



Effects of salinity on methane emissions and methanogenic archaeal communities in different habitat of saline-alkali wetlands

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Abstract

The increase in temperature caused by global climate change has promoted the salinization of wetlands. Inland saline-alkaline wetlands have an environment of over-humidity and shallow water and are hot spots for CH₄ emissions. However, there are few reports on the effect of salinity on CH₄ emissions in inland saline-alkaline wetlands. This study conducted simulation experiments of increased salinity to investigate the impact of salinity, habitat, and their interactions on CH₄ emissions, as well as to examine the response of methanogenic archaea to salinity. Overall, salinity inhibited CH₄ emissions. But there were different responses in the three habitat soils. Salinity decreased the relative abundance of methanogenic archaea and changed the community structure. In addition, salinity changed soil pH and dissolved organic carbon (DOC) and ammonium (NH₄⁺) concentrations, which were significantly correlated with methanogenic archaea. Our study showed that salinity changed the soil physicochemical properties and characteristics of the methanogenic archaeal community, affecting CH₄ emissions.

Keywords Salinity · Habitat · Inland saline-alkaline wetlands · CH₄ emissions · Methanogenic archaea

Introduction

As the second largest greenhouse gas after CO₂, CH₄ contributed about 22% to the greenhouse effect (Wang et al. 2018). Atmospheric CH₄ contents have increased dramatically since the Industrial Revolution, already rising from 719 ppb in 1750 to 1895 ppb in 2021 (IPCC 2021; Lan et al. 2022). Sources of CH₄ include wetland systems (including swamps, sediments, rice fields, etc.), ruminant digestive systems, landfills, leakage during energy production and utilization, and sewage treatment systems (Kirschke et al. 2013). Among them, wetlands produce about 164 Tg CH₄ per year,

contributing about 1/3 of global CH₄ emissions, and are the most important source of CH₄ emissions (Bridgman et al. 2013).

CH₄ is produced by archaea-dominated anaerobic decomposition of organic matter (Hofmann et al. 2016; Gütlein et al. 2018). The known methanogenic archaea are divided into seven orders (Borrel et al. 2014). The community and diversity of methanogenic archaea are influenced by various environmental factors. For example, the abundance of methanogenic *mcrA* genes decreased with increasing pH in acidic rice fields (Luo et al. 2022). In studies with a pH range of 4.0–10.0, extreme pH reduces the relative abundance of acetoclastic methanogens responsible for acetic acid breakdown, while increased those of hydrogenotrophic and hydrogen- or acetic acid-utilizing methanogens (Qiu et al. 2023). In addition, soil organic matter is an important factor affecting methanogenic archaea and CH₄ emissions (Zhang et al. 2018; Wu et al. 2022). Soil dissolved organic carbon (DOC) is an important unstable carbon substrate that provides carbon sources for methanogenic archaea and has a positive effect on CH₄ emissions (Kong et al. 2019; Wang et al. 2021). Nitrate (NO₃⁻) and Fe³⁺ can be used as electron acceptors to participate in the methane oxidation process and affect CH₄ emissions (Fan et al. 2021; Chen et al. 2022). Cover plant is also an important factor affecting

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the methanogen community and CH₄ emissions (Duan et al. 2022; Venturini et al. 2022). The peatlands where vascular plants grow are dominated by acetoclastic methanogens (Ström et al. 2003). In peatlands with non-vascular plants, hydrogenotrophic methanogens are mostly present (Nakagawa et al. 2002).

Methanogenic archaea are sensitive to salinity. In general, increased salinity can inhibit methanogenic archaeal activity, and the number of methanogenic archaea will decrease accordingly, thus reducing the CH₄ emissions in wetlands (Sun et al. 2013). In addition, increased salinity alters microbial community structure (Pattnaik et al. 2000; Feng et al. 2023). In a salinity study of mangrove peat soil, the abundance of microorganisms did not change, but the community structure changed significantly (Chambers et al. 2016). When a large amount of NaCl was input into coastal wetlands, CH₄ emissions from the soil surface was significantly inhibited (Chambers et al. 2011). However, lower concentrations of salt input had some promotion or no significant effect on CH₄ emissions in wetlands (Weston et al. 2011; Krauss and Whitbeck 2012; Konnerup et al. 2014). In the Mobile Bay freshwater swamp, CH₄ emissions did not change significantly in different salinity areas (Wilson et al. 2015). Under high salinity, species with high salinity tolerance can replace species with low salinity tolerance and become the dominant microorganisms in the environment (Rath et al. 2019; Zhang et al. 2019). In conclusion, the effects of salinity on CH₄ emissions and methanogenic archaeal communities were spatially variable. At present, most studies on the influence of salinity on CH₄ emissions and methanogenic archaea focus on coastal wetlands (Dang et al. 2019; Chen et al. 2020b). However, little research has been done in inland saline-alkaline wetlands.

