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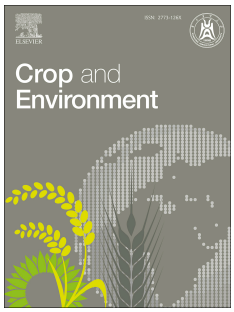

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The response of greenhouse gas emissions, crop yield, and soil health to water scarcity and biochar application in rice cultivation

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ABSTRACT

Projected climate change impacts, such as delayed rainfall and increased drought frequency, threaten rice cultivation and global food security. This study evaluated the effects of water scarcity at critical growth stages and biochar application on greenhouse gas (GHG) emissions, yield, and soil health in Central Thailand using the drought-tolerant cultivar *Pathum Thani* 1.

Treatments included continuous flooding and water scarcity during tillering, reproductive, or both stages, with and without biochar, across wet and dry seasons. Water scarcity significantly reduced methane (CH_4) emissions by inhibiting hydrogenotrophic methanogenesis (*Methanocella*) and acetoclastic methanogenesis (GOM Arc I of *Methanosarcinales*) but increased nitrous oxide (N_2O) emissions via enhanced nitrification. Despite higher N_2O emissions, total GHG emissions, expressed as the global warming potential (GWP), were lower under water-scarce conditions than under continuous flooding, with reductions of 27.1%, 43.0%, and 58.1% during tillering, reproductive, and both stages, respectively. Water scarcity during tillering stage maintained yield, whereas water scarcity during reproductive stage caused a significant reduction in yield. Biochar amendment further mitigated GHG emissions, improved yield (by 12.2%), and enhanced soil health by increasing soil pH, nutrient availability, and soil organic carbon sequestration. Its high porosity and surface area also suppressed methanogenesis and reduced N_2O formation while improving nutrient use efficiency. The strategic use of water restrictions during tillering, combined with biochar, provides a sustainable approach to mitigate GHG emissions, optimize water use, and sustain soil health and productivity. In resource-limited scenarios, prioritizing tillering-stage water scarcity over biochar application is recommended because of its greater GHG mitigation potential.

Keywords: Biochar amendment, Grain yield, Greenhouse gas emissions, Rice, Soil health, Water scarcity

1. Introduction

Anthropogenic greenhouse gas (GHG) emissions have been unequivocally identified as the predominant drivers of global warming and climate change, exerting profound and far-

reaching impacts across environmental, economic, and social systems (IPCC, 2021). Among the diverse sources of GHG emissions, agricultural activities, particularly within agrarian nations, represent a substantial and critical source (IPCC, 2022a). In 2019, Thailand's GHG emissions from agricultural sector were approximately at 28,715 gigagrams of carbon dioxide equivalent (Gg CO_2eq), contributing 7.70% of the nation's total GHG emissions. Within the agricultural sector, rice cultivation alone is responsible for 51.0% of total emissions (ONEP, 2022). The predominant GHG released from rice fields are methane (CH_4) and nitrous oxide (N_2O), which exhibit global warming potentials (GWP) approximately 27 and 273 times that of CO_2, respectively. CH_4 is produced predominantly through the anaerobic decomposition of organic matter facilitated by methanogenic archaea (Conrad, 2002), whereas N_2O is generated through nitrification and denitrification processes under aerobic and anaerobic conditions, respectively (Hayashi et al., 2015). Therefore, mitigating GHG emissions from rice cultivation poses a critical challenge in reducing the future impacts of climate change.

Despite ongoing and future mitigation efforts, the impacts of climate change are inevitable, as indicated by various climate change scenarios (IPCC, 2021). Altered precipitation patterns, such as the absence of rainfall during the wet season or delayed onset of rains, coupled with extreme climatic events, are of particular concern because of their detrimental effects on crop production (IPCC, 2022b; Kumar et al., 2019). Studies from Southeast Asia have demonstrated significant increases in the annual average surface temperature and reductions in precipitation during the wet season, contributing to severe drought conditions across the region (Amnuaylojaroen and Chanvichit, 2019, 2024). These climatic shifts raise concerns regarding food security, heightened water demands, and intensified competition for water resources, particularly in rainfed agricultural areas (Boonwichai et al., 2018; Bouman et al., 2007; Datta et al., 2017).

Thailand also experiences droughts frequently, with future projections indicating increased severity, particularly in agricultural regions (Amnuaylojaroen and Chanvichit, 2024; Boonwichai et al., 2018; Kaewmai et al., 2021). The 2019 drought in Thailand, which resulted in estimated agricultural production losses of approximately 26 billion baht (USD 840 million), was attributed primarily to rice production during the dry season. This reduction was due to government-imposed restrictions on irrigation to prioritize water availability for domestic consumption and ecological management, a decision driven by significantly below-average precipitation (USDA, 2020). These adverse weather conditions have had considerable impacts on crop yields, farmer incomes, and the national economy (Bouman et al., 2007). Field studies evaluating the interplay between crop yield and GHG emissions from rice cultivation under water scarcity, particularly under low rainfall conditions in Thailand, remain limited.

Research conducted in Bangladesh by Moonmoon and Islam (2017) and Hossain et al. (2020) revealed that water deficit conditions and drought stress significantly reduced rice grain yield by affecting key morphological traits, such as plant height, number of effective tillers, spikelets, filled grains, and 1000-grain weight. Drought stress during the panicle initiation stage was identified as a critical factor in yield reduction. Similarly, Zhang et al. (2023) reported that water stress during the heading and flowering stages in China led to average yield reductions of 27.6–46.3% compared to conventional flooding practices. In India, Kumar et al. (2020) demonstrated that cumulative drought stress impaired sugar mobilization, leading to reduced pollen viability and grain yield, with an average yield reduction of 85.7% compared to non-stress conditions. In Southern Thailand, the investigations by Hussain et al. (2022) identified rice genotypes, including Hom Pathum, Sang Yod, Dum Ja, and Pathum Thani 1, as exhibiting high tolerance to drought stress, with reductions in grain yield ranging from 21–52%. However, these studies were conducted under

controlled greenhouse conditions and did not encompass the measurement of GHG emissions. Thus, examining the impact of water scarcity on rice cultivation under field conditions in Thailand, with a focus on GHG emissions, crop yield, and soil health, is of significant scientific interest.

Given the negative impacts of water scarcity on rice cultivation, the application of soil amendments may offer a viable strategy to mitigate these adverse effects. Biochar, in particular, has shown promise as a soil amendment, improving soil quality, increasing grain yield, and reducing GHG emissions (Chew et al., 2020; Jeffery et al., 2011; Sriphirom et al., 2022; Wang et al., 2019; Zhang et al., 2010). The unique properties of biochar, including high porosity and large surface area, enhance water retention, nutrient absorption, and nutrient use efficiency by plants, leading to increased yield production (Chew et al., 2020; Oladele et al., 2019; Oliveira et al., 2017). Biochar's alkalinity can also balance soil pH and improve soil organic C (SOC) sequestration (Koyama and Hayashi, 2019; Sriphirom et al., 2020; Zhang et al., 2020). Additionally, biochar is expected to enhance drought resilience (IPCC, 2022a). Studies by Wang et al. (2019) and Wu et al. (2019) demonstrated that biochar amendments reduced CH_4 emissions by decreasing methanogen populations and increasing methanotrophic activity, thereby minimizing CH_4 production and promoting CH_4 oxidation. Moreover, Cayuela et al. (2013) found that biochar immobilizes NO_3^- in the soil, reducing its availability for N_2O formation. However, the mitigation potential of biochar varies depending on the soil type and application rates (Feng et al., 2012; Wang et al., 2019). Thus, biochar application may provide a strategy to mitigate the adverse effects of water scarcity in rice cultivation.

This study aims to elucidate the effects of water scarcity, imposed during distinct critical phenological stages (tillering, reproductive, or both) with and without biochar application, on GHG emissions, yield performance, and soil conditions. The experiment was conducted over two consecutive growing seasons (one year) using a drought-tolerant rice

variety in a key province of Central Thailand, a nation of global significance in rice production and export. The findings are expected to provide valuable insights into optimizing rice cultivation practices in the context of water scarcity exacerbated by climate change-induced rainfall deficits. This research contributes to the advancement of sustainable agricultural systems by identifying strategies that mitigate GHG emissions while maintaining or enhancing crop productivity and soil health.

2. Materials and methods

2.1. Study site

The study site is situated in Bang Pla Sub-district, Bang Len District, Nakhon Pathom Province, Central Thailand (13˚57'33"N, 100˚09'25"E, at an elevation of 3 m above mean sea level). The soil at the site is classified as Endoaquepts within the Vertisols order, according to the United States Department of Agriculture (USDA) soil taxonomy. The soil texture is characterized as clay, comprising 40.7% sand, 13.3% silt, and 46.0% clay. Baseline physical and chemical properties of the soil, collected from the 0–20 cm depth in April 2023, are detailed in Table 1.

During the study period, from May 2023 to April 2024, the site experienced an annual rainfall of 625 mm, with maximum and minimum air temperatures of 34.5°C and 24.8°C, respectively (Fig. 1A and 2A). Notably, the cumulative annual rainfall was substantially lower than the long-term average of 1,006 mm, reflecting drier-than-average climatic conditions during the study period.

Table 1 here

2.2. Experimental design

This study simulated field conditions of water restriction due to the absence of rainfall for 15 or more consecutive days, representing a precipitation deficit or shortage during rice cultivation (DDPM, 2022). The experiment was conducted during the 2023–2024 growing season, which is characterized by below-average precipitation. Four distinct water regimes were designed: continuous flooding (CO), water scarcity during the tillering stage (DT), water scarcity during the reproductive stage (DR), and water scarcity during both the tillering and reproductive stages (DTR).

All treatments were maintained under continuous flooding with 5 cm of water above the soil surface from 0 to 15 d after transplanting (DAT). The water depth was increased to 10 cm during 16–24 DAT, 55–59 DAT, and at 90 DAT. To induce water scarcity, irrigation was withheld to allow natural drying of the field during the tillering stage (25–54 DAT) and the reproductive stage (60–89 DAT). After 90 DAT, all treatments were naturally dried to prepare for harvest. Water levels above or below the soil surface were monitored using a measuring stick placed within a PVC tube installed in the soil prior to transplanting. The field was irrigated to the target flood level using a pump at fixed intervals (6:00, 13:00, and 18:00). However, rainfall during the wet season partially interfered with the induction of water scarcity during the tillering stage and the drying period before harvest (Fig. 1A and 2A)

The biochar used in this study was derived from bamboo through pyrolysis at 600°C (Sahoo et al., 2021). The bamboo biochar exhibited high porosity and surface area (Odega et al., 2023), with a specific surface area of 192 $m^2\ g^{-1}$ and a specific pore volume of 0.19 $cm^3\ g^{-1}$. Its basic characteristics are presented in Table 1. Biochar was applied at a rate of 20 Mg ha^{-1} (dry weight) per season (Zhang et al., 2010) across all the water regimes. Consequently, the study comprised eight treatments: CO, DT, DR, DTR, CO with biochar application (CO+BI), DT with biochar application (DT+BI), DR with biochar application (DR+BI), and DTR with biochar application (DTR+BI), as illustrated in Fig. S1.

The rice (*Oryza sativa* L.) cultivar Pathum Thani 1 (PTT 1), known for its high drought tolerance and yield potential (Hussain et al., 2022), was cultivated across two growing seasons (wet and dry seasons). The experiment was arranged in a randomized complete block design with three replications, and each plot measured 10 m × 10 m.

