc | 158 ± 0.8 de | 135 ± 1.2 c |
| N2C0       | 12.1 ± 4.8 ef  | 101.3 ± 2.0 bc| 16.5 ± 12.9 cd| 23.4 ± 1.9 bcd| 1296 ± 9.6 bc | 33.1 ± 1.0 b | 24.6 ± 5.8 ab | 104 ± 2.7 f  | 46 ± 0.6 d  |
| N2C1       | 23.0 ± 1.4 b   | 90.4 ± 10.2 cd| 27.7 ± 4.2 bc | 21.3 ± 4.1 cde| 1420 ± 16.1 b | 36.5 ± 1.4 b | 24.3 ± 4.8 ab | 191 ± 3.0 cd | 150 ± 0.8 bc |
| N2C2       | 29.6 ± 0.7 a   | 108.4 ± 2.2 b | 38.2 ± 7.1 b  | 30.1 ± 3.1 a  | 143.9 ± 12.4 b| 37.4 ± 4.4 b | 31.9 ± 10.1 a | 125 ± 3.0 ef | 47 ± 1.6 d  |
| N × Bc     | *** *** *** ***| *** *** *** ***| *** *** *** ***| *** *** *** ***| *** *** *** ***| *** *** *** ***| *** *** *** ***| *** *** *** ***| *** *** *** ***|

The data for vegetable yields in the 1st to 4th vegetable cropping seasons from April 12, 2012, to May 25, 2013, were redrawn from Li et al. (2015a).

†Data in the brackets indicate the vegetable growing days/total days including the following fallow period for each vegetable crop season.

‡Means ± SD with different letters in the same column indicate significant differences according to the Tukey’s multiple range test (P < 0.05) among all treatments.

***Significant at P < 0.001; ** significant at P < 0.01; * significant at P < 0.05; n.s. not significant.
acidification and a decrease in soil quality (Table 2) that had a feedback on vegetable production. Second, the study site had been conventionally, continuously cultivated with vegetables for more than 10 years and is a typical vegetable planting system, which accumulated a high background mineral N content at the beginning of the field trial. Third, irrigation and tillage practices may have caused more N loss by leaching. Overall, appropriate N management should be used to achieve synchrony between N supply and vegetable crop demand without excess or deficiency, and thus increase NUE (Chen et al., 2014). This is the key to optimizing N tradeoffs among yield, economic profit and environmental protection in intensive vegetable cropping systems.

Dynamic effects of biochar amendments on N₂O emissions, crop yields and NUE in vegetable fields

Effects of biochar amendment on the mitigation of N₂O emissions may be either positive or negative depending on the inherent characteristics of the biochar, the addition of exogenous N and the soil water regime (Cayuela et al., 2013, 2014; Li et al., 2015b). Our results show that the significant decreases in N₂O emissions induced by biochar amendment were observed (Fig. 4, \( P < 0.01 \)), which is in agreement with many previous results (Zhang et al., 2010; Cayuela et al., 2013, 2014; Li et al., 2015a). However, the effects of the biochar on N₂O mitigation in individual vegetable crops were different. Biochar amendment significantly decreased the N₂O emissions after the fourth crop season (Table 4), which shows that the biochar had a positive effect on N₂O mitigation after about 1-year incorporation (Table 1) in this intensified vegetable field. As described in Li et al. (2015b), biochar increased cumulative N₂O emissions because a small portion of biochar may serve as an extra carbon source for the heterotrophic nitrification process from the first to fourth vegetable crop seasons, although biochar is always considered to be stable in soil (Lehmann et al., 2011; Wang et al., 2015b). However, Spokas (2013) found that biochar amendments may lose the function of the inhibition of soil N₂O production due to the weathering process, which is inconsistent with our results. The impact of aging and weathering has been hypothesized to be a critical factor for the interaction of biochar with the plant and soil systems, and likely to result in alterations of biochar surface group chemistries (Joseph et al., 2010). This may cause differences in biochar effects on the N₂O suppression and soil improvements for a long-term field experiment (Spokas, 2013). However, we did not monitor the decomposing or weathering processes.