In recent years, global warming has accelerated the evaporation of water from wetlands, resulting in wetland salinization (Jeppesen et al. 2020). Saline groundwater conducts upwards and surface water evaporates, which leads to an increase in wetland salinity (Herbert et al. 2015). Zhalong wetland is located in the Songnen Plain of China, which is an area where inland saline-alkali wetlands are concentrated. The wetland is low-lying and flat, with many swamps and a large amount of water evaporation, which form an ecological environment with a slightly higher salinity. The slightly alkaline soil is suitable for the growth of methanogens, making this wetland a hot spot for CH₄ emissions (Liu et al. 2019). In addition to reed (*Phragmites australis*), the dominant vegetation in Zhalong wetland also includes star grass (*Puccinellia tenuiflora*) and guinea grass (*Leymus chinensis*).

Due to the severe salinization of the Zhalong wetland, we collected soil from three habitats in the wetland to simulate the increase in salinity (Liu et al. 2019; Luo et al. 2022).

The CH₄ emission process and soil physicochemical characteristics were measured. The community composition and relative abundance of archaea were studied by using high-throughput sequencing and quantitative PCR technology. The objectives of this study were to reveal the effects of increased salinity on CH₄ emissions and associated microbes in inland saline-alkaline wetlands and to explain key environmental drivers. This study helps to understand the response of CH₄ emissions and methanogenic archaea to the salinization of wetlands, which will provide a theoretical basis for subsequent research on CH₄ emissions in salinized soils.

Materials and methods

Soil sampling and experimental design

The soil used in this study was collected in July 2022 from the surface (0–20 cm) of the Zhalong wetland (46° 52′–47° 32′ N, 123° 47′–124° 37′ E) in Heilongjiang Province, China. Zhalong wetland has a mid-temperate climate, with an average annual precipitation of 420 mm, an annual average temperature of 3.9 °C, and a freezing period of 7 months (Gao et al. 2018). The specific sampling process was described before (Liu et al. 2019). Each sample site was divided into 3 plots. Homogenous mixing was performed after collecting at least 3 soil samples per plot. The soil samples were stored at low temperatures and transported to the laboratory and were divided into three parts: a part of fresh soil was extracted with 1 mol L⁻¹ KCl to determine the content of inorganic nitrogen (Wang et al. 2023), a part of the soil was air-dried for soil physicochemical analysis, and another part was stored at –80 °C for later experiments. The details of the soil are shown in Table 1. The dominant vegetation in sites was *Puccinellia tenuiflora* (H1), hygrophyte *Phragmites australis* (H2), and aquatic *Phragmites australis* (H3), respectively. The total organic carbon (TOC) and total nitrogen (TN) contents in H2 soils were significantly higher than those in H1 and H3 soils ($P < 0.05$) (Table 1). The soil salinity in the H1 site was significantly higher than that in the other two sites, which were 0.04% (H1), 0.01% (H2), and 0.01% (H3), respectively.

We added 50 mL of sterile anaerobic saline with different NaCl concentrations to 120 mL serum bottles. Fresh soil (equivalent to 10 g of dry soil) was added to the serum bottle in an anaerobic glove box (Coy, USA) to establish anaerobic microcosms. Five treatments were set up for each habitat soil: (i) control without NaCl addition (CK), (ii) 1.0% salinity (S1), (iii) 2.5% salinity (S2), (iv) 3.5% salinity (S3), and (v) 5.0% salinity (S4), with three replicates. The serum

Table 1 Site information and soil properties.

