

## Regulation of nitrogen balance and yield on greenhouse eggplant under biochar addition in Mollisol

YAO WANG<sup>1</sup>, MENG ZHOU<sup>2</sup>, MENG HOU<sup>2</sup>, YIMIN CHEN<sup>2</sup>, YUEYU SUI<sup>2\*</sup>,  
XIAOGUANG JIAO<sup>1\*</sup>

<sup>1</sup>College of Modern Agriculture and Ecological Environment, Heilongjiang University, Harbin, China

<sup>2</sup>Key Laboratory of Mollisols Agroecology, Northeast Institute of Geography and Agroecology, Chinese Academy of Sciences, Harbin, China

Yao Wang and Meng Zhou have contributed equally to this work and share the first authorship.

\*Corresponding authors: [suiyy@iga.ac.cn](mailto:suiyy@iga.ac.cn); [2004086@hlju.edu.cn](mailto:2004086@hlju.edu.cn)

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**Abstract:** Maintaining nitrogen (N) balance and inhibiting N leaching loss in the soil-crop system is crucial to maintaining yield and reducing the environmental pollution. This study investigated the effects of soil  $\text{NO}_3^-$ -N content and accumulation, eggplant yield, N leaching and balance response to biochar addition, including regular fertilisation and irrigation (W + F), biochar addition with regular fertilisation and irrigation (W + F + B), and biochar addition with 20% fertilisation and irrigation reduction (0.8W + 0.8F + B) treatments. Compared with W + F, W + F + B and 0.8W + 0.8F + B increased soil  $\text{NO}_3^-$ -N content in 0–40 cm and soil  $\text{NO}_3^-$ -N accumulation in 0–20 cm, and raised harvest index, N surplus and balance. Simultaneously, 0.8W + 0.8F + B compared to W + F enhanced N use efficiency and N partial factor productivity, conversely, it decreased N dry matter production efficiency, N surplus and balance. Stepwise regression analysis demonstrated that the effect of  $\text{NO}_3^-$ -N leaching lasted in 60 cm under biochar addition in the first year, and lasted in 20 cm without biochar application in the next year. Altogether, biochar addition with 20% fertilisation and irrigation reduction is the most suitable management strategy to decrease N surplus and leaching, and maintain eggplant N uptake in a two-year cycle system on greenhouse vegetables in Mollisols.

**Keywords:** soil profile; soil fertiliser; soil chemical;  $\text{NH}_3$  volatilisation; nitrification and denitrification loss

Nitrogen (N), which is an essential element for plant growth and development, plays an important role in material and energy metabolism, and the regulation of life activities (Wu et al. 2018). In recent decades, the mineral fertiliser, especially the N application amount is rising, which is accounting for 47% of the total fertiliser amounts in China (Editorial Committee of China Agricultural Statistical Yearbook 2006). Furthermore, the amount of mineral fertiliser used in greenhouse vegetable production is usually several times higher than the amount used for field crops, resulting in low fertiliser use effi-

ciency. The fluctuating range of N application loss is very large with between 19% to 95% (Wang et al. 2010), which is related to soil properties, application methods, fertilisation and irrigation amounts, and so on. Among them, excessive fertilisation and irrigation also lead to reduced vegetable yield and quality (Zhang et al. 2014, Xiao et al. 2017). The primary N loss pathways from greenhouse soils include (1) the gaseous emission of  $\text{N}_2$ , NO,  $\text{N}_2\text{O}$ , and  $\text{NH}_3$ , and (2) the leaching of ammonium-nitrogen ( $\text{NH}_4^+$ -N) and nitrate-nitrogen ( $\text{NO}_3^-$ -N). Among these N forms, the  $\text{NO}_3^-$ -N leaching loss is considered to

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be the main pathway under 5-year maize-vegetable rotation in agricultural soils of China, and it is also an important source of nitrate-nitrogen pollution in groundwater, resulting in several environmental problems worldwide (Zheng et al. 2013). Studies have pointed out that 68% and 20% of the residual  $\text{NO}_3^-$ -N in the non-arable and cultivated layer soils enters the groundwater and causes pollution every year (Li et al. 2015). Therefore, reducing soil  $\text{NO}_3^-$ -N leaching and increasing the soil's ability to hold nitrate nitrogen are important ways to increase nitrogen utilisation and reduce water eutrophication (Fan et al. 2014).

Biochar is a product with rich carbon content and stable properties formed by the thermal degradation of organic materials under the condition of complete or partial hypoxia (Lehmann et al. 2011). The high porosity and surface area inside the biochar can reduce soil bulk density, improve soil water permeability, increase the electrostatic adsorption of nutrients, thus, it can increase N effective use, reduce nitrate leaching, and significantly improve plants growth and nutrition (Bell and Worrall 2011).

A significant body of researches demonstrates that the application of biochar can indeed reduce the leaching loss of nitrate N and ammonium N in the soil, and improve the soil's ability to hold N (Chandra et al. 2020, Gao and Deluca 2020). Simultaneously, the effect of N holding capacity is associated with the amount of biochar applied, carbonisation temperature and preparation materials (Yao et al. 2012). Zhou et al. (2011) showed that when the application rate of biochar was 50 and 100 t/ha, the total nitrogen (TN) leaching loss in Mollisols would be reduced by 29% and 74%, indicating that different biochar application amounts have a different inhibitory effect of soil N leaching. Although biochar as a soil amendment is conducive to the maintenance of land productivity and provides valuable findings in the laboratory, few studies address the effects of biochar addition in greenhouse vegetable systems on soils in Northeast China.

The total area of China's greenhouse vegetables has reached more than 3.3 million ha, which is of great significance for accelerating agricultural modernisation and ensuring the "vegetable basket" project (Chang et al. 2011). Eggplant, one of the main vegetables cultivated in the greenhouse in China, has a long growth and fruiting period, and its vegetative growth and reproductive growth overlap (Keya et al. 2020). Although vegetative growth is not equivalent to the formation of economic products, relatively

speaking, the accumulation of dry matter formed during the vegetative growth stage not only controls the biological output, but also the economic output (Medeiros et al. 2019). Soil fertility is the main source of plant nutrients, so the balanced application of nitrogen, phosphorus and potassium has a greater impact on the growth and yield of eggplant (Zhou et al. 2020a, b). However, due to eggplant having a lower nutrient utilisation rate in the soil than other solanaceous fruits, blindly increasing the amounts of fertiliser and irrigation will not only increase the cost but also cause damage to the soil environment (Guo et al. 2010). Therefore, it is necessary to select the most suitable amount of fertiliser application, irrigation water, and biochar addition in the greenhouse eggplant system in Northeast China.