2.3. Crop management

Rice cultivation for the wet season was conducted from June 3 to September 29, 2023, and for the dry season, it was conducted from November 18, 2023 to March 17, 2024. During each cultivation period, the soil underwent two plowing operations: moldboard tillage was performed 25 d before transplanting (DBT), followed by harrow tillage 2 DBT. The final tillage operation included levelling the field as thoroughly as possible and removing aboveground residues from the previous season. Only the stubble below the soil surface was incorporated during the initial tillage to prepare the field for the subsequent season. Biochar was incorporated into the soil in the CO+BI, DT+BI, DR+BI, and DTR+BI treatments during the first tillage.

Rice seedlings were germinated in trays for 20 d prior to transplanting. Vigorous seedlings were transplanted at a spacing of 25 cm × 25 cm with five seedlings per hill on June 3, 2023 and November 18, 2023 for the wet and dry seasons, respectively. Mineral fertilizers were applied twice per season: a basal application of a mixed fertilizer (N–P–K: 15–15–15) at rates of 35 kg N ha^{-1}, 35 kg P ha^{-1}, and 35 kg K ha^{-1} at 21 DAT, and a top-dressing of urea at a rate of 55 kg N ha^{-1} at 60 DAT. All treatments received uniform weed control and pesticide applications as required. Harvesting was carried out simultaneously on September 29, 2023 for the wet season (118 DAT), and on March 17, 2024 for the dry season (120 DAT). The detailed calendar of crop management operations is provided in Table S1.

2.4. CH_4 and N_2O emissions analysis

CH_4 and N_2O emissions were quantified using a closed chamber technique throughout the cultivation period and during the fallow phase (Minamikawa et al., 2015; Sriphirom et al., 2024a). Gas sampling was conducted using acrylic chambers of varying volumes: 0.13, 0.25, or 0.45 m^3 during the growing season, depending on the rice height, and 0.06 m^3 during the fallow period. Chambers were installed in triplicate per plot before transplanting and remained in situ throughout the cultivation season to minimize soil disturbance. During gas sampling, the chamber body was sealed onto the base.

Air samples from the chamber headspace were collected using a 30 mL plastic syringe at intervals of 0, 5, 10, 15, and 20 min after chamber closure. These samples were then transferred into 25 mL evacuated glass vials. Routine analysis was conducted weekly, with additional sampling during fertilizer application and periods of extreme drought. Gas sampling was conducted between 09:00 and 11:00 during the cultivation period and between 12:00 and 14:00 during the fallow period (Minamikawa et al., 2015). Air temperature inside the chamber during sampling was recorded for emission rate calculations.

Gas concentrations were analyzed using a gas chromatography (GC) (7890B, Agilent Technologies, Inc., USA) equipped with a flame ionization detector (FID) and an electron capture detector (ECD) operating at 300°C with a HaySep Q packed column. Nitrogen (N) and helium served as carrier gases for the GC-FID and GC-ECD, respectively (Chidthaisong et al., 2018). CH_4 and N_2O fluxes were calculated from the increase in gas concentration (ppmv) over the 20-min sampling period using linear regression methods as described by Sriphirom et al. (2024b). Seasonal cumulative emissions were estimated through successive linear interpolation and numerical integration of data collected on sampling days (Sriphirom et al., 2024b). CH_4 and N_2O emissions were converted to CO_2 equivalents (CO_2eq) using GWP factors over a 100-year horizon: 27 for CH_4 and 273 for N_2O (IPCC, 2021).

226

227 *2.5. Soil property analysis*

228 Soil samples were collected at a depth of 20 cm at three intervals: pre-cultivation
229 (April 2023; Table 1), post-wet season (October 2023), and post-dry season (April 2024).
230 Triplicate samples per plot were obtained using a 100 cm^3 stainless-steel core sampler, air-
231 dried, sieved (2 mm), and analyzed for pH (1:1 soil/water), electrical conductivity (EC; 1:5
232 soil/water extraction), organic C (OC; Walkley & Black method), and organic matter (OM;
233 calculated as 1.724 × OC). Available P (Bray II method), exchangeable K (ammonium acetate
234 extraction), cation exchange capacity (CEC; ammonium saturation), moisture content and
235 bulk density were measured gravimetrically (105°C drying for 48 h) using a forced-air
236 convection oven (Redline RF 53, Germany). Analyses followed protocols described by Pansu
237 and Gautheyrou (2006).

238 Total C and N were quantified using a CHN analyzer (LECO Corporation, USA) at
239 combustion temperatures of 950–1050°C (Joseph, 2016). Ammonium (NH_4^+) was measured
240 using ion chromatography (IC) with a Dionex Integrion HPIC system (Thermo Scientific,
241 USA) equipped with Dionex IonPac CG16 guard and CS16 analytical columns (Thomas et
242 al., 2002). Nitrate (NO_3^-) was analyzed using IC with Dionex IonPac AG11 guard and AS11
243 analytical columns (Morales et al., 1998). SOC sequestration was estimated as the product of
244 SOC concentration, sampling depth, and bulk density following Lee et al. (2009).

245 During cultivation, soil redox potential (Eh), pH, temperature, NO_3^-, and dissolved
246 organic C (DOC) were monitored at 7–10 d intervals at a depth of 0–10 cm. Eh, pH, and
247 temperature were recorded using a pH/ORP sensor (YSI Professional Plus, USA). DOC was
248 extracted with K_2SO_4 (Dong et al., 2013) and quantified using a total organic C (TOC)
249 analyzer (Multi N/C 2100, Germany) equipped with a non-dispersive infrared (NDIR)
250 detector.

2.6. Soil microbial abundance and community analysis

Soil samples (0–10 cm) were collected (Lee et al., 2015) at 25 DAT and 65 DAT for microbial abundance and community structure analysis. DNA was extracted from 1 g of soil using the DNeasy PowerSoil Pro kit (Qiagen, Germany), with quality confirmed by agarose gel electrophoresis and concentration measured using a NanoPhotometer N60 Touch (Implen, Germany).

Microbial abundances were quantified using quantitative real-time polymerase chain reaction (qRT-PCR) on a CFX96 Touch Real-Time PCR Detection System (Bio-Rad, USA). Each 20-μL reaction contained 10 μL of Luna® Universal qPCR Master Mix (NEB, USA), 0.4 μL each of forward and reverse primers, 1.0 μL of DNA template (10–20 ng), and 8.2 μL of sterile water. Primers sets targeted the methyl coenzyme M reductase (*mcr*A) gene for methanogens (mcrA-F/mcrA-R; Luton et al., 2002), the particulate methane monooxygenase (*pmo*A) gene for methanotrophs (A189F/Mb661R; Kolb et al., 2003), the ammonia monooxygenase (*amo*A) gene for nitrifying bacteria (amoA-1F/amoA-2R; Rotthauwe et al., 1997), and the nitrite reductase (*nir*K) and nitrous oxide reductase (*nos*Z) genes for denitrifying bacteria (nirKF1aCu/nirKR3Cu; Hallin and Lindgren, 1999; nosZ2F/nosZ2R; Henry et al., 2006). Thermal cycling conditions are detailed in Table S2, with standard curves achieving $R^2 > 0.99$.

Bacterial diversity during the reproductive stage was assessed via 16S ribosomal RNA (rRNA) gene amplification using primers specific to methanogenic archaea (1106F/1378R; Watanabe et al., 2007) and type I (197F/533R; Tsien et al., 1990) and type II methanotrophic bacteria (142F/533R; Tsien et al., 1990). Thermal cycling conditions were: for methanogenic archaea, 98°C for 1 min, followed by 30 cycles of 95°C for 10 s, 50°C for 30 s, and 72°C for 35 s, with a final extension at 72°C for 5 min; for methanotrophic bacteria, 94°C for 2 min,

followed by 35 cycles of 94°C for 30 s, 60°C for 30 s, and 72°C for 45 s, with a final extension at 72°C for 5 min. Sequencing libraries were prepared using the Nextera XT Index kit (Illumina, USA) and sequenced on a MiSeq platform (300-bp paired-end) with the MiSeq Reagent Kit v3 (600 cycles).

Amplicon sequence analysis was conducted using Quantitative Insights Into Microbial Ecology (QIIME2) version 2022.2 (Bolyen et al., 2019). Adapters were trimmed with q2-cutadapt (Martin, 2011), and DADA2 (Callahan et al., 2016) was used for error correction and sequence variant calling. Taxonomic classification was performed using the SILVA database version 138 (Bokulich et al., 2018; Quast et al., 2013). Rarefaction ensured uniform sequencing depth, and diversity metrics were computed. Heatmaps were visualized using the Multiple Experiment Viewer version 4.9.0 (Howe et al., 2010).

2.7. Crop growth, yield, and water use measurements

In this study, various parameters of crop growth and yield were assessed, including plant height, tiller count, panicle count, numbers of filled and unfilled grains, 1000-grain weight, aboveground biomass, and grain yield. Plant growth, height, and tiller count were measured and recorded manually throughout the cultivation period. Yield components were determined from a 3 m^2 area in each plot on the designated harvest day, and the dry weights were subsequently determined.

Water use in rice cultivation was quantified by summing the volumes of irrigation and rainfall. Irrigation water was supplied using a water pump (WCM-3705FS, 5HP, Mitsubishi, Thailand), and its application was monitored with a multi-jet water meter (GMK 15 R80, Asahi, Thailand) throughout both the land preparation and cultivation periods. Daily rainfall was recorded using a tipping bucket rain gauge positioned within the study area. Total water use was defined as the cumulative volume of irrigation and rainwater applied to the field,

ensuring that it did not surpass the predetermined flood levels (5 cm or 10 cm, as depicted in Fig. S1). Additionally, irrigation water productivity was estimated by calculating the ratio of marketable grain yield to the total amount of irrigation water used (IWU) according to Eq. (1), which reflects the efficiency of irrigation practices (Fernández et al., 2020).

$$\text{Irrigation water productivity (kg yield m}^{-3}) = \frac{\text{Grain yield (kg ha}^{-1})}{\text{IWU (m}^3\text{ ha}^{-1})} \quad (1)$$

The impact of water scarcity on downstream human users and ecosystems was quantified as the water scarcity footprint (Kaewmai et al., 2021). This footprint, expressed in m^3 H_2Oeq ha^{-1}, was calculated by multiplying the monthly volume of irrigation water used in rice cultivation (from land preparation to harvest) by the monthly water stress index (WSI) according to Eq. (2) (Silalertruksa et al., 2017). For the Tha Chin watershed, where Nakhon Pathom Province is located, the WSI values for each month were as follows: January, 1.00; February, 1.00; March, 0.94; April, 0.04; May, 0.03; June, 0.42; July, 0.76; August, 0.82; September, 0.28; October, 0.04; November, 0.06; and December, 0.69 (Gheewala et al., 2018).

$$\text{Water scarcity footprint (m}^3\text{ H}_2\text{Oeq ha}^{-1}) = \text{Monthly volume of IWU (m}^3\text{ ha}^{-1}) \times \text{Monthly WSI} \quad (2)$$

2.8. Statistical analysis

Principal coordinate analysis (PCoA) of microbial community composition was conducted and visualized using the vegan R package (Dixon, 2003). Functional predictions of microbial communities were performed using phylogenetic investigation of communities by reconstruction of unobserved states (PICRUSt) on the Majorbio I-Sanger cloud platform (http://www.i-sanger.com/). KEGG Orthology data were utilized to estimate the abundance of

key functional enzymes associated with methanogens and methanotrophs, and the relative abundance of these genera was illustrated according to their functional roles.

All results are presented as means ± standard errors. Differences among treatments were assessed using one-way analysis of variance (ANOVA), followed by Tukey's honestly significant difference (HSD) test for post hoc comparisons at a 95% confidence level ($P < 0.05$). Statistically significant differences are indicated by distinct letter annotations. Combined means of key parameters, including CH_4 emissions, N_2O emissions, GWP, grain yield, SOC stock, and irrigation water productivity, were analyzed to evaluate the effects of water management, biochar amendment, and growing season. Statistical analyses were conducted using SPSS version 29.0 (IBM SPSS Statistics, New York, USA).