Sites	H1	H2	H3
Dominant vegetation	<i>Puccinellia tenuiflora</i>	Hygrophyte <i>Phragmites australis</i>	Aquatic <i>Phragmites australis</i>
pH	10.5 ± 0.1a	8.7 ± 0.1c	9.3 ± 0.1b
EC (µs cm ⁻¹)	744.3 ± 1.5a	152.6 ± 1.5b	110.3 ± 0.4c
TOC (g kg ⁻¹)	10.5 ± 0.8b	28.6 ± 0.4a	9.6 ± 0.6b
DOC (mg kg ⁻¹)	1,119.6 ± 5.7c	1,563.6 ± 8.5a	1,407.7 ± 5.5b
NH ₄ ⁺ (mg kg ⁻¹)	1.2 ± 0.3b	3.2 ± 0.4a	0.5 ± 0.1c
NO ₃ ⁻ (mg kg ⁻¹)	2.6 ± 0.4a	1.8 ± 0.1b	1.0 ± 0.1c
TN (g kg ⁻¹)	1.5 ± 0.1b	2.5 ± 0.1a	0.7 ± 0.1c
Salinity (%)	0.04a	0.01b	0.01b

Different letters indicate significant differences among sites ($P < 0.05$)

bottles were sealed with sterile neoprene septa and secured with aluminum caps to maintain an anaerobic environment. All serum bottles were incubated for 68 days at 25 °C in the dark without shaking.

Measurement of soil CH₄ emissions and physicochemical characteristics

The 2 mL of gas samples was taken from the headspace of each bottle, and the concentration of CH₄ was measured by a gas chromatograph (Agilent 8890A, Agilent Technologies, Palo Alto, CA, USA) equipped with a flame ionization detector (FID). 80/100 mesh HayeSep Q column with an inner diameter of 2 mm was used, and the carrier gas was high-purity N₂. The maximum CH₄ emission rate was calculated in the linear range of CH₄ emission increase. The rate of CH₄ emission was calculated using the following formula (Luo et al. 2022):

$$F = \rho \times \frac{V}{m} \times \frac{dc}{dt} \times \frac{273}{273 + T} \times \frac{12}{16} \quad (1)$$

where F is CH₄ emission rate (mg kg⁻¹ d⁻¹), ρ is the density of CH₄ at standard temperature and pressure, V (m³) is the headspace volume of the serum bottle, m (kg) is the dry soil weight, $\frac{dc}{dt}$ (ppm d⁻¹) is the changed concentration of CH₄ in the unit time (d), and T is the incubation temperature.

After the incubation, soil physicochemical characteristics were measured for data analysis. Soil pH was measured with a pH meter. Soil electrical conductivity (EC) was measured with a conductivity meter (DDS-307, Leici, Shanghai, China). Soil dissolved organic carbon (DOC) was measured with a total organic carbon analyzer (Multi-N/C 3100, Analytik Jena, Germany). Ammonium (NH₄⁺) and nitrate (NO₃⁻) concentrations were measured using a continuous flow analyzer (Seal Analytical AA3, Norderstedt, Germany). The concentrations of Fe³⁺ and Fe²⁺ were determined by colorimetry (Wallmann et al. 1993; Haese et al. 1997). Active iron was extracted from the soil with an HCl solution.

Active iron and Fe²⁺ concentrations were determined with a 1, 10-phenanthroline and hydroxylamine hydrochloride. Then, Fe³⁺ content was obtained by calculating the difference between the two.

DNA extraction and high-throughput amplicon sequencing of archaeal 16S rRNA gene

To explore the effect of salinity on the archaeal community, high-throughput sequencing of archaeal 16S rRNA genes was performed on the CK and S4 treated samples of each habitat soil after the incubation. First, DNA in soil (0.5 g) was extracted according to the instruction manual of SPINeasy DNA Kit for Soil (MP Biomedicals, Santa Ana, CA, USA). The concentration of DNA was determined with a NanoDrop (NanoDrop OneC, Thermo Scientific, USA). The DNA samples were stored in a -20 °C refrigerator. PCR amplification on V4–V5 regions of archaeal 16S rRNA gene used 524F10extF (TGYCAGCCGCCGCGGTAA) and Arch958RmodR (YCCGGCGTTGAVTCCAATT) primer pair (Liu et al. 2016). The amplification reaction system (20 µL) included 10 µL 2×Pro Taq, 0.8 µL each for upstream and downstream primers (5 µM), and 10 ng µL⁻¹ DNA template. PCR reaction parameters were 95 °C for 3 min, followed by 35 cycles of 95 °C for 30 s, 55 °C for 30 s, and 72 °C for 45 s, and then 72 °C for 10 min. PCR amplification was performed by a thermocycler (GeneAmp 9700, ABI, USA). PCR products were detected by 2% agarose gel electrophoresis and recovered using the DNA Gel Extraction Kit (AxyPrep, USA). The sequencing was performed on the Illumina MiSeq PE300 platform (Shanghai Majorbio Biopharm Technology Co., Ltd.).