The purpose of our current research was to investigate the variation in soil  $\text{NO}_3^-$ -N content and accumulation, eggplant yield, N uptake, N leaching and N balance response to biochar addition, and try to explore which is a better management strategy to improve N balance and maintain crop yield. We hypothesised that (1) biochar addition would increase soil  $\text{NO}_3^-$ -N content and accumulation across the whole soil profile in 0–100 cm; (2) biochar addition would not only increase the yield but also improve N balance.

## MATERIAL AND METHODS

**Experimental site.** The study was conducted in the Institute of Horticulture, Heilongjiang Academy of Agricultural Sciences, Harbin, China (126°39.050'E, 45°37.836'N, and altitude 173.1 m a.s.l.). The site is in a moderate temperate zone with a semi-humid continental monsoon climate. The annual mean temperature and annual mean precipitation is 4.25 °C and 569 mm, respectively. The soil in the area is a typical Mollisol with a 30 cm topsoil thickness (Xing et al. 2004), and its basic soil physic-chemical properties are given in Table 1.

**Experimental design and management.** The greenhouses were initiated in 2002 orienting from south to north, with an area of 324 m<sup>2</sup> (12 m in width and 27 m in length). The experimental design was a randomised complete block with three replications consisting of 9 plots in total. Each plot was 6 m long and 3 m wide. In each plot, a leachate collecting device was installed in 2016 (Figure 1) and the device details were described by Chen et al. (2018).

The treatments were as followed and cycled in the two-year: (1) regular fertilisation and irrigation (W + F);

Table 1. Basic soil physico-chemical properties in study area

Soil depth (cm)	pH (H <sub>2</sub> O:soil = 2.5:1)	Bulk density (g/cm <sup>3</sup> )	Soil organic carbon	Total nitrogen (g/kg)	Total phosphorus
0–20	6.93	1.07	26.9	2.5	2.1
20–40	6.45	1.28	18.3	1.7	1.1
40–60	6.80	1.25	14.8	1.3	0.6
60–80	6.85	1.33	9.8	0.9	0.4
80–100	6.75	1.41	9.6	0.8	0.6

(2) biochar addition with regular fertiliser and irrigation (W + F + B); (3) biochar addition with reducing 20% fertiliser and 20% irrigation (0.8W + 0.8F + B). The specific fertilisation and irrigation amounts of the three treatments are shown in Table 2 according to the local regular dosages. The irrigation method is drip irrigation under mulch. In terms of W + F + B and 0.8W + 0.8F + B treatments, biochar was applied in 2018, but no biochar was added in 2019, the other management measures were the same, we wanted to explore the sustaining effect of biochar applied into the soil.

Biochar, which was made from fruitwood at 600 °C, was applied to the soil surface together with basal fertilisers before subsoiling and mixed it evenly into the 0–20 cm soil depth when ploughing. Biochar was applied by 30 t/ha once every two years since 2002. The pH value, total carbon, total nitrogen, total phosphorus, and total potassium contents of biochar are 8.87, 715 g/kg, 6.9 g/kg, 1.4 g/kg, and 11.5 g/kg, respectively. The mineral fertiliser of urea (46.0% of N), superphosphate (5.2% of P) and potassium sulfate

(41.5% of K), and commercial organic fertilisers (5.0% of N; 1.3% of P; 2.5% of K) were applied as basal and topdressing fertilisers and drip irrigation systems were used in the current experiment. Irrigation was terminated when the last picking stage reached.

Eggplant (*Solanum melongena* L.) was planted in the greenhouses since 2002, and the eggplant cultivar was Longza 201 in 2018 and 2019. Eggplant seeds were sowed in float trays for the nursery in March 2018 and 2019. The soil was tilled through the 0–20 cm layer by the rotary cultivator. The ridges with all 1 m width were prepared before eggplant transplantation on April 5, 2018, and 2019. A drip tape was placed in the middle of each ridge, and then the ridge was covered with black polyethylene mulch of 1.2 m width. Eggplants were transplanted with 100 cm spacing in lines and 50 cm spacing in rows. The eggplants were harvested by hand when maturing, and the first harvest was in mid-June in 2018 and 2019, and the last in late-September in 2018 and 2019. In total, 10 harvests in each year were done during the period with the frequency of once in ten days.

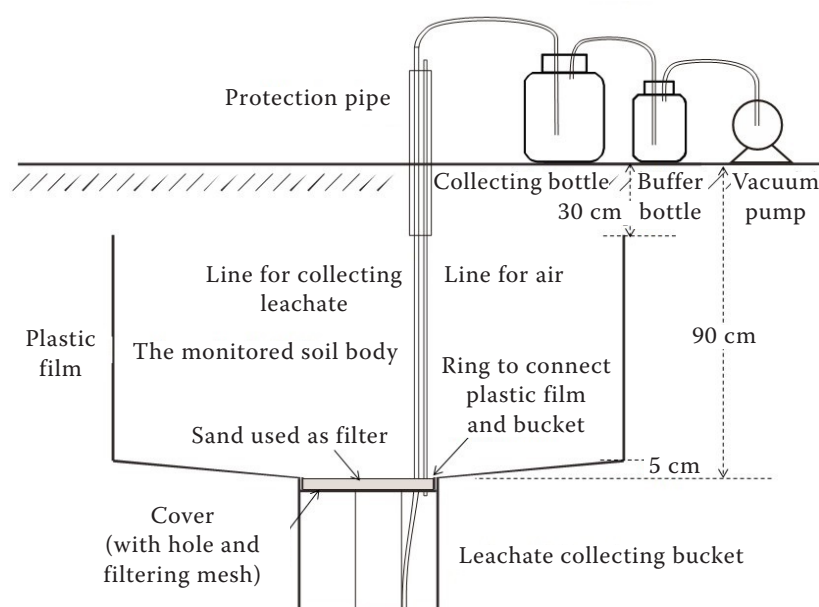


Figure 1. Schematic diagram of the leaching solution acquisition device

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Table 2. Fertilisation and irrigation management for the three treatments

