



Review

Global nitrogen input on wetland ecosystem: The driving mechanism of soil labile carbon and nitrogen on greenhouse gas emissions

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ABSTRACT

Greenhouse gas emissions from wetlands are significantly promoted by global nitrogen input for changing the rate of soil carbon and nitrogen cycling, and are substantially affected by soil labile carbon and nitrogen conversely. However, the driving mechanism by which soil labile carbon and nitrogen affect greenhouse gas emissions from wetland ecosystems under global nitrogen input is not well understood. Working out the driving factor of nitrogen input on greenhouse gas emissions from wetlands is critical to reducing global warming from nitrogen input. Thus, we synthesized 72 published studies (2144 paired observations) of greenhouse gas fluxes and soil labile compounds of carbon and nitrogen (ammonium, nitrate, dissolved organic carbon, soil microbial biomass nitrogen and carbon), to understand the effects of labile carbon and nitrogen on greenhouse gas emissions under global nitrogen input. Across the data set, nitrogen input significantly promoted carbon dioxide, methane and nitrous oxide emissions from wetlands. In particular, at lower nitrogen rates ($<100 \text{ kg ha}^{-1} \cdot \text{yr}^{-1}$) and with added ammonium compounds, freshwater wetland significantly promoted carbon dioxide and methane emissions. Peatland was the largest nitrous oxide source under these conditions. This meta-analysis also revealed that nitrogen input stimulated dissolved organic carbon, ammonium, nitrate, microbial biomass carbon and microbial biomass nitrogen accumulation in the wetland ecosystem. The variation-partitioning analysis and structural equation model were used to analyze the relationship between the greenhouse gas and labile carbon and nitrogen further. These results revealed that dissolved organic carbon (DOC) is the primary factor driving greenhouse gas emission from wetlands under global nitrogen input, whereas microbial biomass carbon (MBC) more directly affects greenhouse gas emission than other labile carbon and nitrogen.

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1. Introduction

In recent decades, nitrogen input into ecosystems has

substantially increased at the global scale due to atmospheric deposition, agricultural input, fossil fuel combustion, and other anthropogenic activities [1–3]. Not only can nitrogen be a limiting nutrient [4], but it can also be a pollutant in many terrestrial ecosystems [5–7]. The wetland ecosystem is the key ecotone between terrestrial and aquatic ecosystems, and nitrogen can move from wetland ecosystems to rivers or lakes, leading to water eutrophication [5,8,9]. The nitrogen trapped by wetlands could also impact element cycling by changing the soil physicochemical properties [10,11] and microbial communities [12,13]. The soil physicochemical properties (e.g. temperature, moisture content, electrical conductivity and so on) and microbial communities usually determine

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Acronyms and symbols			
DOC	dissolved organic carbon	N	the number of observations
SMD	the standardized mean difference	v	variance
MBC	microbial biomass carbon	SEM	The structural equation model
CI	the confidence intervals	X_C	the mean values of an index in the control treatment
CNKI	China National Knowledge Infrastructure	X^2	Chi-Square value
kg	kilogram	X_E	the mean values of an index in the experimental treatment
N_2O	nitrous oxide	df	degree of freedom
ha	Hectares	N_C	the sample size of an index in the control treatment
CH_4	methane	GFI	high goodness-of-fit index
yr	year	N_E	the sample size of an index in the experimental treatment
CO_2	carbon dioxide	CFI	the comparative fit index
NH_4^+/NH_4^+-N	ammonium	S_C	the standard deviation of an index in the control treatment
cm	centimeter	RMSEA	the low root means square errors of approximation
$NO_3^-/NO_3^- - N$	nitrate	S_E	the standard deviation of an index in the experimental treatment
C	the control treatment	MBN	microbial biomass carbon
NH_4NO_3	ammonium nitrate	w_i	weight factor
E	the experimental treatment		
Q_M	the between-group heterogeneity		
d	the effect size		

the availability of soil carbon and nitrogen [14], thereby determining the biomass of vegetation communities [15]. Therefore, soil carbon and nitrogen availability play key roles in the substance cycling of wetland ecosystems. Nitrogen input typically alters the soil nitrogen and carbon availability by affecting labile carbon and nitrogen, including ammonium, nitrate [16,17], dissolved organic carbon [2], soil microbial biomass carbon and nitrogen [18]. However, how the availability of soil carbon and nitrogen in wetlands responds to nitrogen input is often controversial [19–21]. For example, Song et al. [21] pointed that DOC content reduced and ammonium augmented with the increase of nitrogen input rate, but Cui et al. [19] revealed an opposite trend in the peatlands of Northeast China. Song et al. [22] found that nitrogen addition increased nitrate content and MBC, which results contrast with the study of Kastovska et al. [20] and Song et al. [2]. Therefore, exploring the influence of nitrogen enrichment on the availability of soil labile carbon and nitrogen is critically important to understanding substance cycling in wetland ecosystems on a global scale.

Although previous studies have verified that terrestrial ecosystems act a sink/source of greenhouse gases and have quantitatively analyzed the effect of nitrogen input on greenhouse gas emissions [23–25], these results might not accurately describe the effect of wetlands on greenhouse gas emissions because the wetland ecosystem is very different from other types of terrestrial ecosystems [9]. The wetland ecosystem is located at the junction of the terrestrial-aquatic interlaced zone, and has some special characteristics, including frequent changes in water level, great redox fluctuation from highly anaerobic to highly aerobic conditions, and interception of partial nitrogen runoff [26–28]. These characteristics lead to the distinct greenhouse gas emission regulars from wetlands compared to other types of terrestrial ecosystems. Although wetlands occupy only 6%–8% of the earth's land surface, they are an important sink/source of greenhouse gas [29,30]. For example, the IPCC [31] reported that methane emissions from wetlands account for an estimated 63% of all natural methane emissions. Thus, understanding how nitrogen input affects greenhouse gas emissions from wetlands is critically important when attempting to understand the future global climate.

Nitrogen input could not only influence greenhouse gas emissions from wetland ecosystems by altering the soil nitrogen and

carbon cycling [23,32], but could also affect soil microbes due to its influences on soil nitrogen and carbon availability, thereby affecting greenhouse gas emissions [33–35]. Thus, revealing the interactions between soil labile carbon and nitrogen and greenhouse gases will contribute to understanding the mechanism of greenhouse gas emissions from wetland ecosystems to the atmosphere, resulting in elucidating the contribution of wetland ecosystems to the global greenhouse effect. However, previous studies found that soil labile carbon and nitrogen showed both positive and negative effects on greenhouse gas emissions from wetland ecosystems under global nitrogen input [36–39]. The wetland types and climates also affected the relationships between soil labile carbon and nitrogen and greenhouse gas under nitrogen input. Therefore, the mechanism of soil labile carbon and nitrogen on greenhouse gas emissions from wetland ecosystems is complicated and currently not well understood under nitrogen input at global scales. Thus, there is a desperate need to clarify the driving mechanism of labile carbon and nitrogen on greenhouse gas emission under global nitrogen input by combining the conclusions from various studies using a meta-analysis.

To untangle these controversial and uncertain issues, we used a meta-analysis to analyze studies on nitrogen input experiments published prior to September 2019. We used soil carbon dioxide emissions, methane emissions, nitrous oxide emissions, soil labile carbon and nitrogen, and soil microbial biomass to address the following questions: (i) How do the soil greenhouse gas emissions from wetland ecosystems fluctuate as a result of varying nitrogen input in terms of rates, compounds and environmental factors? (ii) What key factors affect soil greenhouse gas emissions as a response to nitrogen input? (iii) What is the major effect of labile compounds on greenhouse gas emissions from wetland ecosystems under global nitrogen input?