Statistical analysis

Statistical analysis of data was performed using SPSS 26.0 (SPSS, Inc., Chicago, IL) (Morgan et al. 2019). One-way analysis of variance (ANOVA) was used to

explore the differences in soil physicochemical properties and CH₄ emission rate in response to salinity. The influence of salinity, soil habitat, and their interaction on CH₄ emissions and soil physicochemical properties was analyzed by multifactor analysis of variance. Correlations between CH₄ emissions, archaeal genus composition, and environmental factors were assessed using Pearson’s correlation analysis. The quality filtering of raw reads was conducted by Fastp software (Chen et al. 2018). FLASH was used to merge paired-end reads (Magoč and Salzberg 2011). Then, the data was processed by using sequence denoising method (DADA2) to obtain amplicon sequence variant (ASVs) representative sequence and abundance information. Sequences with a similarity higher than 97% were classified as operational taxonomic units (OTUs) using Usearch (version 7.1) software (Edgar 2013). A classification was assigned to each sequence using the Ribosome Database Project (RDP) classifier (version 2.2, 70% confidence threshold) based on the SILVA 128 reference database (Quast et al. 2012). Alpha diversity indices were calculated using mothur (version 1.30) software. Canoco 5 was used to perform redundancy analysis (RDA) to explore the relationship between soil physicochemical properties and archaeal communities (Šmilauer and Lepš 2014). Draw various line charts, histograms, and heat maps through Origin 2021 (Moberly et al. 2018). The data were means ± standard deviations (Mean ± SD).