Treatment	Basal fertiliser (kg/ha)				Dressing fertiliser in the early fruit stage (kg/ha)	Dressing fertiliser in the full bearing stage (kg/ha)		Irrigation after transplanting (m <sup>3</sup> /ha)	Irrigation every 7 to 10 days when the plant recovered (m <sup>3</sup> /ha)	Biochar addition (t/ha)
	organic fertiliser	N	P	K	N	N	K			
W + F	5 000	72	32	91	70	23	93	27	45	0
W + F + B	5 000	72	32	91	70	23	93	27	45	30
0.8W + 0.8F + B	5 000	57.6	25	73	56	18.4	75	21.6	36	30

**Sampling and analysis of soil, leachate and plant samples.** Leachate samples were collected from the leachate collecting bucket by a vacuum pump. Soil NO<sub>3</sub><sup>-</sup>-N was analysed using dual-wavelength colorimetry methods. In addition, eggplant fruit was picked at the mature period and then weighed. Total nitrogen content in eggplants was analysed through the H<sub>2</sub>SO<sub>4</sub>-H<sub>2</sub>O<sub>2</sub> distillation method (Bao 2008). NH<sub>3</sub> volatilisation, and nitrification and denitrification loss were measured according to the method by Sun et al. (2013).

**Calculations and statistical analyses.** Soil NO<sub>3</sub><sup>-</sup>-N content and accumulation were calculated using the following Eqs. 1 and 2 (Li et al. 2020):

$$C = C_0 \times V \times A (1 + W_0) / W_1 \quad (1)$$

Where: C – soil NO<sub>3</sub><sup>-</sup>-N content (dry weight, mg/kg); C<sub>0</sub> – detection concentration (mg/kg); V – extraction volume (mL); A – split multiple; W<sub>0</sub> – mass water content (%); W<sub>1</sub> – fresh soil quality (g).

$$M = C \times H \times B / 10 \quad (2)$$

Where: M – soil NO<sub>3</sub><sup>-</sup>-N accumulation (kg/ha); H – soil thickness (cm); B – bulk density (g/cm<sup>3</sup>).

Eggplant yield was calculated by adding the yields from each harvest together. Eggplant biomass was calculated by yield plus the weights of roots, stems, and leaves. In addition, N<sub>up</sub> refers to total nitrogen uptake by eggplant, and it was calculated by the sum of dry weight (kg/ha) multiplied by total nitrogen content (g/kg), including eggplant fruit, root, stem, and leaf.

HI, NHI, NUE, NUE<sub>d</sub>, NUE<sub>g</sub> and PFP<sub>N</sub> were calculated using the following equations 3–8 (Liang et al. 2015, Mauceri et al. 2020):

$$HI (\%) = Y/B \times 100 \quad (3)$$

$$NHI (\%) = N_f / N_{up} \times 100 \quad (4)$$

$$NUE = N_{up} / N_{ap} \quad (5)$$

$$NUE_d = DMA / N_{up} \quad (6)$$

$$NUE_g = Y / N_{up} \quad (7)$$

$$PFP_N = Y / N_F \quad (8)$$

Where: Y – yield (t/ha); B – biomass (t/ha); NHI – N harvest index (%); N<sub>f</sub> – fruit N uptake (kg/ha); NUE – N use efficiency (%); N<sub>up</sub> – total N uptake (kg/ha); N<sub>ap</sub> – rate of fertiliser and irrigation water N application (kg/ha); NUE<sub>d</sub> – N dry matter production efficiency; DMA – dry matter accumulation (including fruit, root, stem, and leaf); NUE<sub>g</sub> – N grain production efficiency; PFP<sub>N</sub> – N partial factor productivity; N<sub>F</sub> – fertiliser (mineral fertilisers and organic fertilisers) N.

$$N \text{ leaching amount (kg/ha)} = (C_i \times V_i \times 10^{-3}) / (1.2 \times 10^{-4}) \quad (9)$$

Where: C<sub>i</sub> – concentration nitrate-nitrogen in leachate (mg/L); V<sub>i</sub> – volume of leachate (L); 1.2 – area of the monitoring (m<sup>2</sup>). 10<sup>-3</sup> and 10<sup>-4</sup> represent the efficiencies to convert g to kg and m<sup>2</sup> to ha, respectively.

N balance and N surplus in the soil-plant system were calculated as the following Eqs. 10–13 (Mauceri et al. 2020):

$$N_{bal} = N_{input} - N_{up} \quad (10)$$

$$N_{input} = N_F + N_O + N_B + N_I + N_S + N_A \quad (11)$$

$$N_{output} = N_{up} + N_L + N_V + N_N \quad (12)$$

$$N_{sur} = N_{input} - N_{output} \quad (13)$$

Where: N<sub>bal</sub> – N balance; N<sub>input</sub> and N<sub>output</sub> – input and output nitrogen from the whole soil-plant system; N<sub>F</sub> – N inputs from chemical fertilisers; N<sub>O</sub> – N inputs from organic fertilisers; N<sub>B</sub> – N input from biochar; N<sub>I</sub> – inputs from irrigation; N<sub>S</sub> – inputs from eggplant seeds; N<sub>A</sub> – N inputs from atmospheric deposition; N<sub>L</sub> – N outputs from leaching (mainly refers to NO<sub>3</sub><sup>-</sup>-N leaching); N<sub>V</sub> – NH<sub>3</sub> volatilisation; N<sub>N</sub> – nitrification and denitrification loss; N<sub>sur</sub> – N surplus.

In the present study, N inputs from eggplant seeds were assumed to be negligible because the sowing

rate in the greenhouses was very low and the small seedlings of eggplant were transplanted into the greenhouses. Similarly, N inputs from atmospheric deposition were considered to be zero because eggplants were covered by the plastic film during the entire growth period. N inputs from fertilisers were calculated from the amount of fertilisers applied, and the forms and concentrations of nutrients present. The N fertilisers used in this study were urea and organic fertilisers. N inputs from irrigation water were calculated from the amount of irrigation water and the N concentrations in irrigation over the year.

The stepwise regression analysis and significant difference analysis were carried out using SPSS 20.0 software (Armonk, USA), and all graphs were generated using Origin 7.5 software (Northampton, USA).