2. Materials and method

2.1. Meta data collection

The IPCC [40] indicated that the increase of nitrogen deposition could promote greenhouse gas emissions into the atmosphere, and Liu et al. [41] and Deng et al. [23] utilized meta-analyses to reveal

the effects of nitrogen input on soil greenhouse gas from terrestrial systems. However, these studies had some limitations and constraints. First, these studies covered a variety of terrestrial ecosystems. However, the wetland ecosystem is the key ecotone between terrestrial and aquatic ecosystems, and therefore its response to nitrogen input is greatly different from that of other types of terrestrial ecosystems. A new meta-analytical study needs to consider the particular pattern of greenhouse gas emissions in wetland ecosystems. Second, Liu et al. [41] and Tian et al. [64] utilized global models to determine the emission patterns and effects of different factor prior to 2009. Deng et al. [23] focused on the relationships between greenhouse gas emissions and carbon pools. Thus, we conducted a literature search in September 2019 for all papers published over the past decade on greenhouse gases, nitrogen input and wetland ecosystems. This literature search used the Web of Science, ScienceDirect, Google Scholar, and CNKI. The keywords for the online search were: (wetland OR peatland OR marsh OR bog OR fen) AND (nitrogen input OR nitrogen addition OR nitrogen enrichment OR nitrogen deposition OR nitrogen fertilizer) AND (greenhouse gas OR nitrous oxide OR N₂O OR methane OR CH₄ OR carbon dioxide OR CO₂). The selected studies satisfied the following criteria: (a) the control experiment was defined by no nitrogen input or atmospheric nitrogen deposition; (b) non-repetitive experimental studies were excluded; (c) non-experimental studies (such as modeling, meta-analyses, and reviews) were excluded.

Based on these criteria, approximately 2144 paired observations (Fig. 1) from 72 papers published from 2009 to 2019 on greenhouse gas emissions (including CO₂, CH₄ and N₂O) and labile carbon or nitrogen (mainly including dissolved organic carbon, ammonium, nitrate, microbial biomass carbon and nitrogen) under global nitrogen input were selected for data collection. Data sources included tables, text, figures and supplementary files. The data in figures were collected using the GetData 2.25 software (<http://getdata-graph-digitizer.com/>). If the key data was not directly acquired, we obtained the data from the authors. The rate of nitrogen input, types of nitrogen input, climate, nitrogen compounds, and types of wetlands were collected. The labile compounds were also collected, including ammonium, nitrate, dissolved organic carbon, soil microbial biomass carbon, and soil microbial biomass nitrogen

from the surface soil (the depth ranges from 0 to 20 cm). We summarized the latitude and longitude of each site from the published papers, or we extracted these data online (<http://www.worldclim.org/>; Table. A1).

2.2. Meta data analysis

The effect size was calculated using Hedges' *d*, which is a measurement of the unbiased standardized mean difference between the control (C) and experimental (E) means [42,43]. The equations for the effect size (*d*) and variance (*v*) are listed as in the follows:

$$S = \sqrt{\frac{(N_E - 1)(S_E)^2 + (N_C - 1)(S_C)^2}{N_E + N_C - 2}} \quad (1)$$

$$d = \frac{(X_E - X_C)}{S} \times \left(1 - \frac{3}{4(N_C + N_E - 2) - 1}\right) \quad (2)$$

$$v = \frac{N_C + N_E}{N_C N_E} + \frac{d^2}{2(N_C + N_E)} \quad (3)$$

X_C and X_E represent the mean values of an index in the control and experimental treatment, respectively. N_C and N_E represent the sample size of an index in the control and experimental treatment, respectively. S_C and S_E represent the standard deviation of an index in control and the experimental treatment, respectively.

The weight factor (*w_i*) was determined as follow:

$$w_i = 1/v \quad (4)$$

The *d* of the control and nitrogen input treatments were used to calculate the weighted standardized mean difference (SMD):

$$SMD = \frac{\sum_{i=1}^n w_i d_i}{\sum_{i=1}^n w_i} \quad (5)$$

where *n* refers to the number of observations, *w_i* and *d_i* represent the weight factor and effect size of observation *i*, respectively.

The calculated mean effect size considered the confidence

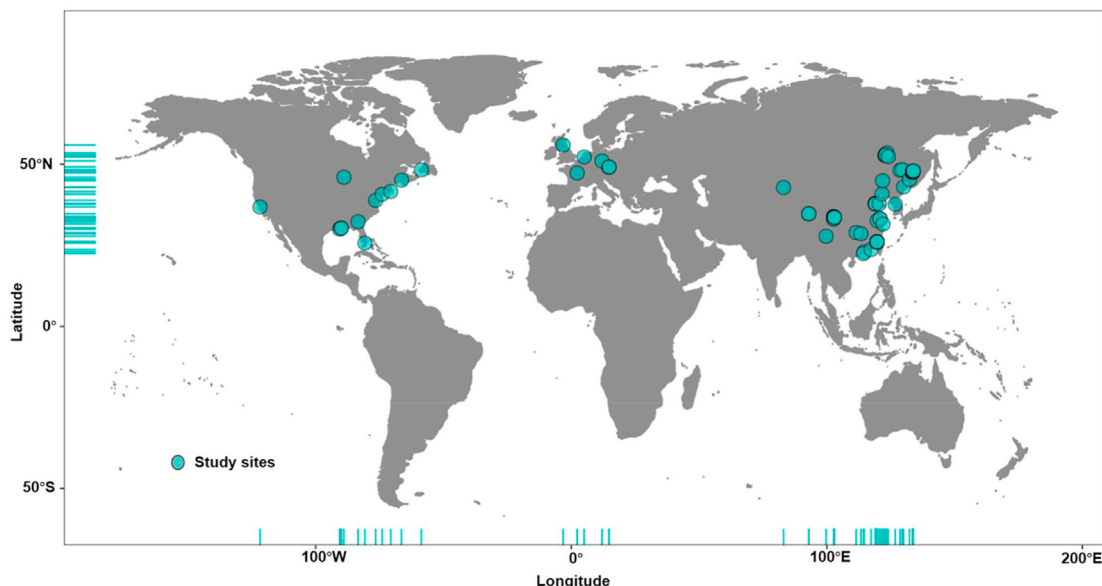


Fig. 1. Geographical distribution of the study sites.

intervals (Bootstrap CIs, bootstrapping by 4999 iterations). If the Bootstrap CIs had nonzero overlap, the nitrogen input significantly influenced the greenhouse gas emissions [44,45]. Negative Hedges'd values indicated that the nitrogen input decreased the greenhouse gas emissions. Positive Hedges'd values indicated that nitrogen input increased the greenhouse gas emissions [46].

To test the effects of nitrogen input on greenhouse gas emissions, we categorized the nitrogen input treatments into four groups: nitrogen input rates (0–50, 50–100, 100–200, 200–300, >300 kg ha⁻¹·yr⁻¹), climate (alpine climate, temperate continental climate, temperate marine climate, monsoon climate of medium latitudes, subtropical monsoon climate, subtropical humid climate), nitrogen compounds (NH₄⁺, NO₃⁻, NH₄NO₃, organic nitrogen fertilizer) and type of wetland (freshwater marsh, alpine wetland, estuary wetland, peatland, salt marsh). The data were analyzed using a mixed-effects model [47]. There are random variations in effect sizes among all the observations, whereas each individual observation is weighted by the reciprocal of the mixed-model variance [48,49]. If the between-group heterogeneity (Q_M) test was smaller than 0.05, it indicates that significant differences exist among the different groups. We tested the Q_M of CO₂, CH₄ and N₂O and the results are shown in Table 1. Meanwhile, we also tested the Q_M of ammonium, nitrate, soil microbial biomass nitrogen, dissolved organic carbon and soil microbial biomass carbon (Table. B1).