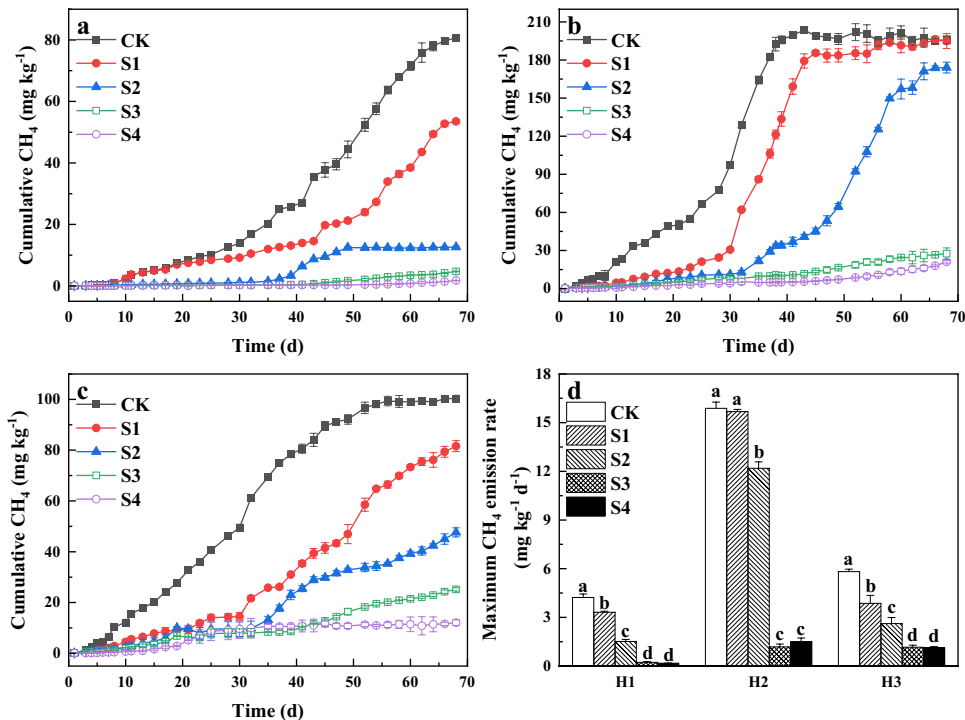
Results

CH₄ emissions and soil physicochemical properties

In H1 soil, there was no significant difference in CH₄ emissions between S1 and CK treatments in the first 25 days. After 25 days, CH₄ emissions of S1 treatment were significantly reduced (Fig. 1a). After 68 days of incubation, cumulative CH₄ emissions with S1 (53.53 ± 0.59 mg kg⁻¹) significantly decreased by 33.67% (Fig. 1a). The maximum CH₄ emission rate of S1 treatment (3.31 ± 0.06 mg kg⁻¹ d⁻¹) was also significantly lower than that of CK (4.23 ± 0.21 mg kg⁻¹ d⁻¹) (Fig. 1d). In the early stage of incubation (the first 40 days), there was no significant increase in CH₄ emissions of S2 treatment. CH₄ emissions of S2 treatment occurred only in the middle of the incubation (40–50 d). S3 and S4 treatments considerably limited CH₄ emissions, which remained at low levels throughout. After anaerobic cultivation, CH₄ accumulations of S2, S3, and S4 treatments were 12.62 ± 0.30, 4.70 ± 0.13, and 1.76 ± 0.05 mg kg⁻¹, respectively (Fig. 1a).

In H2 soil, CH₄ emissions of S1 treatment were delayed, but there was no significant difference in the final CH₄ accumulations compared with CK treatment. At the end of incubation, CH₄ accumulations of S1 and CK treatments were 194.92 ± 5.87 and 195.86 ± 3.14 mg kg⁻¹, respectively (Fig. 1b). There was also no significant difference in the

Fig. 1 The effect of salinity on the CH₄ emission process in H1 (a), H2 (b), and H3 (c) soils and maximum CH₄ emission rate (d). Error bars = SD, *n* = 3. Different letters indicate significant differences among treatments within each habitat (*P* < 0.05)



maximum CH₄ emission rates of the two treatments, which were $15.88 \pm 0.39 \text{ mg kg}^{-1} \text{ d}^{-1}$ (CK) and $15.68 \pm 0.14 \text{ mg kg}^{-1} \text{ d}^{-1}$ (S1), respectively (Fig. 1d). Different from H1 soil, the CH₄ accumulations in H2 soil treated with S2 were less affected by the increased salinity ($174.06 \pm 4.23 \text{ mg kg}^{-1}$). Cumulative CH₄ emissions of S3 and S4 treatments were much lower compared to the other treatments with lower salinity, reaching up to 27.52 ± 4.54 and $21.04 \pm 0.90 \text{ mg kg}^{-1}$, respectively.

Similarly, increased salinity delayed CH₄ emissions in H3 soil. At the initial stage of incubation, CH₄ emission of the treatments with increased salinity was significantly lower than that of CK treatment (Fig. 1c). It mainly occurred in the middle and late stages of incubation (after 30 days). Furthermore, CH₄ emissions decreased gradually with the increase of salinity gradient (Fig. 1c). At the end of incubation, CH₄ accumulations in each treatment were 100.32 ± 1.24 (CK), 81.58 ± 2.14 (S1), 47.62 ± 1.79 (S2), 25.18 ± 0.64 (S3), and $12.07 \pm 1.12 \text{ mg kg}^{-1}$ (S4). The treatments with increased salinity reduced CH₄ emissions by 18.68%, 52.53%, 74.90%, and 87.97%, respectively. The maximum CH₄ emission rate also gradually and significantly decreased with the increase of salinity (Fig. 1d).

Salinity, habitat, and the interaction of these two variables significantly affected cumulative CH₄ emissions ($P < 0.001$) (Table 2). Salinity and habitat had significant effects on soil pH, EC, DOC, NH₄⁺, Fe³⁺, and Fe²⁺ contents ($P < 0.05$). Among them, DOC, Fe³⁺, and Fe²⁺ contents were also affected by the interaction between salinity and habitat ($P < 0.05$). Pearson's correlation analysis showed that CH₄ emissions were significantly affected by pH, EC, DOC, and NH₄⁺

contents ($P < 0.05$) (Table 3). DOC and NH₄⁺ contents were significantly positively and negatively correlated with EC, respectively ($P < 0.01$).