## RESULTS

**Soil  $\text{NO}_3^-$ -N content and accumulation.** In general, the soil  $\text{NO}_3^-$ -N content of each treatment decreases with the soil depth of 0–100 cm increases in 2018 and 2019, among them, 0–40 cm reduced the most (Figure 2). Across the whole soil profile of 0–100 cm, the highest soil  $\text{NO}_3^-$ -N content was always observed in W + F + B treatment for the two years. Specifically, compared with W + F treatment, W + F + B and 0.8W + 0.8F + B treatments significantly ( $P < 0.05$ ) increased soil  $\text{NO}_3^-$ -N content by 68.1% and 32.7% in 0–20 cm, and 137.0% and 70.9% in 20–40 cm, and 100.3% and 52.6% in 40–60 cm soil depths in 2018, respectively. Similarly, W + F + B and 0.8W + 0.8F + B treatments as compared to W + F treatment also significantly ( $P < 0.05$ ) enhanced soil  $\text{NO}_3^-$ -N con-

tent by 32.2% and 13.8% in 0–20 cm, and 76.4% and 44.9% in 20–40 cm soil depths. However, no significant ( $P > 0.05$ ) differences for soil  $\text{NO}_3^-$ -N content were demonstrated in 60–100 cm soil depth of 2018 and 40–100 cm soil depth of 2019 among the three treatments.

Across the whole soil profile of 0–100 cm, the biggest soil  $\text{NO}_3^-$ -N accumulation appeared in 0.8W + 0.8F + B treatment for 2018, but, the treatment of the maximum value on each soil layer in 2019 varied different (Table 3). Moreover, compared to W + F treatment, W + F + B and 0.8W + 0.8F + B treatments significantly ( $P < 0.05$ ) increased soil  $\text{NO}_3^-$ -N accumulation by 21.2% and 41.1% in 2018, and 10.4% and 30.3% in 2019 on 0–20 cm soil depth, respectively. In addition, only 0.8W + 0.8F + B compared to W + F treatment significantly ( $P < 0.05$ ) raised soil  $\text{NO}_3^-$ -N accumulation by 39.2% in 2018 and 28.2% in 2019 on 80–100 cm soil depth.

**Yield, biomass, HI, NHI, NUE,  $\text{NUE}_d$ ,  $\text{NUE}_g$  and  $\text{PFP}_N$ .** Compared with W + F treatment, W + F + B treatment significantly ( $P < 0.05$ ) increased eggplant yield by 6.7% in 2019 (Table 4). However, eggplant yield was not significantly affected among the three treatments in 2018, and between W + F and 0.8W + 0.8F + B treatments in 2019.

No obvious differences ( $P > 0.05$ ) for eggplant biomass,  $\text{N}_{up}$ , NHI, and  $\text{NUE}_g$  were presented among the three treatments in 2018 and 2019 (Table 4). Compared to W + F treatment, both adding biochar practices significantly ( $P < 0.05$ ) enhanced HI by 2.8% and 2.9% in 2018, and also raised it by 3.4% and 3.0% in 2019, respectively. Only 0.8W + 0.8F + B treatment obviously increased ( $P < 0.05$ ) NUE by

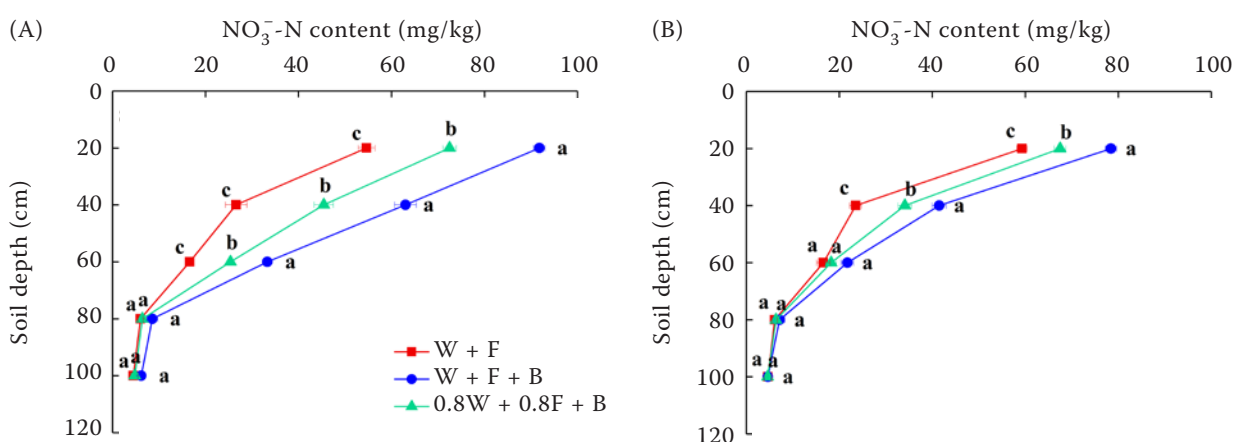


Figure 2. Effect of different treatments on nitrate distribution in the soil profile in (A) 2018 and (B) 2019. W + F – regular fertilisation and irrigation; W + F + B – biochar addition with regular fertilisation and irrigation; 0.8W + 0.8F + B – biochar addition with 20% fertilisation and irrigation reduction

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Table 3. Effect of biochar addition (kg/ha) on  $\text{NO}_3^-$ -N accumulation in soil profile

Year	Soil depth (cm)	W + F	W + F + B	0.8W + 0.8F + B
2018	0–20	$50.09 \pm 1.03^c$	$60.69 \pm 0.74^b$	$70.66 \pm 0.84^a$
	20–40	$10.87 \pm 0.72^a$	$10.48 \pm 0.68^a$	$11.60 \pm 0.17^a$
	40–60	$8.63 \pm 1.35^a$	$9.85 \pm 0.68^a$	$9.58 \pm 0.79^a$
	60–80	$6.20 \pm 0.72^a$	$6.30 \pm 0.28^a$	$6.86 \pm 0.52^a$
	80–100	$5.71 \pm 0.56^b$	$5.91 \pm 0.26^b$	$7.95 \pm 0.45^a$
2019	0–20	$50.10 \pm 0.92^c$	$55.32 \pm 0.67^b$	$65.27 \pm 0.58^a$
	20–40	$11.33 \pm 1.28^a$	$10.24 \pm 1.06^a$	$10.67 \pm 0.88^a$
	40–60	$7.09 \pm 1.11^a$	$7.51 \pm 0.43^a$	$8.58 \pm 0.54^a$
	60–80	$5.64 \pm 1.66^a$	$7.53 \pm 0.33^a$	$7.20 \pm 1.08^a$
	80–100	$7.82 \pm 0.49^{ab}$	$6.74 \pm 0.69^b$	$10.02 \pm 0.61^a$