It is frequently suggested that biochar amendments to soil can increase agricultural productivity (Biederman & Harpole, 2013). In a high proportion of the studies (>90%), biochar-induced increases in crop yield were apparent, which is in agreement with our results (Fig. 4). In this analysis, we found that the biochar significantly increased the soil TN and SOC and decreased the bulk density in the vegetable field (Table 2, \( P < 0.01 \)), which improved soil quality for vegetable growth. Along with the increase in vegetable yield, biochar amendments significantly increased PFPN and ARN in relation to the treatments without biochar (Fig. 5, \( P < 0.01 \)). Therefore, biochar amendment can be an appropriate way to increase NUE (PFPN and ARN) in intensified vegetable cropping systems.

Interactions of N and biochar amendment in intensive vegetable fields

As shown in Fig. 4, biochar amendment significantly decreased cumulative N₂O emissions when N was applied, while increasing N₂O emissions in the...
treatments without N (not significant). Additionally, the relative reduction in cumulative N₂O emissions induced by biochar amendment decreased with the increasing N application rate (Fig. 4). These indicate the significant interactions between the N fertilization and biochar amendment on total cumulative N₂O emissions (Fig. 4, P < 0.01). Both N application and biochar amendments can individually and in combination affect N₂O emissions, primarily by altering N₂O production processes and N₂O product ratios of both nitrification and denitrification (Mørkved et al., 2007). In our study, the vegetable field was frequently irrigated so the WFPS was generally high (Fig. 1a), which can stimulate both nitrification and denitrification processes in vegetable field (Cheng et al., 2015). Although there was no significant influence of biochar on mineral N contents (Figs 2 and 3), biochar’s capacity to take up ammonia is well recognized (Clough & Condron, 2010), which can then decrease N₂O emissions. Moreover, the partial correlations showed that natural logarithms of N₂O fluxes were significantly correlated with NO⁻ content (Table 3, P < 0.01), which indicates that denitrification was the main contributor to N₂O production (Zhang et al., 2015a). Because the function of biochar was reported to be as an ‘electron shuttle’ that facilitates the transfer of electrons to soil denitrifying microorganisms, which together with its liming effect would promote the reduction in N₂O to N₂, it may be more effective to decrease cumulative N₂O emissions in the soils where denitrification was the main pathway of N₂O production (Cayuela et al., 2013; Cheng et al., 2015).

Nitrogen use efficiency was low in our study, but significantly increased by biochar because of the increases in vegetable yields. In addition, treatments with N fertilization and biochar incorporation had higher total vegetable yield than that of the treatments without N fertilization or no biochar (Fig. 4). Moreover, the relative increases in vegetable yields induced by biochar amendment decreased with the increasing application N rate. These indicate the positive interactions between N fertilization and biochar amendment on vegetable yield (Fig. 4, P < 0.01). As a result, biochar effect on increasing NUE decreased as the N fertilizer rate increased (Fig. 4).

Biochar can help to decrease GHGs and N loss and increase total vegetable yield gains and thus improve NUE in intensified vegetable cropping systems in China. Moreover, the relative reduction in total cumulative N₂O emissions, N₂O–N emissions factors and yield-scaled N₂O emissions and the relative increases in vegetable yield and NUE induced by biochar amendments were decreased with increasing N fertilization rate. Overall, since the N1C2 treatment simultaneously achieved the lowest N₂O–N emission factor, yield-scaled N₂O emissions and the highest total vegetable yield and NUE, a biochar amendment rate of 40 t ha⁻¹ with conventional N application is the treatment of choice from our study. We suggest that investigations into reasonable N application and reduction combined with biochar incorporation should be carried out to sustain intensified vegetable agriculture in the future.

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