Pearson's correlation analysis and redundancy analysis (RDA) were employed to investigate the relationships between GHG emission rates (CH_4, N_2O, and GWP) and environmental and biological factors, including soil properties (pH, Eh, temperature, and NO_3^- and DOC contents), plant characteristics (rice height and tiller number), water levels, and microbial abundance (methanogens, methanotrophs, nitrifiers, and denitrifiers). Additionally, the association between CH_4 emission rates and the community composition of methanogenic archaea and methanotrophic bacteria was analyzed. Data for these analyses were collected concurrently throughout the study period. RDA was performed using the vegan R package version 4.3.1 (Kindt, 2020), while Pearson's correlation analysis was conducted using SPSS as described above.

3. Results

3.1. CH_4 and N_2O emissions

Water scarcity during the cultivation season significantly reduced anaerobic soil conditions, substantially mitigating CH_4 emissions. Extended water scarcity further increased

351 CH_4 emission mitigation (Fig. 1B–M), as evidenced by a strong positive correlation ($P < 0.01$)
352 between CH_4 emissions and field water level (Fig. 3A and Table S3). Specifically, compared
353 to continuous flooding (CO and CO+BI), water scarcity during the tillering stage (DT and
354 DT+BI), reproductive stage (DR and DR+BI), and both stages combined (DTR and DTR+BI)
355 reduced seasonal cumulative CH_4 emissions by 20.3%, 41.8%, and 57.6%, respectively, in the
356 wet season, and by 36.6%, 47.9%, and 64.9%, respectively, in the dry season (Table 2).

357 Conversely, water scarcity increased N_2O emissions (Fig. 2B–M), as indicated by a
358 significant negative correlation ($P < 0.01$) between N_2O emissions and field water levels (Fig.
359 3A and Table S3). Seasonal cumulative N_2O emissions under water scarcity during the
360 tillering stage, reproductive stage, and both stages combined increased by 25.1%, 33.2%, and
361 54.3%, respectively, in the wet season and by 31.1%, 50.4%, and 71.7%, respectively, in the
362 dry season relative to continuous flooding. Compared with the wet season, the extended
363 aerobic periods associated with water scarcity during the dry season contributed to lower CH_4
364 emissions but higher N_2O emissions (Table 2).

365 When GHG emissions were assessed in terms of CO_2 equivalents, referred to as GWP,
366 water scarcity consistently demonstrated net environmental benefits, largely due to substantial
367 reductions in CH_4 emissions (Table 2). These findings suggest that rice cultivation under
368 water-scarce conditions, as anticipated under climate change scenarios, represents a more
369 sustainable and environmentally friendly practice.

370 Biochar amendment also exhibited mitigation potential for both CH_4 and N_2O
371 emissions during some periods of the cultivation season (Fig. 1B–M and 2B–M). Compared
372 with no biochar application, biochar application reduced seasonal cumulative CH_4, N_2O, and
373 GWP by 12.0%, 15.3%, and 12.2%, respectively, in the wet season and by 11.9%, 18.7%, and
374 12.2%, respectively, in the dry season (Table 2). Although biochar presents itself as a viable

GHG mitigation strategy, its effectiveness is often surpassed by that of water management interventions.

Fig. 1. here

Fig. 2. here

Table 2 here

Fig. 3. here

3.2. Microbial abundances and diversity

Soil samples analyzed under various management practices revealed that microbial abundance and diversity underwent more pronounced changes during the reproductive stage (65 DAT) than during the tillering stage (25 DAT) (Fig. 4). During the reproductive stage, water scarcity likely reduced the abundance of methanogens and denitrifying bacteria while promoting the proliferation of methanotrophic and nitrifying bacterial populations. Microbial community patterns under water scarcity during the reproductive stage (DR and DR+BI) closely resembled those observed under continuous flooding (CO and CO+BI), likely reflecting similar soil conditions during the sampling period. However, microbial patterns under water scarcity at both stages (DTR and DTR+BI) were consistent with those observed under water scarcity during the tillering stage (DT and DT+BI). Biochar application generally increased the abundance of all the microbial groups, although most of these increases were not statistically significant (Fig. 4).

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Fig. 4. here

Soil drying induced by water scarcity reduced the abundances of methanogenic archaea, including *Methanocella* (5.28%), *Methanocellaceae;* Rice Cluster I (4.43%), *Methanosarcinales;* GOM Arc I (8.62%) and *Methanosaeta* (1.03%). In biochar-amended soils, the abundances of *Methanocella* (6.45%), *Methanocellaceae;* Rice Cluster I (4.07%), *Methanosarcinales;* GOM Arc I (3.35%), and *Methanosaeta* (3.00%) were also reduced (Fig. 5A–B). Conversely, water scarcity stimulated the abundances of methanotrophic bacteria, resulting in increases of 2.65%, 6.20%, 24.3%, 12.4%, 45.6%, 33.4%, and 26.4% for *Methylocystis*, *Methylosinus*, *Methylocella*, *Methylocapsa*, *Methylomonas*, *Methylocaldum*, and *Methylosarcina*, respectively. In biochar-amended soil, these increases were 16.6%, 38.6%, 37.2%, 42.4%, 80.1%, 55.5%, and 40.7%, respectively (Fig. 5C–D).

Fig. 5. here

Pearson's correlation analysis confirmed a significant positive association between the *mcr*A gene of methanogenic archaea and both CH_4 emissions and GWP ($P < 0.01$) (Fig. 3B and Table S4). Among methanogens, GOM Arc I of *Methanosarcinales* was a primary contributor to CH_4 emissions, while *Methylocystis*, *Methylosinus*, and *Methylomonas* were key contributors to CH_4 oxidation, driving treatment-dependent variations in CH_4 emissions (Fig. 3C and Table S5). Furthermore, the *amo*A gene of ammonia oxidizing bacteria (AOB) significantly contributed to variations in N_2O emissions via nitrification during dry soil conditions ($P < 0.05$; Fig. 3B and Table S4).

3.3. Soil properties

Crop management practices altered soil concentrations of soluble NO_3^- and DOC during cultivation. Under flooding-induced anaerobic conditions, NO_3^- concentrations decreased, peaking post-fertilization. In contrast, water scarcity enhanced nitrification, increasing NO_3^- levels during soil desiccation (Fig. 6A–B), which elevated N_2O emissions during fertilization and dry periods (Fig. 2). DOC concentrations were initially high during early cultivation but declined due to microbial assimilation, resurging after the reproductive stage and continuing through the harvest preparation period. However, DOC levels decreased during soil desiccation (Fig. 6C–D), coinciding with an increase in soil Eh under water scarcity (Fig. S2). Biochar amendment under all water scarcity conditions resulted in greater accumulation of NO_3^- and DOC (Fig. 6), along with a marginal increase in soil pH and Eh (Fig. S2–3), compared with those in soils without biochar.

Fig. 6. here

Post-harvest analysis revealed that water scarcity, whether during a single or both growth stages, had no significant effect on key soil characteristics. In contrast, biochar application increased soil pH, EC, OM, OC, total C, nutrient levels, and SOC sequestration due to its alkalinity, high C content, porosity, and surface area, although the impact on nutrient levels was not statistically significant. These effects were more pronounced during the dry season, reflecting the cumulative impact of biochar over both wet and dry seasons (Table 3). Compared with the soil without biochar, its application significantly increased SOC stock by an average of 12.7% in the wet season and 19.9% in the dry season. No significant differences in biochar impacts were observed across water regimes (Table 3). These findings suggest that while water scarcity did not adversely affect soil quality post-harvest, biochar incorporation substantially improved soil health.

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Table 3 here

3.4. Crop growth and yield

Water scarcity during rice cultivation significantly influenced growth and yield, particularly when deficits coincided with critical phenological stages. Growth phenology varied across water scarcity treatments, with a 2–4 d difference (Table S1). Water deficits during the tillering phase preserved tiller production, whereas deficits during the reproductive phase reduced tiller numbers, consistently across both wet and dry seasons (Fig. S4A–B). Water scarcity decreased plant height in all treatments, regardless of season (Fig. S4C–D), but biochar ameliorated the adverse effects on plant height without influencing tiller production (Fig. S4).

Water scarcity during the tillering stage, irrespective of seasonality, preserved yield components—including panicle number, grain weight, grain number, and grain yield—similar to continuous flooding, regardless of biochar application. However, water scarcity during the reproductive stage significantly reduced grain yield and its components across both seasons. While biochar application tended to increase crop yields, these increases were not statistically significant, averaging 5.22% in the wet season and 7.78% in the dry season (Table 4).

Table 4 here

Despite reduced grain yield under water scarcity during the reproductive stage (DR, DTR, DR+BI, and DTR+BI), these treatments mitigated GHG emissions, resulting in lower GHG emissions per kilogram of grain produced compared to continuous flooding. Water scarcity during the tillering stage (DT and DT+BI) and both tillering and reproductive stages

(DTR and DTR+BI) further decreased GHG emissions per unit of grain yield. Biochar enhanced this reduction in GHG emissions and potential yield (Table 4), making water scarcity during the tillering stage, in conjunction with biochar application, a recommended strategy for optimizing food production while minimizing environmental impacts.

3.5. Water use, water productivity, and water scarcity footprint

Water scarcity imposed during the tillering stage, reproductive stage, and combined stages significantly reduced irrigation water in the wet season, enhancing irrigation water productivity. Conversely, in the dry season, higher irrigation water usage under water scarcity treatments decreased irrigation water productivity. Notably, water scarcity during the tillering stage achieved the highest irrigation water productivity in the dry season, attributed to reduced water use and higher yields compared with other treatments.

Water scarcity also reduced the water scarcity footprint, benefiting downstream users and ecosystems. Specifically, water scarcity during the tillering stage, reproductive stage, and combined stages reduced the water scarcity footprint by an average of 1,310, 1,725, and 2,980 m^3 H_2Oeq ha^{-1} in the wet season and by 1,125, 1,730, and 2,935 m^3 H_2Oeq ha^{-1} in the dry season, respectively (Table 5). Biochar application did not significantly affect irrigation water use, irrigation water productivity, or water scarcity footprint (Table 5).

Table 5 here

4. Discussion

4.1. Water scarcity mitigated GHG emissions more significantly than biochar application

Water scarcity, exacerbated by climate change-induced rainfall delays, is projected to intensify across regions like Thailand, posing challenges for agriculture while simultaneously

offering opportunities to mitigate GHG emissions in rice cultivation (Amnuaylojaroen and Chanvichit, 2024; USDA, 2020). This study revealed a positive correlation between CH_4 emissions and field flooding ($r = 0.645$, $P < 0.01$; Table S3), with water scarcity significantly reducing CH_4 emissions (Table S6). Imposing water scarcity during the tillering or reproductive stages reduced CH_4 emissions by 28.4–45.0%, while applying it during both stages achieved a 61.1% reduction in CH_4 emissions compared with continuous flooding. These findings align with those of Zhang et al. (2024), who reported 70–90% reductions in CH_4 emissions under water-saving practices.