2.3. Publication bias

Publication bias means there is a higher possibility of publishing highly positive or negative results or not reporting non-significant effects [43]. We tested the publication bias for greenhouse gas emissions using weighted histograms and a fail-safe number. Weighted histograms consist of the effect sizes and weight of data (Eq. (4)), rather than the frequency of effect size [50]. The fail-safe number is substantially larger than 5 N + 10 (N represent the number of observations in this study), where 5 N + 10 was defined using the acceptable threshold in the literature. The results indicate that the observations from this study can be treated as a reliable estimate of the true effect [43,51]. Therefore, the results shown in Fig. B1 indicate that there were no biases in the selected publications.

2.4. Statistic analysis

All of the standardized mean differences, the between-group heterogeneity, Bootstrap CIs and fail-safe number were counted using MetaWin 2.1.3 software (<http://www.metawinsoft.com/>, Sinauer Associates Inc., Sunderland, MA, USA). The figures were

Table 1
Results of statistical comparisons among groups for greenhouse gas.

Item	CO ₂		CH ₄		N ₂ O	
	Q _M	p-value	Q _M	p-value	Q _M	p-value
Rate of nitrogen input	24.03	<0.001	13.94	<0.001	28.21	<0.001
Climate	37.52	<0.001	28.52	<0.001	31.18	<0.001
Nitrogen compounds	9.81	<0.05	30.29	<0.001	38.09	<0.001
Types of wetlands	29.00	<0.001	24.35	<0.001	43.01	<0.001

Notes: All data were grouped into five nitrogen input rates (0–50, 50–100, 100–200, 200–300, >300 kg ha⁻¹·yr⁻¹), six climate types (alpine climate, temperate continental climate, temperate marine climate, monsoon climate of medium latitudes, subtropical monsoon climate, subtropical humid climate), four nitrogen compounds types (NH₄⁺, NO₃⁻, NH₄NO₃, organic nitrogen fertilizer) and five wetlands types (freshwater marsh, alpine wetland, estuary wetland, peatland, salt marsh). CO₂ is carbon dioxide, CH₄ is methane, and N₂O is nitrous oxide. Q_M: heterogeneity in group cumulative effect sizes.

constructed using OriginPro 2017 and R (3.6.1) software. The variation-partitioning analysis was conducted using R (3.6.1) software for the effects of soil labile carbon and nitrogen on greenhouse gas emissions. The Pearson correlation analysis was performed using SPSS 20.0 (IBM Corporation, Armonk, NY, USA) for indicating the relationships between greenhouse gas and labile carbon and nitrogen, where *p* values smaller than 0.05 are considered statistically significant. The regression analysis was conducted using the OriginPro 2017 software for the effect size of greenhouse gas emission and mean annual precipitation, mean annual temperature at the global level.

The structural equation model (SEM) could reveal the driving factors and impacts of greenhouse gas emissions under nitrogen input and be constructed using Amos (Version 21). Several tests were used to determine the adequacy of model fitting, including the X² test (0.05 < *p* < 1.00, 0 ≤ X²/df ≤ 2), high goodness-of-fit index (GFI, 0.9 < GFI < 1.0), the comparative fit index (CFI, 0.9 < CFI < 1.0), and the low root means square errors of approximation (RMSEA, < 0.05). The effect value and pathway of the model were obtained after the model was constructed. The obtained test results, including X²/df < 2, *p* > 0.05, GFI and CFI close to 1, and RMSEA < 0.05, for the SEM (Fig. 7) indicated that the SEM could be considered to be a perfect fit.

3. Results

3.1. Greenhouse gas emissions from wetland ecosystems under nitrogen input

Across all observations, the overall standardized mean difference (SMD) of carbon dioxide (CO₂), methane (CH₄) and nitrous oxide (N₂O) were 1.41, 0.58 and 1.74, respectively (Fig. 2; Bootstrap CIs of 1.24–1.57, 0.31 to 0.85, and 1.52 to 1.97, respectively), and presented a significantly positive effect because the Bootstrap CIs had nonzero overlap. Nitrogen input increased greenhouse gas emissions for all types of wetlands, except for the Alpine wetland, which had significantly decreased methane emission (SMD = -1.41, Bootstrap CIs = -2.14 to -0.74). Compared to CO₂ and CH₄, all types of climate significantly and positively promoted N₂O emissions. Specially, the SMD of CO₂ and CH₄ were negative under temperate continental climate. This suggested that nitrogen input under temperate continental climate decreased CO₂ and CH₄ emissions in comparison to that no nitrogen addition in the wetland.

Nitrogen input via NH₄⁺ and NH₄NO₃ significantly promoted greenhouse gas emissions (Fig. 2). However, organic nitrogen fertilizer significantly reduced CH₄ emissions (SMD = -0.76, Bootstrap CIs = -1.36 to -0.30). We also found that different nitrogen input rates had a positive effect on the CO₂ and N₂O emissions in wetlands ecosystem (SMD = 0.069 to 2.32 in Bootstrap CIs). Specially, the nitrogen input rate of 50–100 kg ha⁻¹·yr⁻¹ had the largest impact on CO₂ and N₂O emissions among all nitrogen input rates. Meanwhile, nitrogen input rate of 0–50 kg ha⁻¹·yr⁻¹ (SMD = 1.16 in Bootstrap CIs) had the largest effect on CH₄ emissions among all nitrogen input rates. This suggested that lower nitrogen input rates (<100 kg ha⁻¹·yr⁻¹) significantly promoted greenhouse gas emissions.

3.2. Changes in soil labile carbon and nitrogen under nitrogen input in wetland ecosystems

As illustrated in Fig. 3, the overall SMD of dissolved organic carbon (DOC), ammonium (NH₄⁺-N), and nitrate (NO₃⁻-N) range from 0.64 to 3.58 indicated that the nitrogen input augmented soil labile carbon and nitrogen contents in wetland ecosystems. For all

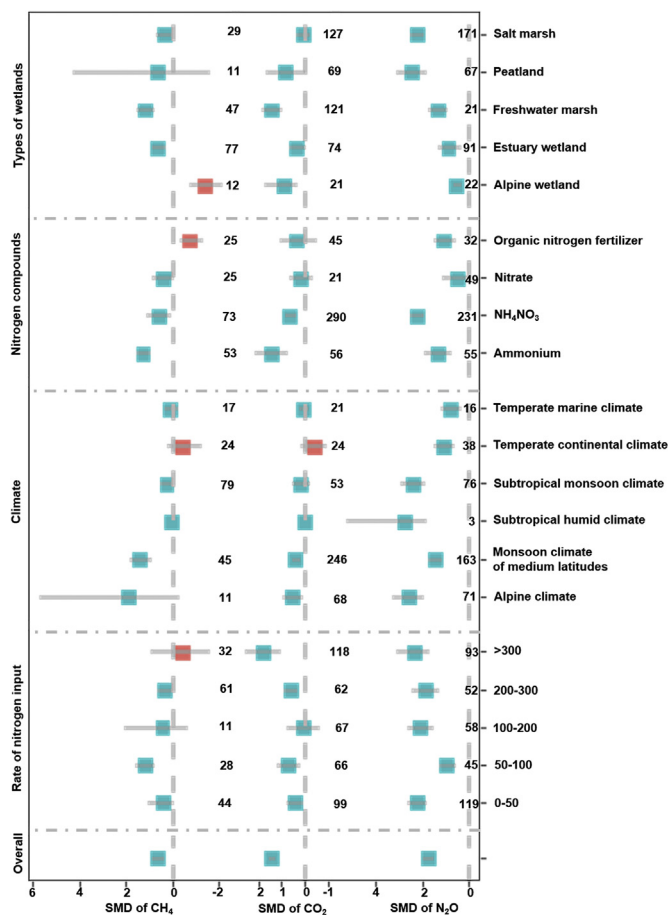


Fig. 2. Standardized mean difference for greenhouse gases emissions from different wetland environments under nitrogen inputs. The numbers in the figure represent the number of case studies. A standardized mean difference >0 reveals a positive effect on greenhouse gas emissions, whereas values < 0 reveal negative effects. Error bars are the bootstrap confidence intervals (CIs). CIs that do not include 0 and do not overlap indicate a significant effect on greenhouse gas emissions and significant differences among groups, respectively. SMD represents the standardized mean difference, which is a type of effect size. The unit of nitrogen input rate is $\text{kg} \cdot \text{ha}^{-1} \cdot \text{yr}^{-1}$.