Different lowercase letters in the same row indicate significant differences among different treatments at every soil depth ( $P < 0.05$ ; Tukey's *HSD* (honestly significant difference) test). W + F – regular fertilisation and irrigation; W + F + B – biochar addition with regular fertilisation and irrigation; 0.8W + 0.8F + B – biochar addition with 20% fertilisation and irrigation reduction

Table 4. Yield, biomass, harvest index, nitrogen (N) harvest index and production efficiency response to various managements

Year	Indicator	W + F	W + F + B	0.8W + 0.8F + B
2018	yield (t/ha)	$52.85 \pm 0.33^a$	$55.04 \pm 1.55^a$	$52.97 \pm 1.03^a$
	biomass (t/ha)	$61.09 \pm 0.63^a$	$61.86 \pm 1.46^a$	$59.48 \pm 1.08^a$
	$N_{up}$ (kg/ha)	$153.02 \pm 1.82^a$	$161.91 \pm 2.89^a$	$154.74 \pm 3.71^a$
	HI (%)	$86.52 \pm 0.36^b$	$88.96 \pm 0.58^a$	$89.05 \pm 0.14^a$
	NHI (%)	$13.34 \pm 0.50^a$	$12.83 \pm 0.03^a$	$13.34 \pm 0.27^a$
	NUE (%)	$33.05 \pm 0.39^b$	$34.97 \pm 0.62^{ab}$	$36.84 \pm 0.88^a$
	$NUE_d$	$52.71 \pm 1.01^a$	$49.90 \pm 0.50^{ab}$	$48.81 \pm 0.85^b$
	$NUE_g$	$345.39 \pm 1.91^a$	$339.94 \pm 7.85^a$	$342.40 \pm 3.90^a$
2019	$PFP_N$	$137.34 \pm 0.80^b$	$130.19 \pm 1.50^b$	$140.29 \pm 1.37^a$
	yield (t/ha)	$52.64 \pm 0.65^b$	$56.19 \pm 0.93^a$	$52.79 \pm 0.62^b$
	biomass (t/ha)	$60.97 \pm 0.90^a$	$62.96 \pm 1.07^a$	$59.36 \pm 0.51^a$
	$N_{up}$ (kg/ha)	$145.05 \pm 12.17^a$	$153.07 \pm 3.79^a$	$151.01 \pm 6.50^a$
	HI (%)	$86.34 \pm 0.26^b$	$89.24 \pm 0.04^a$	$88.94 \pm 0.38^a$
	NHI (%)	$13.81 \pm 1.09^a$	$13.93 \pm 0.46^a$	$13.80 \pm 0.44^a$
	NUE (%)	$29.78 \pm 1.47^b$	$33.06 \pm 0.82^{ab}$	$35.96 \pm 1.55^a$
	$NUE_d$	$52.51 \pm 0.42^a$	$48.71 \pm 0.45^b$	$49.17 \pm 0.49^b$
	$NUE_g$	$350.91 \pm 13.60^a$	$367.32 \pm 6.31^a$	$350.66 \pm 12.94^a$
	$PFP_N$	$126.83 \pm 1.57^b$	$135.40 \pm 2.24^a$	$138.20 \pm 1.62^a$

Different lowercase letters in the same row indicate significant differences among different treatments ( $P < 0.05$ ; Tukey's *HSD* (honestly significant difference) test). W + F – regular fertilisation and irrigation; W + F + B – biochar addition with regular fertilisation and irrigation; 0.8W + 0.8F + B – biochar addition with 20% fertilisation and irrigation reduction;  $N_{up}$  – total nitrogen uptake by eggplant; HI – the ratio of yield to biomass; NHI – nitrogen harvest index; NUE – nitrogen use efficiency;  $NUE_d$  – nitrogen dry matter production;  $NUE_g$  – nitrogen grain production efficiency;  $PFP_N$  – nitrogen partial factor productivity