The mitigation of CH_4 emissionswas attributed to suppressed methanogenesis, including reduced activity of hydrogenotrophic (*Methanocella*) and acetoclastic (GOM Arc I of *Methanosarcinales*) methanogenesis (Fig. 5A–B) (Mohamad Shahimin et al., 2021). This suppression is consistent with diminished *mcr*A gene abundance under prolonged desiccation and alternate wetting and drying (AWD) practices (Hester et al., 2022; Reim et al., 2017). While methanogen abundance during the tillering stage did not decrease significantly, microbial connectivity and the abundance of methanogenic drivers within the microbial network were reduced (Zhang et al., 2024), leading to a significant reduction in CH_4 emissions during the tillering stage. Enhanced CH_4 oxidation by methanotrophs further contributed to CH_4 reductions ($P < 0.01$) (Fig. 3C, and Table S5), particularly by type I methanotrophs (Fig. 5C–D). Type I methanotrophs (*Methylocaldum*, *Methylomonas*, and *Methylosarcina*) oxidize CH_4 via methane monooxygenases (MMOs) through the ribulose monophosphate (RuMP) pathway under oxygen-rich conditions and require N for growth. Type II methanotrophs (*Methylocella*), which employ the serine pathway in CH_4-rich and oxygen-limited environments (Chidambarampadmavathy et al., 2015), also supported CH_4 oxidation.

Conversely, water scarcity increased N_2O emissions due to intensified nitrification and denitrification processes (Hayashi et al., 2015), driven by NH_4^+ depletion and NO_3^-

accumulation and associated with increased activity of nitrifying bacteria (Oo et al., 2018; Qiu et al., 2022; Wu et al., 2023). The positive correlation between N_2O emissions and *amo*A-AOB gene abundance (Table S4) indicates the role of nitrifying bacteria. N fertilization, which increased soil NO_3^- availability, further exacerbated N_2O emissions under drained conditions. The most pronounced increases in N_2O emissions occurred during the reproductive stage compared to the tillering stage, attributed to higher activity and abundance of nitrifying bacteria (Fig. 4C), as well as greater rice height and tiller number (Table S3).

Despite the rise in N_2O emissions, the reduction in CH_4 emissions dominated the GHG profile, resulting in an overall reduction in GWP, as shown by the linear correlation between CH_4 emissions and GWP ($P < 0.01$) (Fig. 5 and Tables S3–S4). The greatest GWP reductions were observed during the dry season, when prolonged drying periods were uninterrupted by rainfall (Table S6). Allowing paddy fields to dry during the cultivation season thus represents an effective strategy for mitigating GHG emissions from rice cultivation.

Biochar application further enhanced GHG mitigation (Table S6), reducing emissions by an average of 12.2%. Bamboo biochar applied at 20 Mg ha^{-1} decreased CH_4 emissions by suppressing hydrogenotrophic and acetoclastic methanogenesis, targeting *Methanocella*, Rice Cluster I of *Methanocellaceae*, and GOM Arc I of *Methanosarcinales* (Fig. 5A–B). Biochar increased electron acceptor availability and enhanced soil Eh under anaerobic conditions (Fig. S2), thereby slowing CH_4 production (Sriphirom et al., 2022). This effect was facilitated by the high surface area and porosity of biochar, which increased the availability of oxygen and other oxidants, especially within the rhizosphere (Chew et al., 2020; Joseph et al., 2013; Oliveira et al., 2017). The improved soil oxygenation, combined with the habitat provided for microbes, stimulated CH_4 oxidation by methanotrophs (Chen et al., 2017; Han et al., 2016), including *Methylosinus*, *Methylocapsa*, *Methylomonas*, *Methylocaldum*, and *Methylosarcina* (Fig. 5C–D).

Biochar also mitigated N_2O emissions (Table S6) by decreasing NO_3^- availability through immobilization on its surface, thereby limiting N availability for N_2O formation (Cayuela et al., 2013) and facilitating N_2O reduction to N_2 in anaerobic soils (Aamer et al., 2020; Cayuela et al., 2014). Aamer et al. (2020) demonstrated that biochar mitigated N_2O emissions by increasing the abundance of *nos*Z and *nir*K genes and elevating soil pH, which aligns with findings of this study (Fig. 4D–E). Increased *nos*Z-related bacterial abundances led to greater N_2O consumption, whereas *nir*K-related bacteria promoted N_2 production over N_2O production. However, the extent of biochar's mitigation effects varies depending on soil type, feedstock origin, biochar production conditions, and application rate (Feng et al., 2012; Zhang et al., 2010).

4.2. Effects of water scarcity and biochar on rice growth and yield

This study demonstrates that water scarcity during the tillering stage and biochar application, individually and combined, can sustain crop growth and yield comparable to conventional practices by preserving key yield components, including tiller numbers, filled grain count, and 1,000-grain weight (Table 4 and Fig. S4). Norton et al. (2017) demonstrated that AWD—a water management approach allowing the soil to dry naturally to a depth of approximately 15–20 cm during the tillering stage—maintains tiller numbers and yields equivalent to continuous flooding by promoting root system development (Thakur et al., 2011) and increasing leaf abscisic acid concentrations, which are critical for tiller retention (Howell et al., 2015; Norton et al., 2017). AWD also optimizes shoot-root activity, regulates vegetative growth, and modulates hormone signaling pathways, thereby stabilizing yield (Davies et al., 2011). These findings are corroborated by studies conducted across diverse regions, including the USA (LaHue et al., 2016) and India (Oo et al., 2018).

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However, water scarcity during the reproductive stage significantly reduces yield (Tables 4 and S6). Drought stress during flowering impairs rice physiology, with reductions in spikelet number and grain filling driving yield losses. Lower leaf water potential under drought hinders panicle development, whereas elevated soil temperatures under drought conditions suppress root and microbial activity, accelerate leaf senescence, and diminish photosynthetic, stomatal conductance, and transpiration (Yang et al., 2019). Strategic inclusion of drying phases during the tillering stage, as part of the AWD methodology, can mitigate these adverse impacts (Siopongco et al., 2013).

Biochar enhances soil-water-nutrient dynamics through its high porosity and large surface area (Oladele et al., 2019; Uchida et al., 2019). In this study, biochar applied at 20 Mg ha^{-1} increased grain yield by 5.22% and 7.78% in the wet and dry seasons, respectively (Table 4), consistent with Zhang et al. (2012), who observed similar yield increases in China. Yield improvement is attributed to the ability of biochar to increase nutrient storage (Table 3), enhance nutrient uptake (Joseph et al., 2010), and reduce plant energy demands for nutrient acquisition. Root exudates diffusing into biochar pores stimulate microbial activity, thereby increasing nutrient availability for uptake (Bhattacharjya et al., 2015; Chew et al., 2020). Biochar's interaction with the rhizosphere further facilitates nutrient uptake via root hairs and water-filled macropores (Joseph et al., 2013). However, biochar efficacy depends on its physicochemical properties, which are influenced by feedstock type and production conditions (Chen et al., 2021). Its benefits are more pronounced in low-fertility soils, such as acidic, nutrient-deficient, and coarse-textured soils (Bekchanova et al., 2024; Jeffery et al., 2011). Long-term studies are essential to fully evaluate biochar's potential to sustainably enhance rice productivity (Zhang et al., 2020).

4.3. Biochar application improved soil quality

599 Biochar is a soil amendment distinguished by its unique properties, including a
600 combination of organic, carbonate, and inorganic alkalinities. These characteristics enable
601 biochar to effectively modify soil pH, particularly in acidic soils (Fidel et al., 2017). For
602 example, Zhang et al. (2012) reported that biochar with a pH (H_2O) of 10.4, applied at rates
603 of 10, 20, and 40 Mg ha^{-1}, increased soil pH by 0.21, 0.24, and 0.30 units, respectively. The
604 high porosity and surface area of biochar, facilitated by mechanisms such as hydrogen
605 bonding, cation bridging, covalent bonding, hydrophobic interactions, and oxygenated
606 functional groups (e.g., carboxylic, phenolic, and hydroxyl groups), contribute to increased
607 soil CEC (Adhikari et al., 2024) and improved nutrient availability (Chew et al., 2020; Joseph
608 et al., 2010). Adhikari et al. (2024) further demonstrated that biochar derived from hardwood
609 or cellulosic biomass significantly enhanced the CEC of clayey soils, functioning as a
610 reservoir of soil nutrients and serving as a potential slow-release fertilizer. Similarly,
611 Bekchanova et al. (2024) observed that biochar application increased soil nutrient
612 availability—namely, N, P, and K—by 36%, 34%, and 15%, respectively, and also enhanced
613 soil CEC by 18%.

614 The high C content of biochar, particularly in its stabilized form, is instrumental in
615 long-term SOC sequestration. Stabilized C in biochar is resistant to biological and chemical
616 degradation, allowing for gradual decomposition and sustained enhancement of soil C levels
617 (Tables 3 and S6) (Lehmann, 2007). Ding et al. (2023) reported that biochar amendment
618 significantly increased native and recalcitrant SOC contents, with 39%–51% of the biochar
619 remaining in the topsoil (0–30 cm) even after 11 years. Additionally, the potential for SOC
620 sequestration was notably greater during the dry season, likely due to the residual
621 accumulation of biochar applied during the preceding wet season.

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4.4. Water scarcity affected irrigation water productivity and decreased water scarcity footprint

Water scarcity, driven by restricted irrigation and inadequate rainfall, serves as a proxy for the anticipated impacts of climate change, substantially reducing water use in rice cultivation. While water scarcity reduced grain yield, it increased irrigation water productivity, particularly during the wet season (Tables 5 and S6). During this period, water scarcity achieved grain yields statistically comparable to those under continuous flooding but with reduced irrigation water input. In contrast, during the dry season, higher evaporation rates necessitated increased irrigation across all treatments, resulting in irrigation water productivity levels similar to those of continuous flooding. Notably, the highest yield per cubic meter of irrigation water during the dry season was observed under the water scarcity imposed at the tillering stage (DT and DT+BI). These results corroborate the findings of Hussain et al. (2022), who reported reduced water use efficiency under drought stress in Southern Thailand.

Water scarcity during the tillering stage emerges as a viable strategy for balancing water savings and yield, highlighting the importance of managing water scarcity during critical growth stages. Optimizing soil drying levels and durations can reduce irrigation water use, enhance water use efficiency, and maintain food security while mitigating GHG emissions. Mallareddy et al. (2023) emphasized that integrated approaches—such as maximizing rainfall utilization, optimizing limited irrigation, and improving crop water use efficiency —can further enhance water productivity. These strategies include upgrading irrigation systems, land levelling, conjunctive use of surface and groundwater, rotational water distribution, and ensuring access to drought-tolerant seeds and other critical inputs. Such measures are essential for sustaining agricultural productivity under climate change.

Strategic water restrictions during the tillering stage, without significant yield loss, also reduce the water scarcity footprint, increasing water availability for downstream human

and ecosystem needs. This finding aligns with Sriphirom et al. (2019), who demonstrated that AWD practices reduce the water scarcity footprint, increasing water availability for non-agricultural purposes.

5. Conclusions

Rice cultivation under water scarcity driven by climate change poses substantial challenges, necessitating the adoption of environmentally sustainable practices that mitigate GHG emissions while maintaining or enhancing crop yield and soil quality to ensure food security. The use of biochar as a soil amendment has been identified as a promising strategy for mitigating these challenges and supporting farmer adoption under water-limited conditions. This study demonstrated that imposing water scarcity during the tillering stage effectively sustains crop growth and yield comparable to those achieved under continuous flooding. Grain yield, a critical determinant for farmer acceptance, was maintained under this water management practice, which also reduced GHG emissions and improved irrigation water productivity by an average of 20.7% and 51.1% during the wet season and 34.4% and 23.4% during the dry season, respectively, relative to continuous flooding. Furthermore, the combined application of biochar and water scarcity during the tillering stage further enhanced GHG mitigation and increased grain yield by an average of 12.7% and 2.4%, respectively, compared with those in soils not amended with biochar. Soil health metrics were also significantly improved with biochar application, as evidenced by increases in soil pH, CEC, nutrient availability, and SOC sequestration. These benefits highlight the dual role of biochar in improving soil quality and contributing to long-term climate resilience in rice cultivation systems. Consequently, the implementation of water restrictions during the tillering stage is recommended as a practical strategy for supporting net-zero GHG emissions with minimal adverse effects on water availability, even in scenarios where biochar production capacity is

limited. However, in regions where biochar resources are accessible, its application should be prioritized to maximize mitigation potential and improve soil quality, thereby supporting sustainable agricultural production in the context of climate change.