wetland types, nitrogen input dwindled the soil $\text{NH}_4^+ \text{-N}$ and $\text{NO}_3^- \text{-N}$ contents. For peatland, the soil DOC was significantly increased by nitrogen input, whereas the soil DOC was reduced by nitrogen input for freshwater marshes, alpine wetlands and salt marshes. Fig. 3 shows that for all types of climates except the temperate marine climate, the nitrogen input significantly augmented the soil $\text{NO}_3^- \text{-N}$ contents. The nitrogen input significantly increased the soil $\text{NH}_4^+ \text{-N}$ content for the alpine climate, temperate continental climate, monsoon climate of medium latitudes and subtropical monsoon climate. The soil DOC was significantly added by nitrogen input for the alpine climate (SMD = 1.72, Bootstrap CIs = 1.36 to 2.11), whereas the nitrogen input under the temperate marine climate diminished the soil DOC content.

For all types of nitrogen compounds, nitrogen input added the soil $\text{NH}_4^+ \text{-N}$ and $\text{NO}_3^- \text{-N}$ (Fig. 3). The effect of nitrogen input in terms of NH_4^+ , NH_4NO_3 and NO_3^- on the soil $\text{NH}_4^+ \text{-N}$ and $\text{NO}_3^- \text{-N}$ content was larger than that of organic nitrogen fertilizer. This means that inorganic nitrogen input significantly and directly promotes soil $\text{NH}_4^+ \text{-N}$ and $\text{NO}_3^- \text{-N}$ formation. Similarly, the soil DOC content was increased by adding inorganic nitrogen, and significantly lessened by adding organic nitrogen fertilizer. We also revealed that all nitrogen input rates promoted $\text{NH}_4^+ \text{-N}$ and $\text{NO}_3^- \text{-N}$ formation. However, nitrogen input rate of 0–50 and

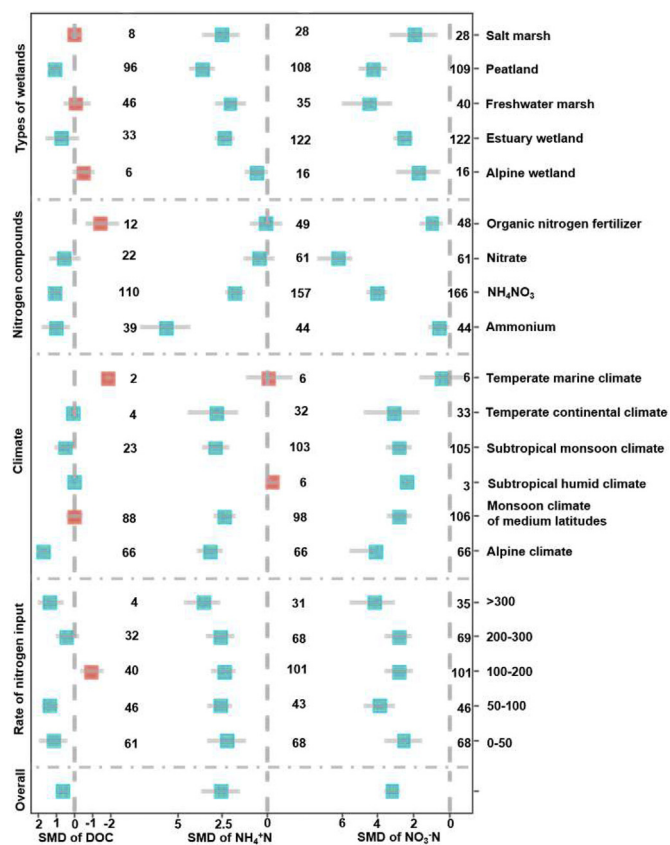


Fig. 3. Standardized mean difference for DOC, $\text{NH}_4^+ \text{-N}$, $\text{NO}_3^- \text{-N}$ from different wetland environments under nitrogen inputs. The numbers in the figure represent the number of case studies. For details on effect size interpretation, refer to Fig. 2.

$50\text{--}100 \text{ kg ha}^{-1} \cdot \text{yr}^{-1}$ had larger impacts on the soil $\text{NH}_4^+ \text{-N}$ and $\text{NO}_3^- \text{-N}$ formation than other input rates. It signifies that lower nitrogen input rates significantly expanded the soil nitrogen availability. Similarly, lower nitrogen input rates showed significant and positive effects on the soil DOC contents. In contrast, a nitrogen input rate of $100\text{--}200 \text{ kg ha}^{-1} \cdot \text{yr}^{-1}$ (SMD = -0.96 , Bootstrap CIs = -1.66 to -0.35) significantly decreased the soil DOC content.

3.3. Changes in soil microbial biomass under nitrogen input in wetland ecosystems

Fig. 4 shows that the overall SMD of soil microbial biomass carbon (MBC) and nitrogen (MBN) were 1.24 and 3.01 (Bootstrap CIs of 0.87–1.61 and 2.47 to 3.56, respectively), and presented positive effects of nitrogen input on MBC and MBN significantly. The results showed that for estuary wetlands and peatlands, nitrogen input significantly added the MBC (means of SMD = 3.54 to 4.33 in Bootstrap CIs) and the MBN (means of SMD = 1.91 to 3.65 in Bootstrap CIs). For freshwater marsh and alpine wetland, nitrogen input reduced the MBC and MBN. Compared to other climates, the alpine climate had the largest SMD of MBC (SMD = 2.10, Bootstrap CIs = 1.51 to 2.73) and MBN (SMD = 3.44, Bootstrap CIs = 2.80 to 4.14 in). It suggests that for the alpine climate, nitrogen input significantly multiplied the activity of soil microbes. Similarly, the effect of nitrogen deposition on soil microbial biomass was greater than that of fertilization.

As shown in Fig. 4, inorganic nitrogen input significantly increased the MBC (SMD range from 1.79 to 4.27 in Bootstrap CIs) and MBN (means of SMD range from 1.00 to 1.88 in Bootstrap CIs).