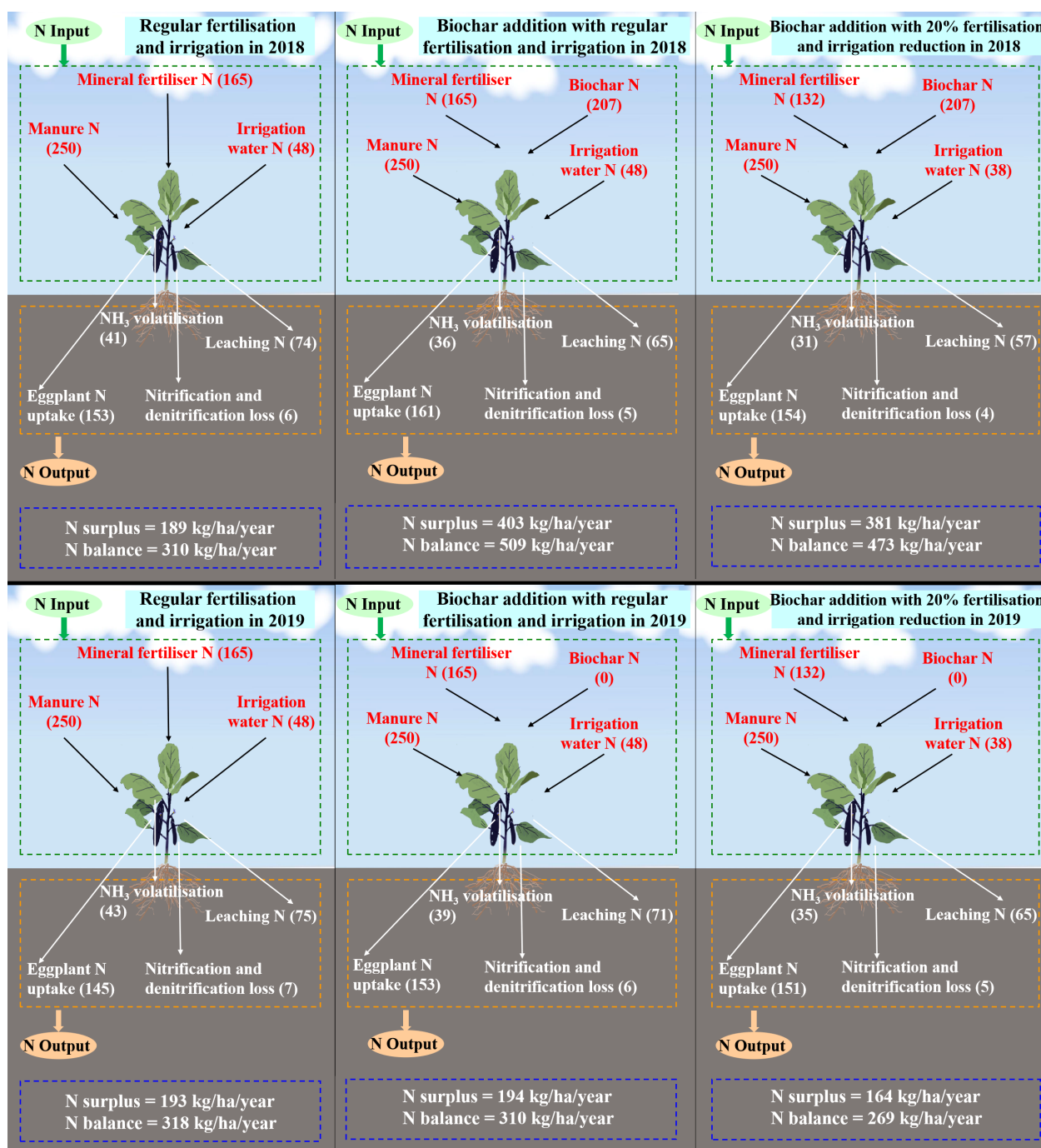


Figure 3. Schematic diagram of nitrogen (N) balance in two-year cycle system on greenhouse vegetable in Mollisols

11.5% and 20.7% in 2018 and 2019 as compared to W + F treatment. Furthermore, compared to W + F treatment, 0.8W + 0.8F + B treatment significantly ( $P < 0.05$ ) enhanced PFP<sub>N</sub> by 10.2% in 2018, while, W + F + B and 0.8W + 0.8F + B treatments both increased it by 6.8% and 9.0% in 2019. Conversely, compared with W + F treatment, 0.8W + 0.8F + B treatment significantly ( $P < 0.05$ ) decreased NUE<sub>d</sub>

by 7.4% in 2018, but, W + F + B and 0.8W + 0.8F + B treatments both decreased it by 7.3% and 6.4% in 2019.

**N balance.** In general, total N input in the eggplant greenhouse ranged from 463 to 670 kg/ha in 2018 and ranged from 420 to 463 kg/ha in 2019 across the three treatments (Figure 3). Simultaneously, the highest input was observed in the W + F + B treatment with 670 kg/ha in 2018 and 463 kg/ha in 2019.

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Furthermore, N inputs from mineral and organic fertilisers were the main N sources in the greenhouse, accounting for 60.9–89.6% of total N inputs in 2018, and 89.6–91.0% in 2019; next is biochar N with 30.9–33.0% in 2018; however, irrigation water, and seed, accounted for less than 11% of the total N input across the three treatments (Table 5).

Eggplant N uptake was the primary N output pathway in this vegetable cropping system in the greenhouse, which accounted for 55.8–62.6% of total N outputs in 2018, and 53.7–59% in 2019 (Figure 3). Subsequently was leaching N with 23.2–27.8% and

NH<sub>3</sub> volatilisation with 12.6–15.9%, and nitrification and denitrification loss was the smallest proportion with 1.6–2.6% in the two years.

In addition, compared to W + F treatment, leaching N and NH<sub>3</sub> volatilisation were both significantly ( $P < 0.05$ ) decreased by 12.2% and 12.2% under W + F + B treatment, and reduced them in sequence by 23% and 24.4% under 0.8W + 0.8F + B treatment in 2018. Whereas, only 0.8W + 0.8F + B as compared to W + F treatment significantly ( $P < 0.05$ ) decreased leaching N and NH<sub>3</sub> volatilisation by 13.3% and 18.6% in 2019 (Table 5). No significant difference ( $P > 0.05$ )

Table 5. Nitrogen (N) balance response to various managements in the greenhouse vegetable system (kg/ha/year)

Year	Indicator	W + F	W + F + B	0.8W + 0.8F + B
2018	N inputs	mineral fertiliser N	165	165
		manure N	250	250
		biochar N	0	207
		irrigation water N	48	48
		seed N	nd <sup>a</sup>	nd <sup>a</sup>
		atmospheric deposition N	nd <sup>b</sup>	nd <sup>b</sup>
		total N input	463	670
	N outputs	eggplant N uptake	153 <sup>a</sup>	161 <sup>a</sup>
		leaching N	74 <sup>a</sup>	65 <sup>b</sup>
		NH <sub>3</sub> volatilisation	41 <sup>a</sup>	36 <sup>b</sup>
		nitrification and denitrification loss	6 <sup>a</sup>	5 <sup>a</sup>
		total N output	274 <sup>a</sup>	267 <sup>a</sup>
	N surplus	189 <sup>c</sup>	403 <sup>a</sup>	381 <sup>b</sup>
	N balance	310 <sup>c</sup>	509 <sup>a</sup>	473 <sup>b</sup>
2019	N inputs	mineral fertiliser N	165	165
		manure N	250	250
		biochar N	0	0
		irrigation water N	48	48
		seed N	nd	nd
		atmospheric deposition N	nd	nd
		total N input	463	463
	N outputs	eggplant N uptake	145 <sup>a</sup>	153 <sup>a</sup>
		leaching N	75 <sup>a</sup>	71 <sup>a</sup>
		NH <sub>3</sub> volatilisation	43 <sup>a</sup>	39 <sup>a</sup>
		nitrification and denitrification loss	7 <sup>a</sup>	6 <sup>a</sup>
		total N output	270 <sup>a</sup>	269 <sup>a</sup>
	N surplus	193 <sup>a</sup>	194 <sup>a</sup>	164 <sup>b</sup>
	N balance	318 <sup>a</sup>	310 <sup>a</sup>	269 <sup>b</sup>

Different lowercase letters in the same row indicate significant differences among different treatments ( $P < 0.05$ ; Tukey's *HSD* (honestly significant difference) test). W + F – regular fertilisation and irrigation; W + F + B – biochar addition with regular fertilisation and irrigation; 0.8W + 0.8F + B – biochar addition with 20% fertilisation and irrigation reduction; nd – not determined

was observed for eggplant N uptake, nitrification and denitrification loss, and total N output among the three treatments in 2018 and 2019.