Abbreviations

*amo*A	ammonia monooxygenase
AOB	ammonia-oxidizing bacteria
AWD	alternate wetting and drying
CH_4	methane
DAT	days after transplanting
DOC	dissolved organic carbon
Eh	soil redox potential
GHG	greenhouse gas
GWP	global warming potential
IWU	irrigation water use
*mcr*A	methyl coenzyme M reductase
NH_4^+	ammonium
*nir*K	nitrite reductase
NO_3^-	nitrate
N_2O	nitrous oxide
*nos*Z	nitrous oxide reductase
*pmo*A	particulate methane monooxygenase
SOC	soil organic carbon

Availability of data and materials

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Not applicable.

Authors' contributions

P.S.: conceptualization, methodology, investigation, data curation, writing of original draft, reviewing, and editing, funding acquisition, and project administration; B.R.: methodology and investigation; R.O. and A.C.: conceptualization, reviewing, and editing.

Declaration of competing interest

The authors declare that they have no conflicts of interest.

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Table 1

Basic physical and chemical properties of the soil collected prior to the study and biochar.

Parameter (unit)	Soil	Biochar
Sand (%)	40.7 ± 0.5	–
Silt (%)	13.3 ± 0.1	–
Clay (%)	46.0 ± 0.5	–
pH [H_2O]	6.21 ± 0.06	8.64 ± 3.70
Electrical conductivity (dS m^{-1})	0.80 ± 0.06	–
Organic matter (%)	1.13 ± 0.66	–
Organic carbon (%)	0.66 ± 0.03	–
Total carbon (%)	1.65 ± 0.03	68.8 ± 2.0
Total nitrogen (%)	0.11 ± 0.01	0.65 ± 0.04
Ammonium (mg kg^{-1})	8.67 ± 0.49	–
Nitrate (mg kg^{-1})	86.5 ± 4.9	–
Available phosphorus (mg kg^{-1})	39.1 ± 1.9	8,467 ± 404
Exchangeable potassium (mg kg^{-1})	96.9 ± 5.2	9,367 ± 493
Cation exchange capacity (cmol kg^{-1})	10.8 ± 0.4	41.2 ± 2.5
Bulk density (g cm^{-3})	1.43 ± 0.03	–
Moisture content (%)	19.5 ± 0.7	5.38 ± 0.06
Soil organic carbon sequestration (Mg ha^{-1})	18.8 ± 0.8	–
Ash content (%)	–	4.27 ± 0.37
Specific surface area (m^2 g^{-1})	–	192 ± 9
Specific pore volume (cm^3 g^{-1})	–	0.19 ± 0.02

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Table 2

Seasonal cumulative CH_4 and N_2O emissions and global warming potential (GWP) for the wet and dry seasons.

Treatment	Wet season			Dry season		
	CH_4 emissions (kg CH_4 ha^{-1})	N_2O emissions (g N_2O ha^{-1})	GWP (Mg CO_2 eq. ha^{-1})	CH_4 emissions (kg CH_4 ha^{-1})	N_2O emissions (g N_2O ha^{-1})	GWP (Mg CO_2 eq. ha^{-1})
CO	98.4 ± 9.5 a	257 ± 12 c	2.73 ± 0.25 a	92.2 ± 4.0 a	254 ± 8 d	2.56 ± 0.11 a
DT	77.2 ± 4.7 b	296 ± 17 bc	2.16 ± 0.13 b	58.9 ± 4.0 b	326 ± 17 bc	1.68 ± 0.11 b
DR	56.0 ± 2.7 cd	313 ± 11 b	1.60 ± 0.07 cd	48.0 ± 1.4 bc	342 ± 9 b	1.39 ± 0.04 bc
DTR	40.6 ± 1.5 de	366 ± 24 a	1.20 ± 0.04 de	32.3 ± 2.5 de	394 ± 8 a	0.98 ± 0.04 de
CO+BI	83.7 ± 8.7 ab	193 ± 8 d	2.31 ± 0.23 ab	81.6 ± 8.6 a	184 ± 1 e	2.25 ± 0.23 a
DT+BI	49.8 ± 9.9 bc	259 ± 29 c	1.90 ± 0.27 bc	51.3 ± 2.4 bc	246 ± 20 d	1.45 ± 0.06 bc
DR+BI	49.8 ± 5.3 cde	278 ± 12 bc	1.42 ± 0.15 cde	41.9 ± 3.7 cd	305 ± 10 c	1.21 ± 0.10 cd
DTR+BI	36.4 ± 3.0 e	320 ± 11 ab	1.07 ± 0.08 e	29.0 ± 1.2 e	346 ± 11 b	0.88 ± 0.03 e

Note: CO refers to continuous flooding, DT to water scarcity during the tillering stage, DR to water scarcity during the reproductive stage, and DTR to water scarcity during both the tillering and reproductive stages. +BI indicates the addition of biochar to the same water regimes. Data are presented as mean ± standard error. Different letters indicate significant differences between treatments within each season.

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Table 3

Physical and chemical properties of soil after the wet and dry cultivation seasons.

Treatment	pH	EC (dS m^{-1})	OM (%)	OC (%)	Total C (%)	Total N (%)	NH_4^+ (mg kg^{-1})	NO_3^- (mg kg^{-1})	Avail. P (mg kg^{-1})	Exch. K (mg kg^{-1})	CEC (cmol kg^{-1})	Bulk density (g cm^{-3})	Moisture content (%)	SOC stock (Mg ha^{-1})
After wet season harvest														
CO	6.17 ± 0.03 a	0.78 ± 0.04 b	1.15 ± 0.03 b	0.67 ± 0.02 b	1.56 ± 0.05 b	0.11 ± 0.01 a	7.20 ± 1.77 b	36.9 ± 6.5 ab	26.3 ± 5.6 a	49.7 ± 9.2 a	10.7 ± 0.4 a	1.49 ± 0.05 a	19.9 ± 1.5 a	20.0 ± 1.2 bc
DT	6.21 ± 0.04 a	0.80 ± 0.02 b	1.14 ± 0.06 b	0.66 ± 0.03 b	1.59 ± 0.05 b	0.11 ± 0.01 a	7.65 ± 1.72 b	32.4 ± 3.1 b	27.8 ± 6.5 a	52.8 ± 11.3 a	10.5 ± 0.3 a	1.50 ± 0.04 a	19.4 ± 0.9 a	19.8 ± 0.7 bc
DR	6.19 ± 0.06 a	0.79 ± 0.03 b	1.14 ± 0.03 b	0.66 ± 0.01 b	1.57 ± 0.04 b	0.10 ± 0.01 a	6.48 ± 0.95 b	29.6 ± 4.0 b	29.8 ± 5.8 a	51.4 ± 9.4 a	10.8 ± 0.4 a	1.49 ± 0.05 a	19.8 ± 1.3 a	19.8 ± 0.6 bc
DTR	6.20 ± 0.04 a	0.79 ± 0.03 b	1.13 ± 0.02 b	0.66 ± 0.01 b	1.58 ± 0.03 b	0.10 ± 0.01 a	7.47 ± 0.79 b	29.7 ± 7.7 b	28.8 ± 8.2 a	51.9 ± 9.0 a	10.5 ± 0.4 a	1.49 ± 0.05 a	19.5 ± 0.9 a	19.6 ± 0.8 c
CO+BI	6.24 ± 0.03 a	0.86 ± 0.04 ab	1.28 ± 0.06 a	0.74 ± 0.03 a	1.78 ± 0.03 a	0.12 ± 0.01 a	12.6 ± 2.1 a	55.3 ± 11.5 a	37.4 ± 6.9 a	62.9 ± 9.3 a	11.6 ± 0.5 a	1.48 ± 0.04 a	20.3 ± 1.0 a	22.0 ± 0.5 ab
DT+BI	6.26 ± 0.02 a	0.85 ± 0.02 ab	1.33 ± 0.05 a	0.77 ± 0.03 a	1.76 ± 0.06 a	0.12 ± 0.01 a	12.5 ± 1.0 a	45.2 ± 7.5 ab	37.8 ± 7.5 a	59.7 ± 10.5 a	11.7 ± 0.6 a	1.48 ± 0.04 a	20.1 ± 1.1 a	22.8 ± 0.4 a
DR+BI	6.24 ± 0.04 a	0.86 ± 0.02 ab	1.31 ± 0.06 a	0.76 ± 0.04 a	1.73 ± 0.05 a	0.12 ± 0.01 a	11.7 ± 1.0 a	40.2 ± 6.9 ab	34.8 ± 8.1 a	59.0 ± 12.7 a	11.4 ± 0.8 a	1.48 ± 0.03 a	20.2 ± 0.6 a	22.5 ± 1.0 a
DTR+BI	6.24 ± 0.03 a	0.88 ± 0.03 a	1.28 ± 0.05 a	0.74 ± 0.03 a	1.72 ± 0.02 a	0.12 ± 0.01 a	9.66 ± 0.96 ab	35.7 ± 6.1 ab	36.3 ± 7.3 a	57.3 ± 11.0 a	11.3 ± 0.5 a	1.47 ± 0.03 a	20.2 ± 1.4 a	21.8 ± 0.9 abc
After dry season harvest														
CO	6.16 ± 0.04 b	0.76 ± 0.03 b	1.16 ± 0.04 b	0.67 ± 0.02 b	1.52 ± 0.05 b	0.11 ± 0.01 a	8.53 ± 0.95 b	39.1 ± 5.1 b	25.9 ± 5.1 b	51.5 ± 3.4 a	10.8 ± 0.6 bc	1.48 ± 0.04 a	14.8 ± 1.3 a	19.9 ± 0.6 b
DT	6.20 ± 0.02 b	0.75 ± 0.01 b	1.15 ± 0.04 b	0.67 ± 0.02 b	1.54 ± 0.04 b	0.11 ± 0.01 a	9.50 ± 0.97 ab	39.9 ± 5.6 b	26.3 ± 0.8 b	52.8 ± 8.4 a	10.5 ± 0.4 c	1.49 ± 0.03 a	15.2 ± 1.0 a	19.9 ± 0.9 b
DR	6.18 ± 0.03 b	0.75 ± 0.03 b	1.16 ± 0.05 b	0.67 ± 0.03 b	1.53 ± 0.05 b	0.11 ± 0.01 a	9.30 ± 2.43 ab	40.0 ± 6.7 b	26.7 ± 6.5 b	50.5 ± 4.9 a	10.9 ± 0.4 bc	1.49 ± 0.03 a	15.9 ± 1.3 a	20.0 ± 1.0 b
DTR	6.16 ± 0.02 b	0.75 ± 0.01 b	1.13 ± 0.05 b	0.66 ± 0.03 b	1.52 ± 0.06 b	0.11 ± 0.01 a	9.16 ± 1.45 b	38.4 ± 3.3 b	26.0 ± 3.5 b	51.9 ± 3.3 a	10.4 ± 0.4 c	1.49 ± 0.03 a	15.3 ± 0.8 a	19.6 ± 1.2 b
CO+BI	6.28 ± 0.02 a	0.87 ± 0.03 a	1.39 ± 0.06 a	0.81 ± 0.03 a	1.81 ± 0.05 a	0.12 ± 0.01 a	13.2 ± 3.0 ab	60.5 ± 11.2 a	36.0 ± 2.7 ab	62.8 ± 5.5 a	11.9 ± 0.5 ab	1.48 ± 0.02 a	16.1 ± 0.8 a	23.9 ± 1.2 a
DT+BI	6.30 ± 0.02 a	0.91 ± 0.03 a	1.40 ± 0.07 a	0.81 ± 0.04 a	1.81 ± 0.06 a	0.12 ± 0.01 a	14.2 ± 1.7 a	62.0 ± 7.1 a	41.7 ± 1.1 a	60.2 ± 5.3 a	12.1 ± 0.3 a	1.49 ± 0.03 a	16.3 ± 0.5 a	24.2 ± 1.6 a
DR+BI	6.27 ± 0.02 ab	0.86 ± 0.02 a	1.37 ± 0.05 a	0.79 ± 0.03 a	1.80 ± 0.07 a	0.12 ± 0.01 a	13.2 ± 1.3 ab	56.4 ± 5.1 ab	36.8 ± 6.1 ab	63.4 ± 1.8 a	11.9 ± 0.5 ab	1.49 ± 0.03 a	15.8 ± 0.6 a	23.7 ± 1.2 a
DTR+BI	6.26 ± 0.03 ab	0.87 ± 0.04 a	1.36 ± 0.02 a	0.79 ± 0.01 a	1.82 ± 0.09 a	0.12 ± 0.01 a	11.7 ± 0.8 ab	55.2 ± 4.8 ab	34.9 ± 5.7 ab	55.0 ± 9.5 a	12.0 ± 0.6 ab	1.47 ± 0.04 a	16.0 ± 0.7 a	23.3 ± 0.5 a