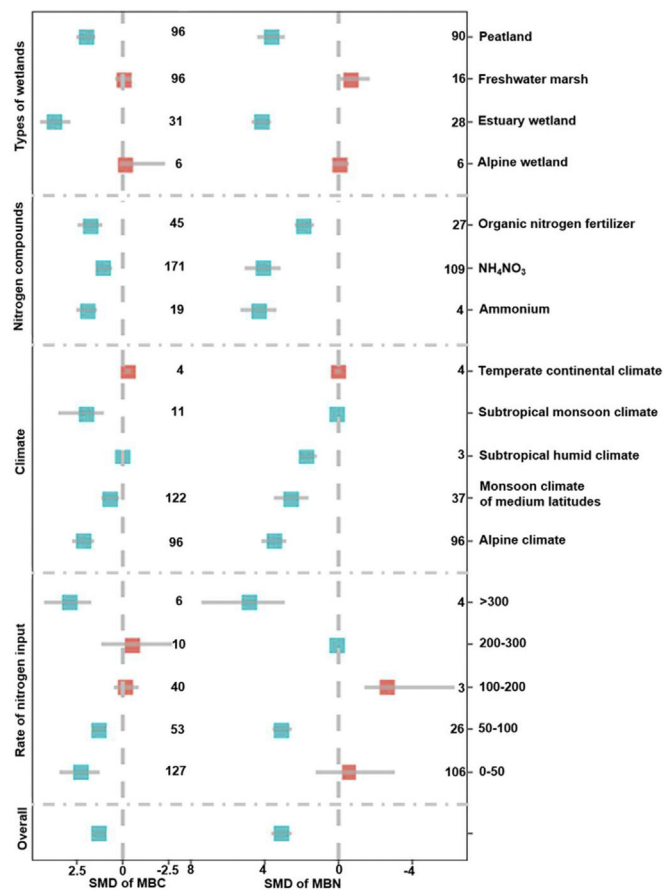


Fig. 4. Mean effect size for MBC and MBN from different wetland environments under nitrogen inputs. The numbers in the figure represent the number of case studies. For details on effect size interpretation, refer to Fig. 2.

Specially, the SMD was the largest for nitrogen input as NH₄⁺ compared to the other compounds. This means that NH₄⁺ input could significantly multiply soil microbial biomass formation. As seen from Fig. 4, nitrogen input rates of 0–100 kg ha⁻¹·yr⁻¹ significantly added the MBC (means of SMD = 1.28, 2.86 in Bootstrap CIs) and MBN (means of SMD = 3.01, 4.79 in Bootstrap CIs). In contrast, nitrogen input rates of more than 100 kg ha⁻¹·yr⁻¹ dwindled the MBC and MBN, except for the case of nitrogen input rates of more than 300 kg ha⁻¹·yr⁻¹. This indicated that lower nitrogen input rates significantly increased the soil microbial biomass contents.

4. Discussion

4.1. Impact of soil labile carbon and nitrogen compounds on greenhouse gas emissions

4.1.1. The effect of soil labile carbon and nitrogen on greenhouse gas emissions

The meta-analysis indicated that the nitrogen input significantly augmented the soil labile carbon and nitrogen content at the global scale (Fig. 3). The increase of soil labile carbon and nitrogen maybe because the nitrogen input changed the stability of soil aggregates and promoted the leaching of DOC, NH₄⁺-N and NO₃⁻-N from soil [39,52]. The meta-analysis also revealed that nitrogen input significantly multiplied decomposition of organic matter and subsequent gas formation (Figs. 2 and 3). However, nitrogen input influenced the activity of soil microbe by altering the ratio of

available carbon to nitrogen [53]. This study clarified that NH₄⁺-N and DOC play leading roles in greenhouse gas emissions according to a variation-partitioning analysis (Fig. 5). Higher NH₄⁺-N and DOC promoted CH₄ emission due to the increase in carbon availability, which resulted in more substrate being available for methanogens [54]. However, excessive NH₄⁺-N competitively inhibited CH₄ oxidation [55,56]. Meanwhile, ammonium oxidation produced toxic byproducts that noncompetitively inhibit CH₄ oxidation [57]. Additionally, the DOC could regulate carbon availability, thereby affecting the soil microbial activity [14,58]. The increase of DOC also altered the content of the soil inorganic nitrogen under the rewetting system because changing carbon availability would affect organic nitrogen mineralization and inorganic nitrogen assimilation [59]. Therefore, the effect of DOC on the CH₄ and CO₂ emissions is more important than NH₄⁺-N, and higher DOC content could stimulate bacteria that are responsible for organic matter decomposition and methanogenesis [54], leading to promote CH₄ and CO₂ emissions.

It is known that N₂O is mainly produced during nitrification but some N₂O can also be formed during denitrification, which is affected by nitrogen availability [60]. However, with the increase of anthropogenic activities, nitrogen input disrupted the balance of soil elemental stoichiometry, thereby affecting nitrogen availability [61,62]. The soil elemental stoichiometry determines the concentration and fractions of soil carbon and nitrogen [63,64]. A Pearson correlation analysis revealed the effect of soil labile nitrogen and carbon on N₂O emissions. The results showed that DOC had a significant and positive effect on N₂O emissions (Table 2) because higher DOC increased the nitrogen utilization rate and microbial activity [14]. Meanwhile, NH₄⁺-N also showed a significant and positive effect on N₂O emissions (Table 2) because soil microbes utilize NH₄⁺-N at a lower energy cost than NO₃⁻-N [14,65].

To elucidate which soil labile carbon and nitrogen are the main drivers of greenhouse gas emissions from wetland ecosystems under global nitrogen input, a structural equation model was established. Structural equation models are often utilized to investigate “latent” effects among various measured variables [9,66]. This research indicated that NH₄⁺-N showed a positive and

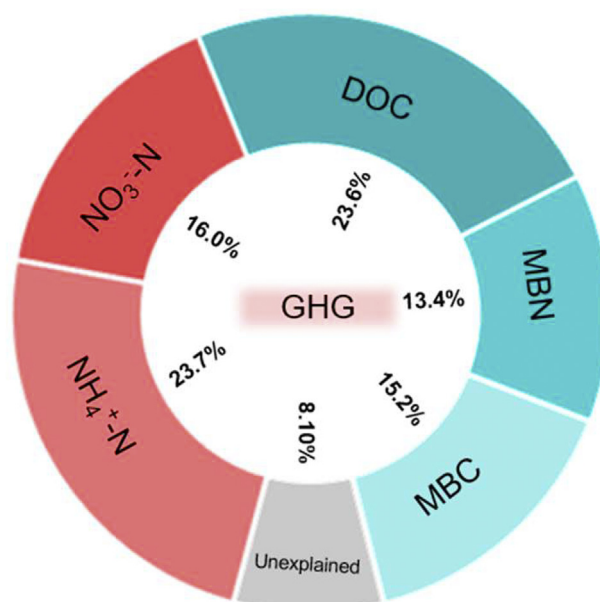


Fig. 5. Variation-partitioning analysis of the effects of soil labile carbon and nitrogen on greenhouse gas emissions. NH₄⁺-N, ammonium; NO₃⁻-N, nitrate; DOC, dissolved organic carbon; MBN, microbial biomass nitrogen; MBC, microbial biomass carbon.