Across the three treatments, N balance and N surplus varied from 189 to 403 kg/ha/year and 269–509 kg/ha/year (Figure 3). Compared with WF treatment, W + F + B and 0.8W + 0.8F + B treatments significantly ( $P < 0.05$ ) enhanced N surplus by 1.1 fold and 1 fold in 2018, while, 0.8W + 0.8F + B treatment significantly ( $P < 0.05$ ) decreased it by 15% in 2019. Similarly, compared to W + F treatment, W + F + B and 0.8W + 0.8F + B treatments significantly ( $P < 0.05$ ) increased N balance by 64.2% and 52.6% in 2018, but, 0.8W + 0.8F + B treatment obviously ( $P < 0.05$ ) decreased it by 15.4% in 2019 (Table 5).

**Stepwise regression analysis.** Linear regression models between  $\text{NO}_3^-$ -N leaching in 0–100 cm soil depth and soil  $\text{NO}_3^-$ -N accumulation in the different soil depths, obtained by stepwise regression analysis, were listed in Table 6. The data indicated that  $\text{NO}_3^-$ -N leaching in 0–100 cm soil depth was significantly and negatively correlated to soil  $\text{NO}_3^-$ -N accumulation in 0–20 cm and 40–60 cm soil depths in 2018, as shown in Eq. (1) ( $R^2 = 0.890$ ;  $P = 0.047$ ). Simultaneously, a significant and negative correlation was also obtained between  $\text{NO}_3^-$ -N leaching in 0–100 cm soil depth and soil  $\text{NO}_3^-$ -N accumulation in 0–20 cm soil depth in 2019, as shown in Eq. (2) ( $R^2 = 0.849$ ;  $P = 0$ ).

## DISCUSSION

**Effects of different management on  $\text{NO}_3^-$ -N content and accumulation.** Biochar plays a vital role in adsorption reaction due to its loose pores (Cao et al. 2018), resulting in affecting nutrient retention including  $\text{NH}_4^+$ -N and  $\text{NO}_3^-$ -N (Knowles et al. 2011). In the current study, the significant decreasing  $\text{NO}_3^-$ -N leaching in soil solution, and increasing

soil  $\text{NO}_3^-$ -N content and accumulation at 0–100 cm soil depth in the biochar addition treatments than without biochar application verified the above research results. This may be because that biochar can enhance soil water retention ability, and  $\text{NO}_3^-$ -N can remain in soil solution within biochar pores (Basso et al. 2013). Additionally, the reduction in  $\text{NO}_3^-$ -N leaching of the biochar-applied soils may be due to the longer retention of  $\text{NO}_3^-$ -N in the rhizosphere, thus caused increasing N uptake by plants (Kameyama et al. 2012). Furthermore, Razzaghi et al. (2020) demonstrated that biochar application changed the soil pore size and distribution, and altered the residence time, N percolation mode and flow path of soil water. Simultaneously, biochar particles can provide a suitable environment for the growth of microorganisms (Haider et al. 2021), and these microorganisms transform organic N into mineral N that vegetables can adsorb effectively, resulting in inhibiting N leaching loss (Warnock et al. 2007). Therefore, biochar amendment may indirectly or directly affect  $\text{NO}_3^-$ -N leaching *via* its interaction with nutrients and functional properties, including large surface area, highly porous structure, and strong ion exchange capacity, etc. (Nan et al. 2016).

Biochar can affect the soil water holding capacity and nutrient leaching, also depending on soil texture, biochar addition duration and amount, and biochar materials, etc. (Wang et al. 2017). Several researchers found that  $\text{NO}_3^-$ -N leaching may be substantially reduced soon after biochar application on Templeton Silt Loam, Ashley Dene Silt Loam and Brickfield soils (Bell and Worrall 2011), and in a Haplic Calcisol (a silty clay loam texture) (Ventura et al. 2013). In our present research, biochar was applied in 2018, but not added in 2019. Whereas, the  $\text{NO}_3^-$ -N content, accumulation, and leaching effect lasted for two years.

Biochar addition inhibits the migration of soil solution and improves the soil water-holding capacity due

Table 6. Relationship among  $\text{NO}_3^-$ -N leaching and soil  $\text{NO}_3^-$ -N accumulation in the soil profile ( $n = 9$ )

Year	Indicators in Y	Indicators in X	Regression model (Equation number)	$R^2$	$F$	$P$
2018	$\text{NO}_3^-$ -N leaching in 0–100 cm soil depth	X2: soil $\text{NO}_3^-$ -N accumulation in 0–20 cm soil depth X4: soil $\text{NO}_3^-$ -N accumulation in 40–60 cm soil depth	$Y = -0.746X_2 - 1.495X_4 + 124.562$ (1)	0.890	6.244	0.047
2019	$\text{NO}_3^-$ -N leaching in 0–100 cm soil depth	X2: soil $\text{NO}_3^-$ -N accumulation in 0–20 cm soil depth	$Y = -1.085X_2 + 127.204$ (2)	0.849	45.923	0

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to its high porosity and large specific surface area, thus reducing soil water percolation and the volume of the leaching solution, resulting in enhancing soil nitrate-nitrogen content (Liu et al. 2019). However, in our experiment, this phenomenon only appeared in shallow soils, which might be because the water movement was mainly concentrated in shallow soils due to the small amount of high-frequency irrigation practices under mulch drip irrigation, thereby making the deep soils unable to replenish water in time. Simultaneously, our results found that no significant difference was observed in the water content of deep soils across the three treatments (data not shown), which indicates that the biochar addition had no obvious effect on the nitrate-nitrogen leaching of deep soils, ultimately leading to the nitrate-nitrogen content in deep soils was not statistically significant between biochar addition and without biochar application practices.

Inconsistent with our hypothesis, based on biochar addition, the effect of soil depth for soil  $\text{NO}_3^-$ -N content was 0–60 cm in 2018 and 0–40 cm in 2019 (Figure 2), while, for soil  $\text{NO}_3^-$ -N accumulation was 0–20 cm and 80–100 cm (Table 3). This phenomenon demonstrated that although biochar application increased  $\text{NO}_3^-$ -N content in surface and sub-surface greenhouse eggplant soil, it enhanced  $\text{NO}_3^-$ -N accumulation in surface and deep soil, especially for the combined application of biochar and reducing fertiliser and irrigation water. Furthermore, the significant and negative correlation between  $\text{NO}_3^-$ -N leaching and soil  $\text{NO}_3^-$ -N accumulation on 0–20 cm and 40–60 cm soil depths in 2018, and the same relationship between  $\text{NO}_3^-$ -N leaching and soil  $\text{NO}_3^-$ -N accumulation on 0–20 cm soil depth in 2019 (Table 6) demonstrated that  $\text{NO}_3^-$ -N leaching along the 0–100 cm soil profile decreased with soil  $\text{NO}_3^-$ -N accumulation increased, but the reaction of specific soil depth in different years varied. These results also indicated that the effect of  $\text{NO}_3^-$ -N leaching lasted in 60 cm soil depth when applying biochar in the first year, while, the impact lasted only in 20 cm soil depth when no biochar was added in the second year.