Note: CO refers to continuous flooding, DT to water scarcity during the tillering stage, DR to water scarcity during the reproductive stage, and DTR to water scarcity during both the tillering and reproductive stages. +BI indicates the addition of biochar to the same water regimes. Data are presented as mean ± standard error. Different letters indicate significant differences between treatments within each season. EC is electrical conductivity, OM is organic matter, OC is organic carbon, NH_4^+ is ammonium, NO_3^- in nitrate, CEC is cation exchange capacity, and SOC is soil organic carbon.

Table 4

Components of crop yields and grain yield-scaled greenhouse gas (GHG) emissions in rice cultivation during wet and dry seasons.

Treatment	Panicle number (panicle m^{-2})	1000-grain weight (g)	Grain number (grain $panicle^{-1}$)		Aboveground biomass weight (Mg ha^{-1})	Grain yield (Mg ha^{-1})	GHG intensity (Mg CO_2 eq Mg $yield^{-1}$)
			Filled grain	Unfilled grain			
Wet season							
CO	586 ± 5 bcd	27.1 ± 0.5 a	116 ± 8 a	7.00 ± 1.00 c	13.3 ± 0.5 ab	4.37 ± 0.14 a	0.62 ± 0.05 a
DT	587 ± 7 bcd	27.1 ± 0.3 a	113 ± 6 a	9.67 ± 1.53 c	12.2 ± 0.2 c	4.42 ± 0.10 a	0.49 ± 0.04 bc
DR	573 ± 6 cd	25.9 ± 0.4 b	80.0 ± 5.6 b	20.7 ± 4.2 a	12.4 ± 0.3 bc	3.04 ± 0.08 bc	0.53 ± 0.02 ab
DTR	569 ± 7 d	25.7 ± 0.5 b	73.3 ± 3.2 b	22.3 ± 3.8 a	12.0 ± 0.3 c	2.44 ± 0.12 d	0.49 ± 0.03 bc
CO+BI	604 ± 8 ab	27.1 ± 0.5 a	119 ± 5 a	12.7 ± 8.1 bc	13.6 ± 0.3 a	4.49 ± 0.15 a	0.51 ± 0.04 b
DT+BI	610 ± 8 a	27.2 ± 0.3 a	116 ± 3 a	8.67 ± 1.15 c	12.4 ± 0.2 bc	4.45 ± 0.16 a	0.43 ± 0.05 bc
DR+BI	588 ± 6 bc	26.0 ± 0.3 b	81.3 ± 5.0 b	19.0 ± 4.4 ab	12.5 ± 0.3 bc	3.22 ± 0.06 b	0.44 ± 0.04 bc
DTR+BI	578 ± 6 cd	25.7 ± 0.3 b	74.7 ± 4.5 b	19.7 ± 3.2 ab	12.2 ± 0.6 c	2.72 ± 0.10 cd	0.39 ± 0.02 c
Dry season							
CO	594 ± 8 bc	27.1 ± 0.3 a	112 ± 6 a	7.33 ± 1.53 d	13.1 ± 0.3 ab	4.38 ± 0.09 a	0.58 ± 0.03 a
DT	588 ± 7 bcd	27.1 ± 0.2 a	109 ± 6 a	7.00 ± 1.00 d	12.0 ± 0.3 c	4.30 ± 0.08 a	0.39 ± 0.02 cde
DR	577 ± 5 cd	25.5 ± 0.1 b	80.0 ± 5.6 b	17.0 ± 2.0 bc	12.2 ± 0.3 c	3.06 ± 0.08 b	0.46 ± 0.02 bc
DTR	570 ± 6 d	25.3 ± 0.2 b	75.3 ± 3.1 b	23.0 ± 2.7 a	11.6 ± 0.4 c	2.34 ± 0.09 d	0.42 ± 0.02 cd
CO+BI	616 ± 7 a	27.1 ± 0.3 a	113 ± 4 a	6.00 ± 1.00 d	13.4 ± 0.3 a	4.48 ± 0.05 a	0.50 ± 0.05 b
DT+BI	605 ± 7 ab	27.1 ± 0.4 a	110 ± 3 a	8.67 ± 1.15 d	12.2 ± 0.3 c	4.47 ± 0.13 a	0.32 ± 0.02 e
DR+BI	577 ± 9 cd	25.7 ± 0.3 b	83.3 ± 3.1 b	15.3 ± 3.2 c	12.4 ± 0.3 bc	3.24 ± 0.09 b	0.37 ± 0.02 de
DTR+BI	580 ± 10 cd	25.4 ± 0.2 b	77.7 ± 4.9 b	21.3 ± 2.1 ab	11.7 ± 0.3 c	2.78 ± 0.05 c	0.32 ± 0.02 e

Note: CO refers to continuous flooding, DT to water scarcity during the tillering stage, DR to water scarcity during the reproductive stage, and DTR to water scarcity during both the tillering and reproductive stages. +BI indicates the addition of biochar to the same water regimes. Data are presented as mean ± standard error. Different letters indicate significant differences between treatments within each season.

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Table 5

Water use, irrigation water productivity, and water scarcity footprint of rice cultivation in wet and dry seasons.

Treatment	Water use in rice cultivation		Irrigation water productivity (kg yield m^{-3})	Water scarcity footprint (m^3 H_2Oeq ha^{-1})
	Irrigation (m^3 ha^{-1})	Rain (m^3 ha^{-1})		
Wet season				
CO	6,080 ± 51 a	3,280	0.72 ± 0.02 d	4,270 ± 37 a
DT	4,070 ± 14 b	3,480	1.09 ± 0.05 b	2,930 ± 5 b
DR	3,900 ± 59 cd	3,280	0.78 ± 0.02 cd	2,520 ± 39 c
DTR	1,990 ± 22 e	3,480	1.23 ± 0.05 ab	1,260 ± 16 d
CO+BI	5,980 ± 108 a	3,280	0.75 ± 0.02 cd	4,190 ± 74 a
DT+BI	4,040 ± 55 bc	3,480	1.10 ± 0.05 b	2,910 ± 40 b
DR+BI	3,850 ± 59 d	3,280	0.84 ± 0.02 c	2,490 ± 39 c
DTR+BI	1,960 ± 31 e	3,480	1.39 ± 0.04 a	1,240 ± 23 d
Dry season				
CO	7,230 ± 24 a	1,080	0.61 ± 0.02 c	5,320 ± 21 a
DT	5,740 ± 37 b	1,080	0.75 ± 0.02 a	4,170 ± 27 b
DR	5,490 ± 23 c	1,080	0.56 ± 0.05 c	3,590 ± 18 c
DTR	3,920 ± 73 d	1,080	0.60 ± 0.02 c	2,360 ± 63 d
CO+BI	7,170 ± 30 a	1,080	0.62 ± 0.02 c	5,270 ± 27 a
DT+BI	5,750 ± 22 b	1,080	0.78 ± 0.02 a	4,170 ± 22 b
DR+BI	5,430 ± 17 c	1,080	0.60 ± 0.02 c	3,540 ± 6 c
DTR+BI	3,920 ± 17 d	1,080	0.71 ± 0.02 b	2,360 ± 9 d

Note: CO refers to continuous flooding, DT to water scarcity during the tillering stage, DR to water scarcity during the reproductive stage, and DTR to water scarcity during both the tillering and reproductive stages. +BI indicates the addition of biochar to the same water regimes. Data are presented as mean ± standard error. Different letters indicate significant differences between treatments within each season.

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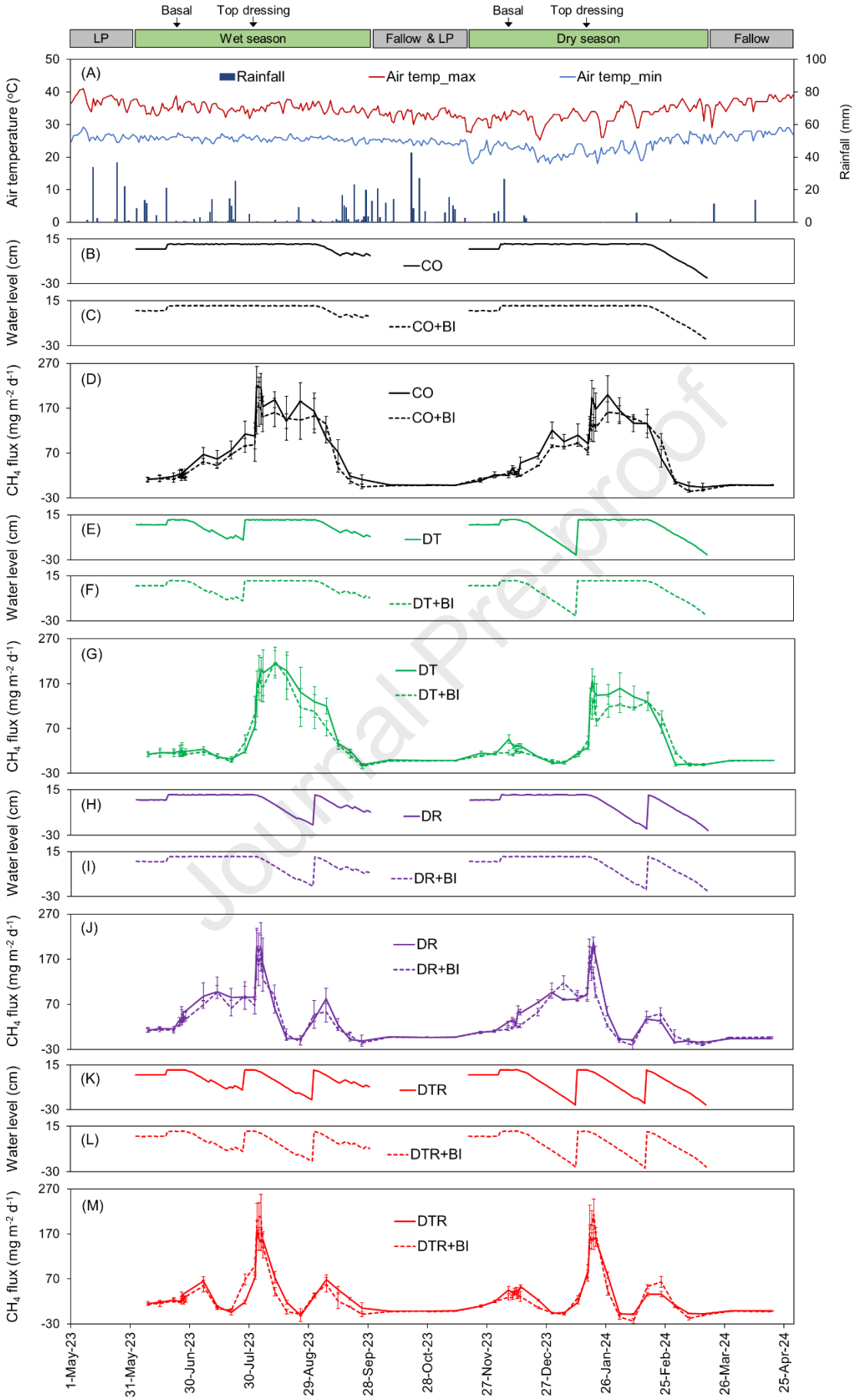

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Fig. 1. (A) Air temperature and rainfall during the study period. Water level in the field using (B) continuous flooding according to conventional practice (CO), (C) biochar application (CO+BI), (E) water scarcity during tillering stage (DT), (F) DT combined with BI (DT+BI), (H) water scarcity during reproductive stage (DR), (I) DR combined with BI (DR+BI), (K) water scarcity during tillering and reproductive stages (DTR), and (L) DTR combined with BI (DTR+BI). Variation of CH_4 emissions throughout the study period of (D) CO and CO+BI, (G) DT and DT+BI, (J) DR and DR+BI, and (M) DTR and DTR+BI. LP signifies land preparation, basal is application of basal fertilizer, and top dressing is application of top-dressing fertilizer.