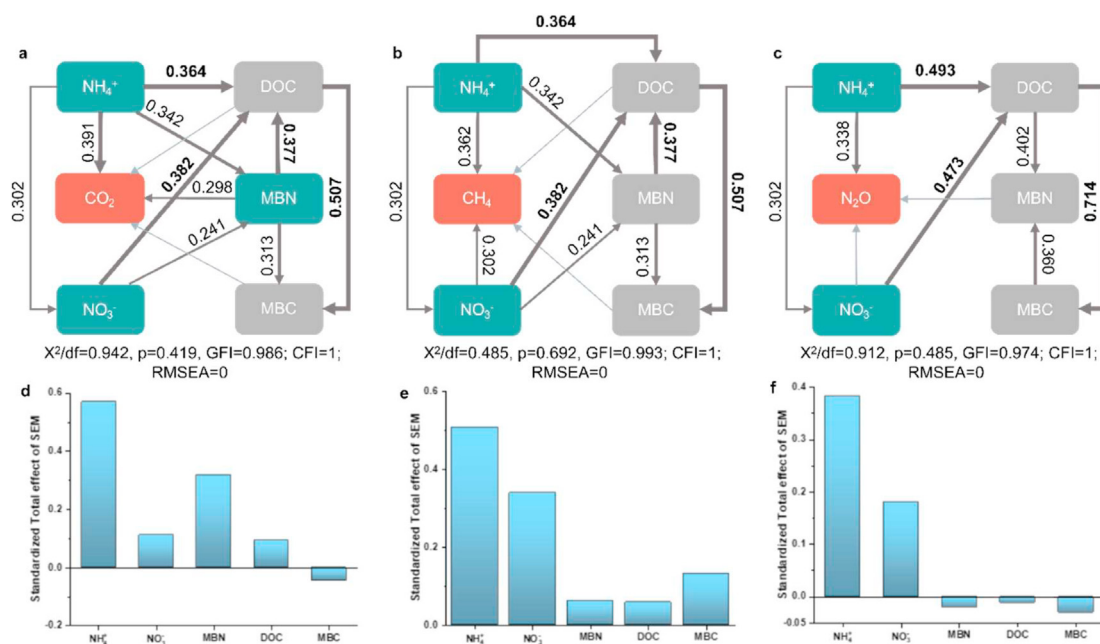


Fig. 6. Structural equation model (SEM) evaluating the direct effects on greenhouse gases (a–c) and the standardized total effect (direct plus indirect effects) derived from the SEM (d–f) on a global scale. The number represents the direct effects on greenhouse gas emissions. The various widths of the gray lines represent $p < 0.001, p < 0.005, p < 0.01$ and $p > 0.01$.

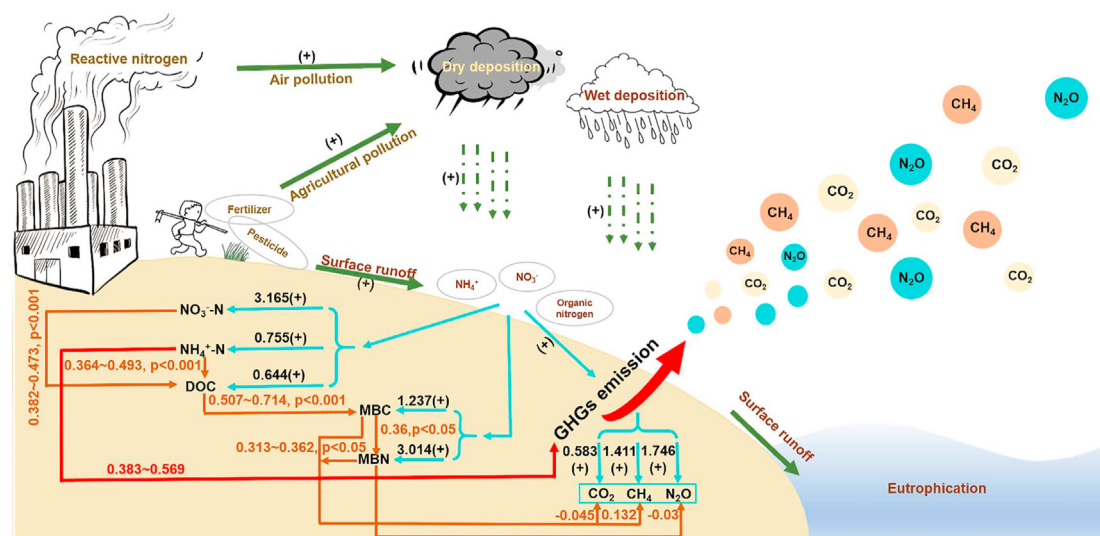


Fig. 7. The driving mechanisms of labile carbon and nitrogen on greenhouse gas emissions from wetland ecosystems under nitrogen input. The black numbers represent the effect size of various parameters, the red numbers represent the total effects of ammonium on greenhouse gas emissions, the brown numbers represent the direct effects of labile carbon and nitrogen on greenhouse gas emissions and the direct effects between various labile carbon and nitrogen.

significant direct effect on CO₂ emissions based on the structural equation model (Figs. 6 and 7), because higher NH₄⁺-N could increase the availability of carbon [67,68]. In addition, higher soil NH₄⁺-N could also promote plant photosynthesis and increase plant biomass, leading to an increase in the autotrophic respiration of plants [41]. However, the study revealed that NH₄⁺-N significantly and indirectly affected CH₄ and N₂O emissions by affecting the DOC (Table 3, Fig. 6). Additionally, the total effect of NO₃⁻-N (0.112–0.339) on a single greenhouse gas emission was larger than that of DOC (–0.02 to 0.093) according to Fig. 6(d, e, f) and Fig. 7. The effect of NO₃⁻-N on greenhouse gas emissions was significant and was indirectly shown by its effect on DOC (Table 3). These results reveal that DOC was the most important factor for

greenhouse gas emission. The results also suggested that DOC affected microbial activity more directly than NH₄⁺-N and NO₃⁻-N. The soil DOC is an organic carbon source directly utilized by microbes [69,70], and it is the main substrate and energy source for microbial metabolism [71]. Thus, DOC concentrations directly determine greenhouse gas emissions by regulating microbial metabolism in comparison to soil ammonium and nitrate [72–74].

4.1.2. The effect of soil microbial biomass on greenhouse gas emission

Although soil microbial biomass only accounts for 1%–5% of soil organic matter, it plays an important role in promoting material transformation and energy flow in the soil [18,75]. This research

Table 2
The Pearson relationships between greenhouse gas and labile carbon and nitrogen.

Parameters		NH ₄ -N	NO ₃ -N	DOC	MBN	MBC
CO ₂	Pearson Correlation	0.567	0.381	0.53	0.508	0.428
	p-value	<0.001	0.002	<0.001	<0.001	<0.001
CH ₄	Pearson Correlation	0.512	0.459	0.501	0.394	0.431
	p-value	<0.001	<0.001	<0.001	0.001	<0.001
N ₂ O	Pearson Correlation	0.383	0.282	0.43	0.175	0.277
	p-value	0.001	0.022	<0.001	0.159	0.024

showed that the global nitrogen input significantly added the soil microbial biomass carbon and nitrogen in wetland ecosystems (Figs. 4 and 7). It might be that nitrogen input multiplies nitrogen sources for microbial metabolism [76]. Nitrogen input could also change greenhouse gas production and emission by increased decomposition rates [77,78]. Thus, based on the Pearson correlation analysis (Table 2), the MBC and MBN showed a significant relationship with CH₄ and N₂O ($p < 0.01$). The soil microbial biomass is a sensitive measure of microbial activity [79]. Soil microbial biomass is more easily utilized by the microorganism for mineralization and assimilation than other fractions of soil organic matter [80]. Therefore, soil microbial biomass influenced greenhouse gas production and emission by changing the substrate content for organic matter mineralization and methanogenesis.

To clearly elucidate the effect of soil microbial biomass carbon and nitrogen on greenhouse gas emission, this research utilized the structural equation model to determine that MBN significantly and indirectly influenced the CH₄ and CO₂ emission by affecting MBC (Fig. 7, Table 3). The total effect of MBC on greenhouse gas showed that MBC negatively affected CO₂ and N₂O emissions and positively affected CH₄ emissions. Nitrogen input typically accelerated the anaerobic decomposition of MBC [14], and greater MBC provided more biologic residues as substrates for methanogens, which promoted the CH₄ generation [21,81]. This SEM also illustrated that MBC is the main pathway by which DOC affects greenhouse gas emissions (Figs. 6 and 0.507 to 0.714, $p < 0.001$). Soil microbial biomass carbon is the labile fraction of soil organic carbon, and has some particular characteristics including poor stability, fast turnover rate, easy mineralization and decomposition [82,83]. MBC can act as metabolism substrate for soil microbes and can sensitively affect the activity of functional microorganisms, resulting to promote greenhouse gas production and emission. By combining a variation-partitioning analysis and a structural equation model, this study inferred that MBC was the most direct indicator of the response of greenhouse gas emissions to nitrogen input in wetlands ecosystems.