The current research demonstrated that under biochar addition, the combined application of reducing irrigation and fertilisation amounts by 20% decreased nitrate-nitrogen content as compared to the full amounts of irrigation and fertilisation. On the one hand, water deficiency might promote plant root growth (Noack et al. 2010), which increase water and nutrients uptake by plants (Ashraf et al. 2005) and reduce soil N leaching. On the other hand,

water and fertiliser deficiency might decrease soil microbial biomass and diversity, and inhibit enzyme activities (Bastida et al. 2017), resulting in inhibiting the transformation of ammonium nitrogen to nitrate nitrogen, thus reducing soil nitrate-nitrogen content.

**Effects of different management on N uptake, yield and NUE.** A large number of studies showed that biochar application promoted crop growth, increased crop yield, and improved soil environment (Zhang et al. 2019a, Kamala and Bastin 2021). In our study, though biochar addition did not increase eggplant yield in 2018, the combined application of regular fertilisation and biochar significantly enhanced eggplant yield by 6.7% than without biochar addition in 2019, which may predict that the effect of biochar addition on yield can be observed in the second year. Simultaneously, the HI, NUE, and  $\text{PFP}_\text{N}$  were all enhanced by adding biochar in 2018 and 2019. Firstly, biochar can inhibit the formation and emission of  $\text{N}_2\text{O}$  in the process of nitrogen nitrification and denitrification by improving soil aeration conditions (Zhang et al. 2012). Secondly, biochar can reduce the ammonia concentration in the soil solution, thereby reducing the volatilisation of nitrogen in the form of ammonia (Lehmann et al. 2011). Finally, biochar particles have large cation adsorption capacity, porosity and high specific surface area. Nitrogen in fertilisers, especially ammonium ions, is easily adsorbed and the biochar is compounded to achieve a slow-release effect, so as to promote the continuous absorption and utilisation of plants (Lehmann and Joseph 2009). Consequently, the application of biochar enhanced N availability in soil and plant N uptake, and eventually increased nitrogen use efficiency and promoted yield, to a certain extent.

Furthermore, the combined application of biochar, and reducing fertiliser and irrigation by 20% had the highest NUE in the three managements, which speculated that based on applying biochar, reducing the input of fertiliser and irrigation water is the most appropriate practice to improve NUE and maintain yield of greenhouse eggplant in Mollisols. On the contrary, Cao et al. (2018) demonstrated that it is not appropriate to reduce N fertiliser rates in the presence of biochar amendments on orchard soils, given that the reduction in biomass accumulation when N fertiliser was decreased from 450 to 50 mg  $\text{NO}_3^-$ -N/kg. Whereas, biochar application enabled enhanced plant biomass and NUE to maintain productivity through decreasing N loss and promoting N uptake in the root system of *Malus hupehensis*

on orchard soil when the N fertiliser rate reached 300 mg  $\text{NO}_3^-$ -N/kg. The inconsistent results uncover that when combining the application of fertiliser and biochar, the NUE effect is dependent on the initial N fertiliser rate and soil type.

No significant difference was presented for eggplant N uptake, NUE, and biomass between biochar addition with 20% fertilisation and irrigation reduction practice and regular amounts in 2018 and 2019, which might be because the essential nutrient content in the vegetable field was relatively high, and the continuous application of large amounts of fertiliser made the soil nutrient sufficient for the eggplant's N needs. In 2018, the eggplant yield was decreased under biochar addition with 20% fertilisation and irrigation reduction as compared to full amounts, but the difference was not significant. However, the reduction in 2019 reached statistical significance, indicating that the biochar addition had a greater effect on crop yield (biochar was added in 2018 and no-biochar was added in 2019). Simultaneously, water deficiency measures cannot provide the normal water demand of eggplant plants, resulting in a reduction in the actual water consumption of eggplants, and consequently a decrease in yield.

#### Effects of different management on N balance.

In the present research, leaching N loss refers to  $\text{NO}_3^-$ -N. On the one hand, due to mineralisation and nitrification,  $\text{NO}_3^-$ -N accumulates in the soil, which increases the soil N content, promotes mineralisation and nitrification, and produces a strong leaching loss, thus makes the  $\text{NO}_3^-$ -N leaching loss greater than  $\text{NH}_4^+$ -N (Clément et al. 2020). On the other hand, soil colloids are negatively charged and have weak adsorption performance for  $\text{NO}_3^-$ -N, leading to the mobility of  $\text{NO}_3^-$ -N is stronger than  $\text{NH}_4^+$ -N, and the organic matter content is higher,  $\text{NH}_4^+$ -N is easily adsorbed by soil colloids and is not easy to be leached (Drury et al. 2012).

Under the current experimental conditions, the surplus N mainly refers to soil N residue, followed by N leaching (Table 5). Several studies indicated that the continuous leaching of residual inorganic N in the soil into the deep soil is the main direction of N surplus, thus posing a huge threat to groundwater (Cheng et al. 2016). Therefore, based on the monitoring of this study and previous research results, the amount of N fertiliser should be controlled and the single application of quick-acting N fertiliser should be avoided, so that the amount of N fertiliser can be matched with the absorption of crops (Zhang et al. 2019b).

In 2018 and 2019, the combined application of biochar and 20% irrigation and fertilisation reduction as compared to regular amounts, only irrigation water N was decreased in N input path. Whereas, in the N output path, leaching N and  $\text{NH}_3$  volatilisation were both significantly reduced, but no significant change was observed in nitrification and denitrification loss and total N output. In accordance with our hypothesis to some extent, the increasing N surplus and balance in 2018, and decreasing them in 2019 of 0.8W + 0.8F + B treatment as compared to without biochar application (W + F) treatment indicates that under biochar addition, reducing irrigation and fertilisation by 20% could reduce the risk of N loss in the agricultural ecosystem. Furthermore, this practice promoted the sustainability or efficiency of N management as well as the ability to maintain soil fertility in crop systems.

Altogether, the combined application of biochar and reducing 20% fertiliser and 20% irrigation water is an effective way and the most suitable management strategy to decrease N surplus and leaching loss, and maintain crop N uptake in a two-year cycle system on greenhouse Mollisols.

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