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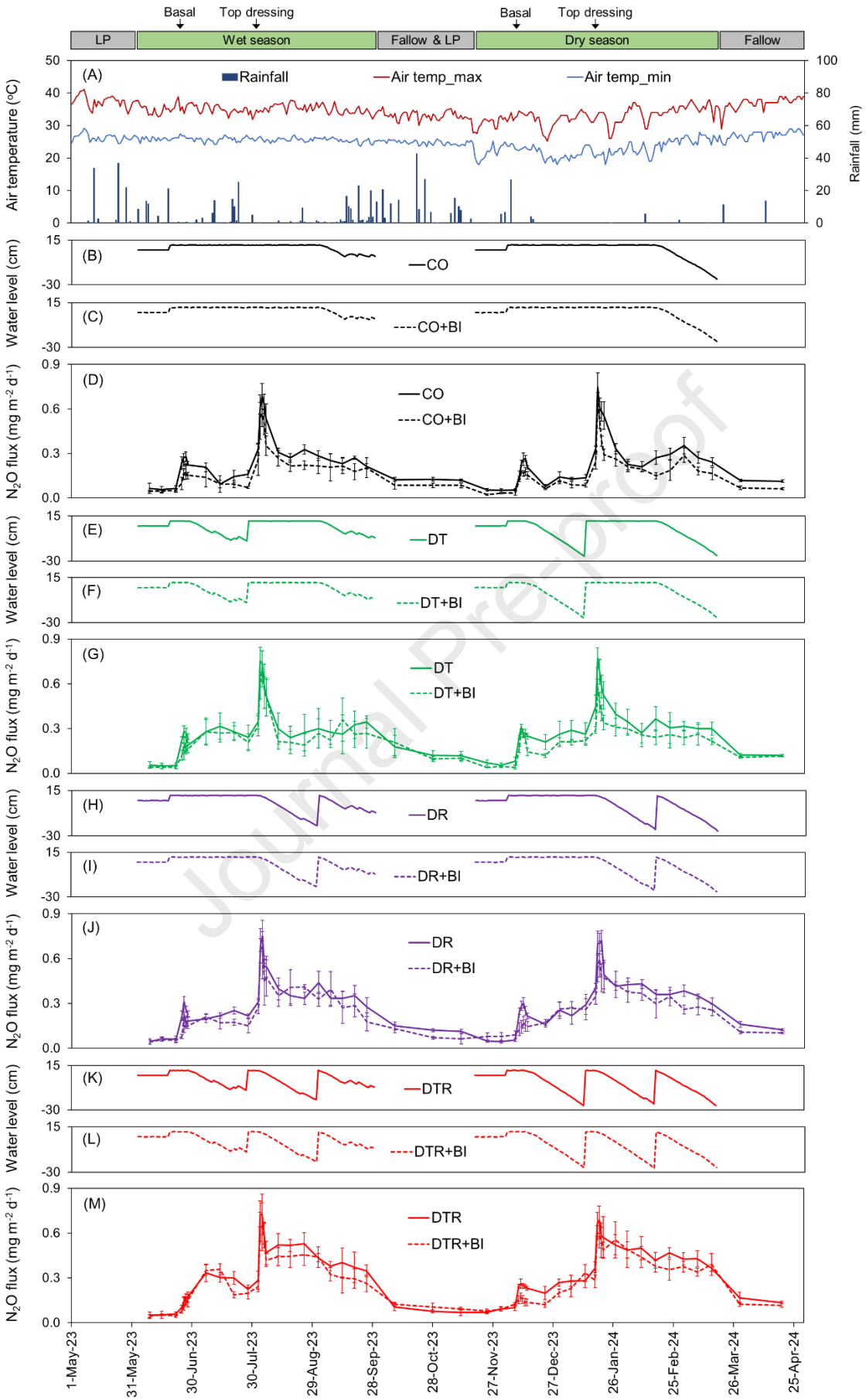

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Fig. 2. (A) Air temperature and rainfall during the study period. Water level in the field using (B) continuous flooding according to conventional practice (CO), (C) biochar application (CO+BI), (E) water scarcity during tillering stage (DT), (F) DT combined with BI (DT+BI), (H) water scarcity during reproductive stage (DR), (I) DR combined with BI (DR+BI), (K) water scarcity during tillering and reproductive stages (DTR), and (L) DTR combined with BI (DTR+BI). Variation of N_2O emissions throughout the study period of (D) CO and CO+BI, (G) DT and DT+BI, (J) DR and DR+BI, and (M) DTR and DTR+BI. LP signifies land preparation, basal is application of basal fertilizer, and top dressing is application of top-dressing fertilizer.

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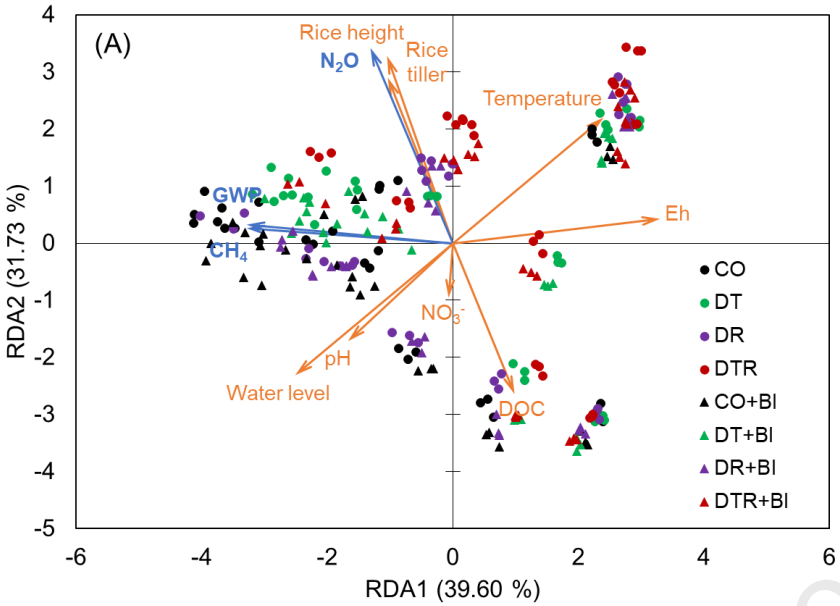

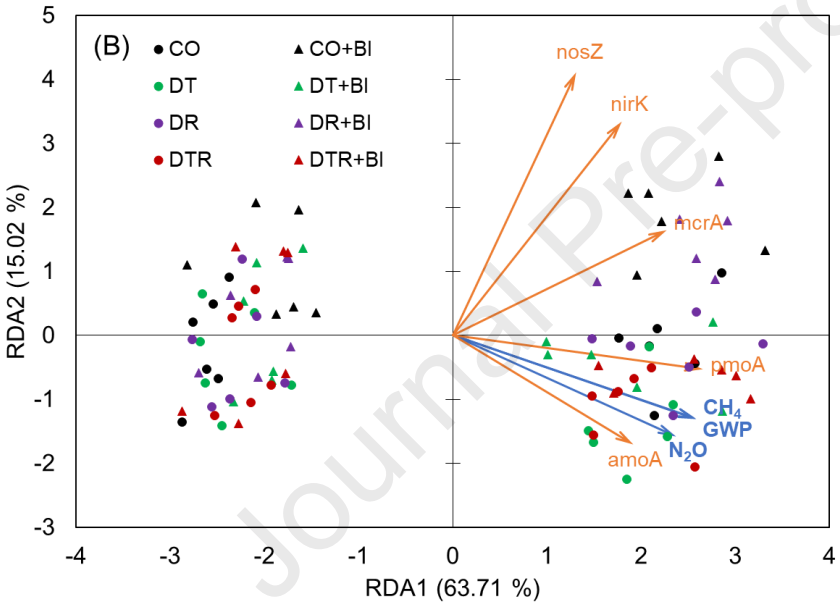

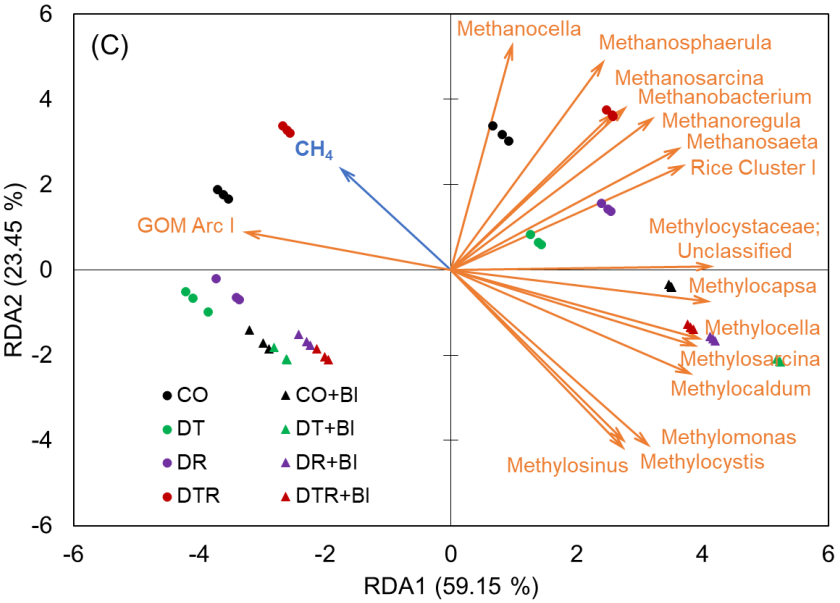

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Fig. 3. Redundancy analysis (RDA) illustrating the relationships between (A) greenhouse gas (GHG) emissions (CH_4, N_2O, and global warming potential (GWP)) and soil and plant conditions observed throughout the cultivation period; (B) GHG emissions (CH_4, N_2O, and GWP) and the abundances of soil microbial communities at 25 and 65 d after transplanting (DAT); and (C) CH_4 emissions and the abundances of methanogenic archaea and methanotrophic bacteria at 65 DAT. CO refers to continuous flooding, DT to water scarcity during the tillering stage, DR to water scarcity during the reproductive stage, and DTR to water scarcity during both the tillering and reproductive stages. +BI indicates the addition of biochar to the same water regimes. DOC is dissolved organic carbon, Eh is soil redox potential, and NO_3^- in nitrate. All associations are significant at $P < 0.05$.