4.2. Impact of environmental factors on greenhouse gas emissions

4.2.1. The effect of wetland type and climates on greenhouse gas emissions

Greenhouse gas emission is affected by the different physico-chemical properties (including plant types, water table, saline level and so on) of wetlands [84–86]. As seen in Fig. 2, for freshwater wetlands, nitrogen input significantly promoted CH₄ and CO₂

Table 3
The indirect effect of labile carbon and nitrogen to greenhouse gas emissions from wetland ecosystem under nitrogen input.

Parameters	NH ₄ -N	NO ₃ -N	MBN	DOC	MBC
CO ₂	0.177	0.112	0.021	-0.023	0
CH ₄	0.145	0.037	0.063	0.067	0
N ₂ O	0.045	-0.009	-0.02	-0.011	0

emissions compared to other types of wetlands. This may be because freshwater wetlands could decrease the effect of osmotic stress due to their lower saline level, leading to multiplying microbial activity and the decomposition rate of organic matter [30,87]. These results also revealed that for peatland, nitrogen input significantly promoted N₂O emission compared to other wetlands (Fig. 2). Although peatlands cover only 3% of the Earth's surface, they store one-third of the global organic carbon pool and conserve the higher nitrogen stocks [88,89]. Previous studies indicated natural peatlands display negligible N₂O emissions and can even act as net sinks for N₂O [89]. The external nitrogen enhanced microbial activity and triggered a priming effect that further facilitated the release of available nitrogen [105]. Thus, nitrogen input has a greater effect on N₂O emission from peatlands than other wetlands.

Climate can influence greenhouse gas emissions from wetlands by changing rainfall and temperature. A regression analysis revealed that greenhouse gas emission reduced as the mean annual precipitation (MAP) increased (Fig. 8 a, c, e). Fig. 2 shows that for temperate continental climate, nitrogen input dwindled CH₄ and CO₂ emissions. Although nitrogen input promoted N₂O emissions for all types of climate, it was significantly lower for temperate continental climate than other types of climates. These results indicated that nitrogen input promoted greenhouse gas emissions the least for the temperate continental climate. In particular, the regression analysis of the effect size and MAP as it ranged from 400 mm to 700 mm (typical for the temperate continental climate) indicated that the CH₄ and N₂O emissions increased with the increase of mean annual precipitation (Fig. 8). Therefore, lower greenhouse gas emissions in the temperate continental climate were likely due to soil drought and osmotic stress caused by lower MAP [90], which would destroy the microbial community and restrain the microbial activity in the wetland ecosystem [91].

The regression analysis revealed that greenhouse gas emissions reduced with the increase of mean annual temperature (MAT), except for N₂O (Fig. 8 b, d, f). As we know, temperature could influence the soil microbial activity and thereby affect N₂O emissions. It is likely that higher temperatures altered the content of the soil oxygen and available carbon, thus producing anoxic conditions for denitrifying bacteria [92,93]. In addition, when the soil temperature ranged from 10 to 35 °C, the denitrification activity increased with the increase in environmental temperature [94,95]. Contrary to N₂O emissions, the CO₂ and CH₄ emissions decreased as the environment warming. It is likely that the interaction of warming and nitrogen input increased the content of soil available nitrogen and carbon and decreased the soil pore water, leading to promote the activity of methanotrophs higher than methanogens [36]. Warming increased the soil carbon mineralization and nitrogen turnover rate, whereas nitrogen input promoted the assimilation of labile carbon by soil microbes and led to carbon sequestration and lower rates of nitrogen cycling [96,97]. It was probably that the soil carbon sequestration was greater than mineralization under the interaction of warming and nitrogen input, leading to a decrease in CO₂ emissions.

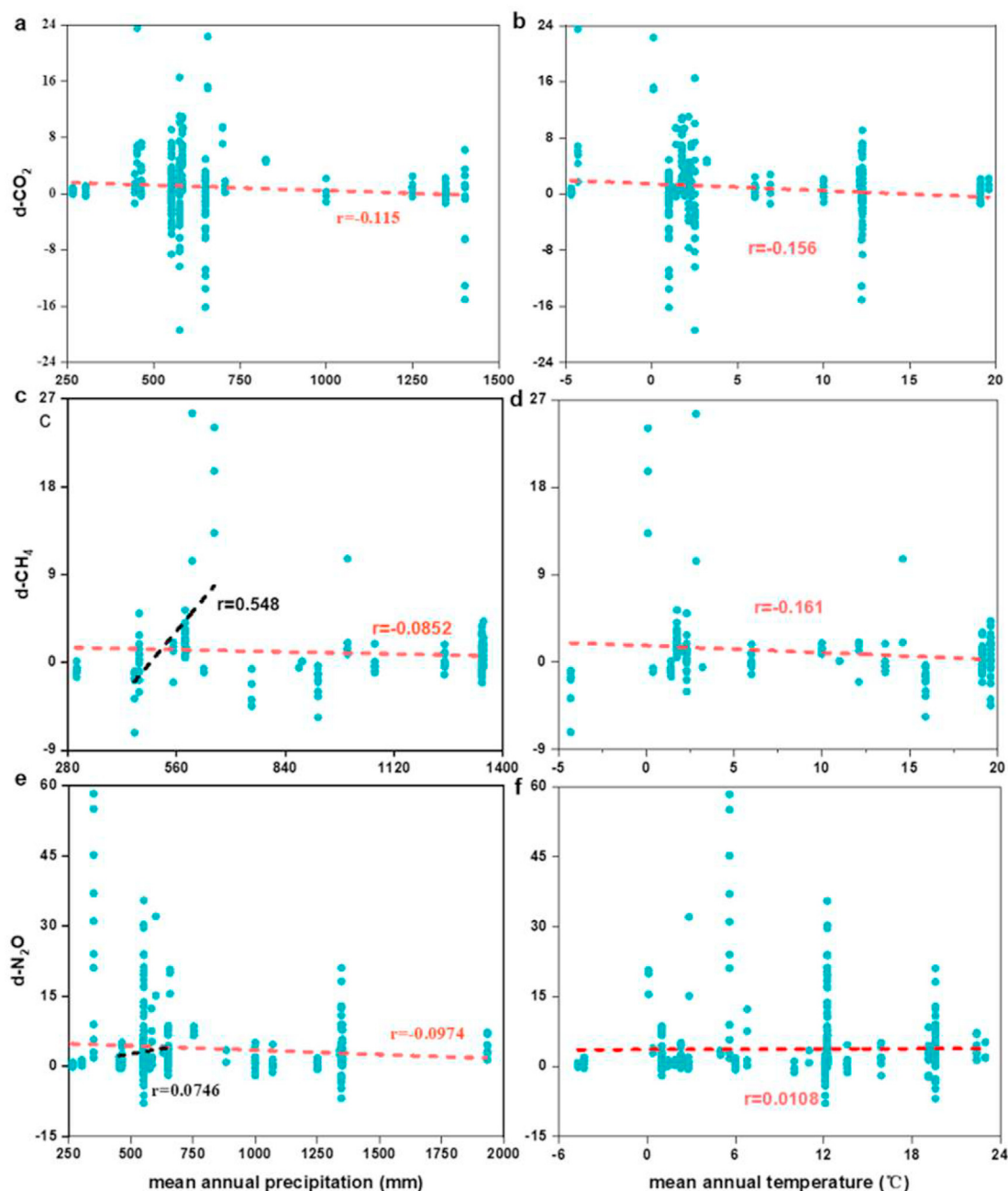


Fig. 8. Regression analysis for the effect size of greenhouse gas emission and mean annual precipitation, mean annual temperature at the global level. The red short dash represents the regression analysis of the effect size and mean annual precipitation and mean annual temperature among all studies. The black short dash represents the regression analysis of the effect size and mean annual precipitation in the range from 400 to 700 mm. d-CO₂ is the effect size of carbon dioxide, d-CH₄ is the effect size of methane, d-N₂O is the effect size of nitrous oxide.

4.2.2. The effect of nitrogen input on greenhouse gas emission

The continual increase of anthropogenic nitrogen inputs has already altered the rates of nitrogen cycling and nitrogen availability [19,98,105], which are affected by different nitrogen compounds and nitrogen input rates [99–101]. For different nitrogen compounds (Fig. 9 a, c, e), greenhouse gas emissions are promoted by nitrogen input as inorganic nitrogen (NH₄⁺-N, NO₃⁻-N and NH₄NO₃). Nitrogen input as organic nitrogen fertilizer suppresses CH₄ and CO₂ emissions and promotes N₂O emissions. This may be because soil microorganisms have different capacities to use the various nitrogen compounds, leading to the differences in greenhouse gas emissions [47]. Compared to other nitrogen compounds,

nitrogen input as NH₄⁺-N and NH₄NO₃ largely promoted greenhouse gas emissions (Fig. 9 a, c, e). The microbial utilization of ammonium is preferred over nitrate due to the low energy cost, implying that soil ammonium oxidation and organic matter decomposition were stimulated with the input of ammonium [102]; Tao et al., 2018). Notably, nitrogen input rates of 0–50 kg ha⁻¹·yr⁻¹ significantly promoted CH₄ emissions than other nitrogen input rates (Figs. 2 and 9 d). The CO₂ and N₂O emissions for nitrogen input rates of 50–100 kg ha⁻¹·yr⁻¹ were larger than for the other nitrogen input rates (Fig. 9 b, f). These results illustrated that lower nitrogen input significantly promoted greenhouse gas emissions from wetland ecosystems. This arose

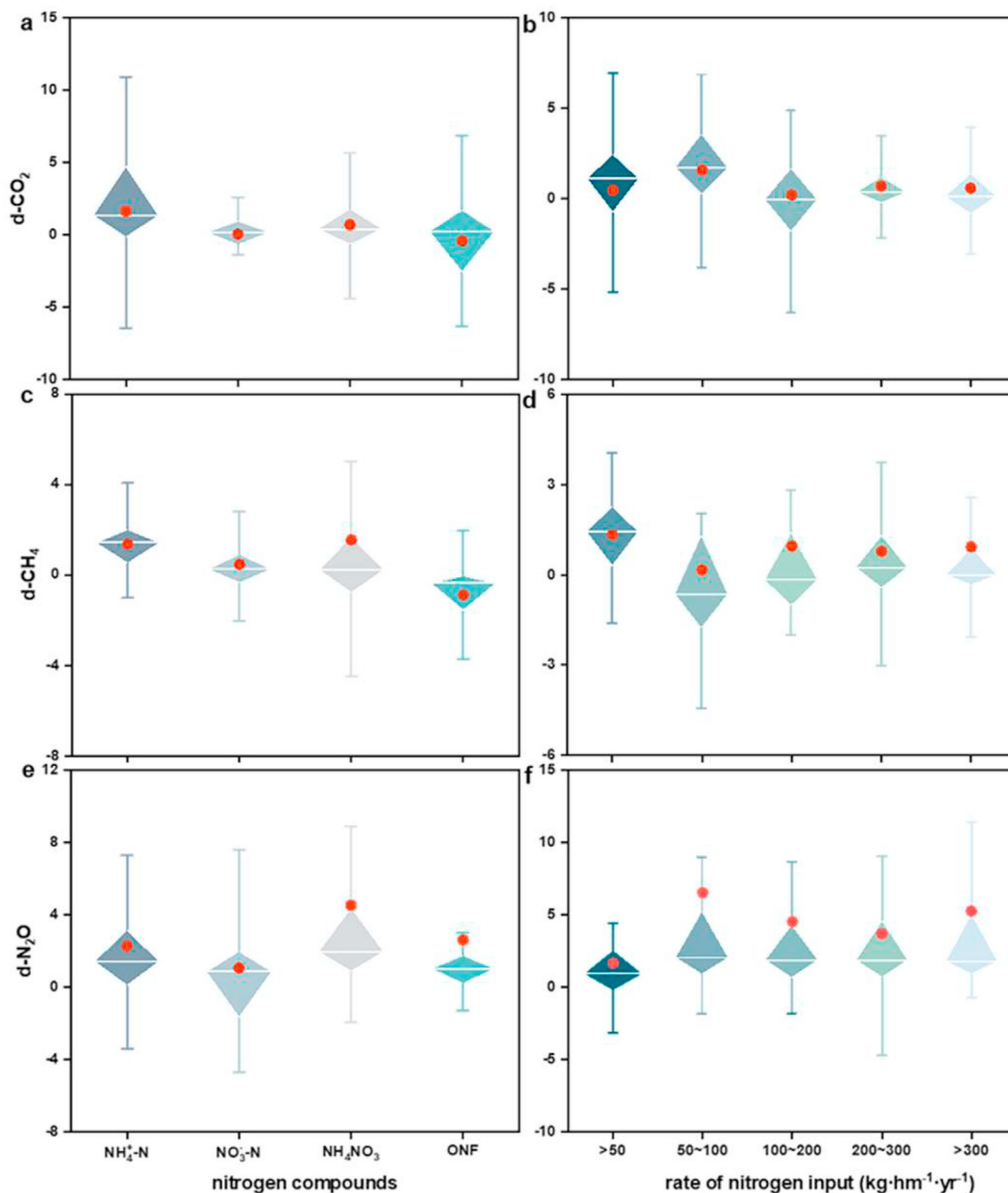


Fig. 9. The effect size of greenhouse gas emission among different nitrogen compounds and the nitrogen input rate. d-CO₂ is the effect size of carbon dioxide, d-CH₄ is the effect size of methane, and d-N₂O is the effect size of nitrous oxide. NH₄⁺-N represents ammonium, NO₃⁻-N represents nitrate, NH₄NO₃ represents ammonium nitrate, ONF represents organic nitrogen fertilizer. The red point represents the mean effect size of greenhouse gas emission under different nitrogen inputs and nitrogen input rates.

mainly because higher nitrogen input reduced the microbial biomass and activity by increasing the effect of osmotic stress and electrical conductivity [30,87]. Additionally, continuous and massive nitrogen input led to soil acidification, thereby directly or indirectly affecting the composition of the soil microbial diversity and community [103,104].

5. Conclusions

This meta-analysis found that nitrogen input significantly promoted greenhouse gas emissions from wetlands on a global scale. The driving effect of soil labile carbon and nitrogen and nitrogen

inputs on greenhouse gas emissions from wetlands ecosystems are summarized as follows:

- (1) DOC is the most important driving factor for greenhouse gas emissions from wetlands under global nitrogen input.
- (2) MBC is the most direct driving factor for greenhouse gas emissions from wetlands under global nitrogen input.
- (3) Nitrogen input to freshwater wetlands shows the most significant and positive effects on CH₄ and CO₂ emissions from wetlands under global nitrogen input, whereas nitrogen input to peatland largely and significantly promotes N₂O emissions compared to other wetlands.

- (4) Nitrogen input as ammonium compounds and at lower rates show the most significant and positive effects on greenhouse gas emissions from wetlands under global nitrogen input.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at <https://doi.org/10.1016/j.ese.2020.100063>.

Author contributions

Mengli Chen and Yi Chen designed the study. Mengli Chen, Lian Chang, Junmao Zhang and Fucheng Guo collected references and got data. Mengli Chen wrote and modified this article. Yi Chen, Jan Vymazal and Qiang He reviewed this article.

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