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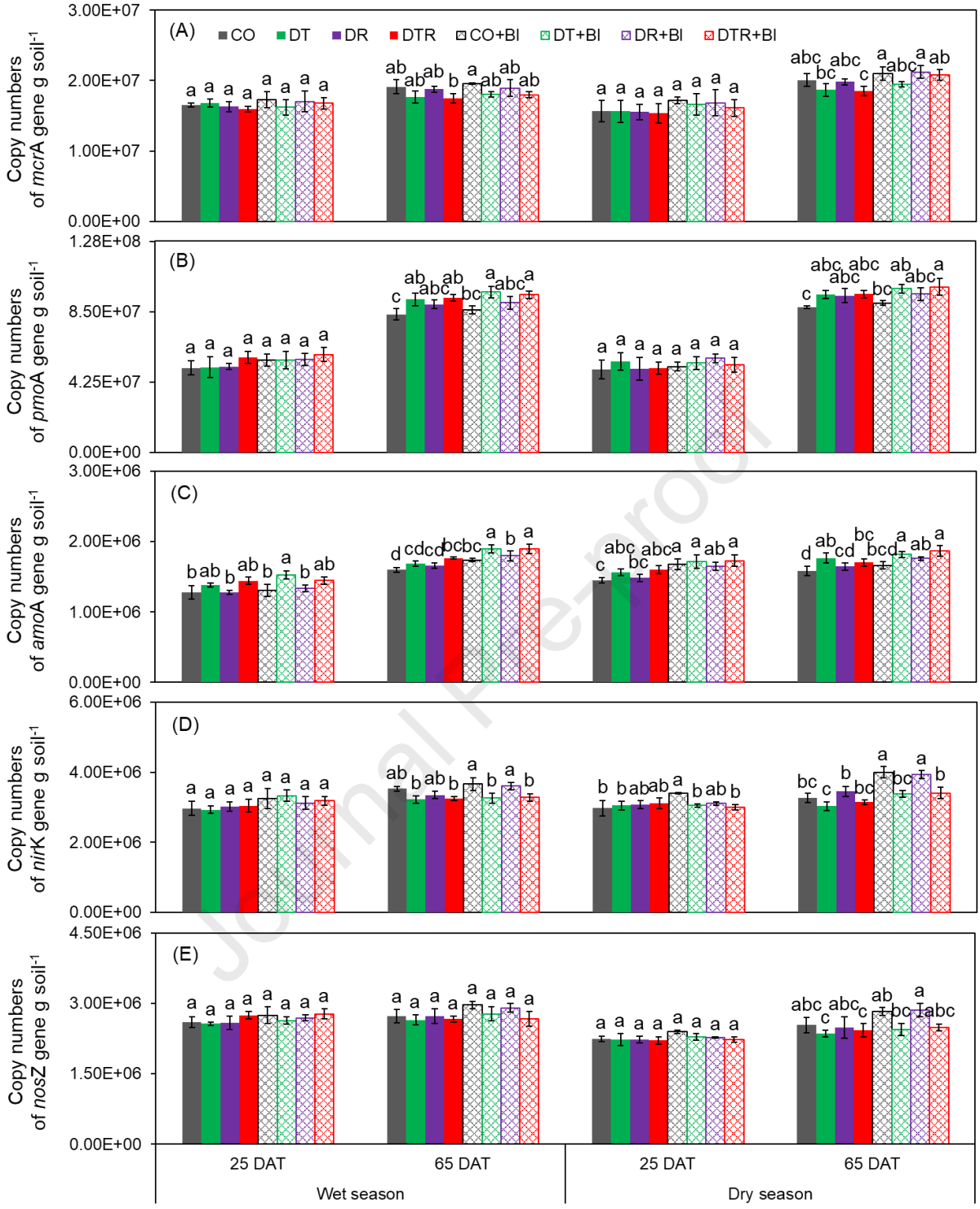

Fig. 4. Abundances of (A) methanogens (*mcr*A gene), (B) methanotrophs (*pmo*A gene), (C) nitrifying bacteria (*amo*A gene), (D) nitrite reductase (*nir*K), and (E) nitrous oxide reductase (*nos*Z) genes of denitrifying bacteria during the tillering (25 d after transplanting (DAT)) and reproductive (65 DAT) stages in both wet and dry seasons. CO refers to continuous flooding, DT to water scarcity during the tillering stage, DR to water scarcity during the reproductive

stage, and DTR to water scarcity during both the tillering and reproductive stages. +BI indicates the addition of biochar to the same water regimes. Data are presented as mean ± standard error. Different letters indicate significant differences between treatments within each season.

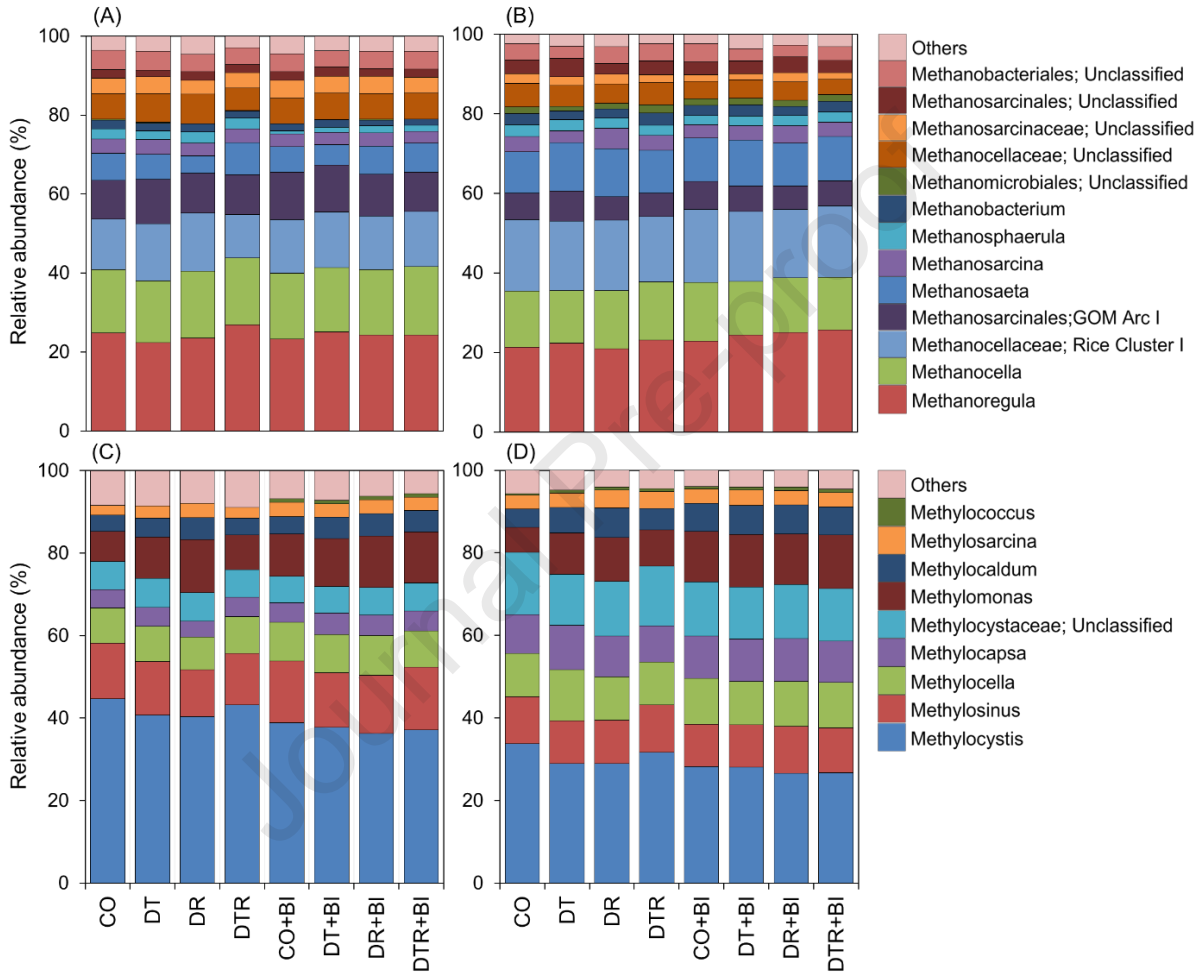

Fig. 5. Taxonomic profiles (genus level) of methanogenic archaea at 65 d after transplanting (DAT) in (A) the wet and (B) dry seasons, and methanotrophs at 65 DAT in (C) the wet and (D) dry seasons. CO refers to continuous flooding, DT to water scarcity during the tillering stage, DR to water scarcity during the reproductive stage, and DTR to water scarcity during both the tillering and reproductive stages. +BI indicates the addition of biochar to the same water regimes.

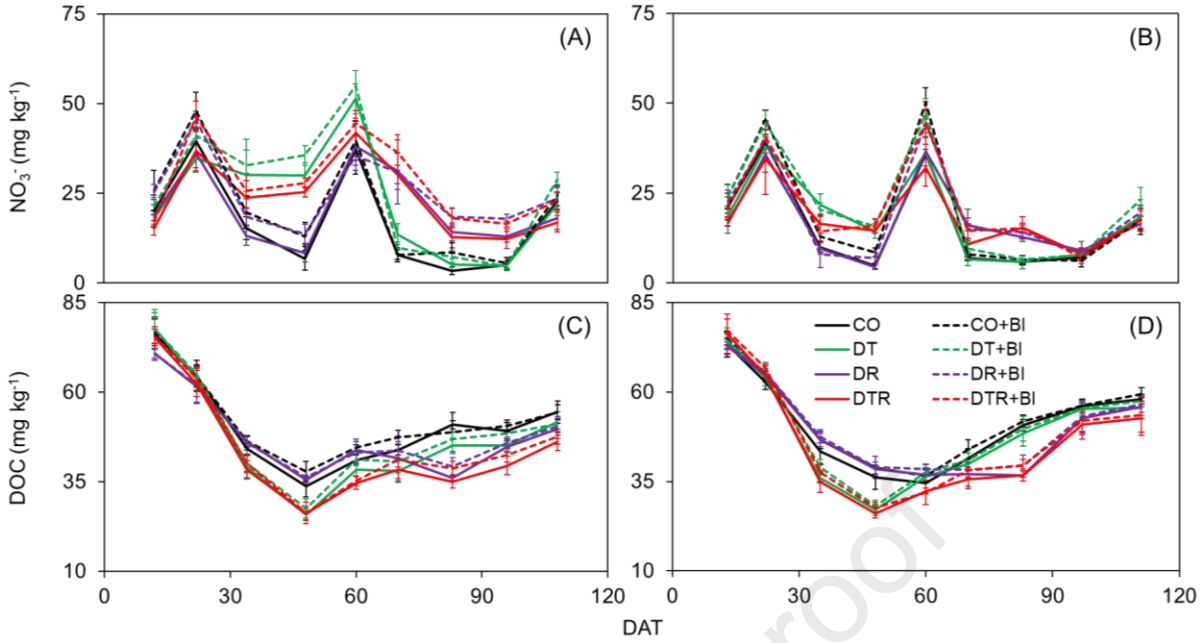

Fig. 6. Soluble (A, B) nitrate (NO_3^-) and (C, D) dissolved organic carbon (DOC) in soil during the (A, C) wet and (B, D) dry cultivation seasons. CO refers to continuous flooding, DT to water scarcity during the tillering stage, DR to water scarcity during the reproductive stage, and DTR to water scarcity during both the tillering and reproductive stages. +BI indicates the addition of biochar to the same water regimes. Data are presented as mean ± standard error.

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