Analysis of archaeal communities

Archaeal community analysis was performed by high-throughput sequencing of 16S rRNA genes, and 7 major lineages were found in samples at the order level of archaea (Fig. 2). As salinity increased, the summed relative abundance of methanogenic orders decreased in all three sites, by 35.25% (H1), 29.31% (H2), and 14.34% (H3), respectively. There were mainly 6 orders of methanogenic archaea: *Methanobacteriales* (12.64~41.79%), *Methanosarciniales* (0.54~41.93%), *norank_c_Bathyarchaeia* (1.25~42.31%), *Methanomicrobiales* (0~4.23%), *Methanocellales* (0.04~2.53%), and *Methanomassiliicoccales* (0.04~2.54%). In H1 and H3 soils, the relative abundance of *Methanosarciniales* was most affected by increasing salinity, decreasing by 41.39% and 20.00%, respectively. In H2 soil, the relative abundance of *Methanosarciniales* was reduced by only 7.30%, while that of *Bathyarchaeia* was reduced by 23.78%.

We calculated the total archaeal richness and diversity of each sample using alpha-diversity analysis (coverage, Chao1 diversity, Shannon index, and Simpson index) (Fig. 3). The community coverage was >99%, indicating that the sequencing depth was sufficient to cover most species information in the sample. For soils in the three habitats, the Chao1 diversity was significantly reduced by increasing salinity ($P < 0.05$) (Fig. 3a), indicating that elevated salinity reduced the richness of soil archaeal communities. Especially in the H2 soil with the most drastic

Table 2 Effects of salinity, habitat, and their interactions on soil characteristics.

Treatments	CH ₄	pH	EC	DOC	NH ₄ ⁺	NO ₃ ⁻	Fe ³⁺	Fe ²⁺
Salt	***	*	***	***	***	*	***	**
Habitat	***	***	*	***	***	ns	***	***
Salt × habitat	***	ns	ns	***	ns	*	***	*

“ns,” “*,” “**,” and “***” stand for no significant, $P < 0.05$, $P < 0.01$, and $P < 0.001$, respectively

Table 3 Pearson's correlation analysis of soil CH₄ emissions and physicochemical properties.

	CH ₄	pH	EC	DOC	NH ₄ ⁺	NO ₃ ⁻	Fe ³⁺	Fe ²⁺
CH ₄	1	-0.34*	-0.69**	-0.72**	0.86**	0.25	-0.22	-0.16
pH		1	-0.16	-0.13	-0.48**	0.14	0.68**	0.09
EC			1	0.93**	-0.65**	-0.26	-0.19	0.22
DOC				1	-0.55**	-0.40**	0.02	0.44**
NH ₄ ⁺					1	0.08	-0.12	0.03
NO ₃ ⁻						1	-0.02	-0.35*
Fe ³⁺							1	0.67**
Fe ²⁺								1

* $P < 0.05$; ** $P < 0.01$

Fig. 2 The community composition of archaea at order level

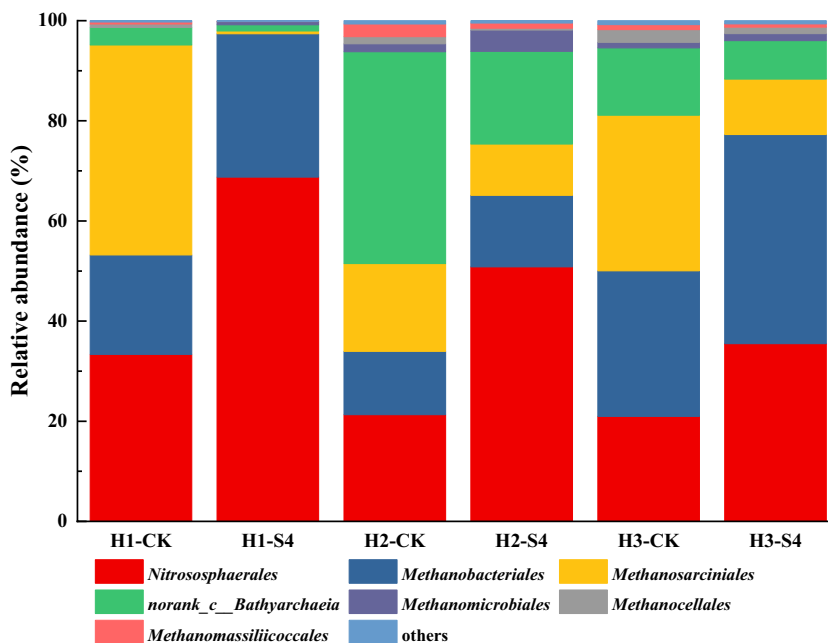
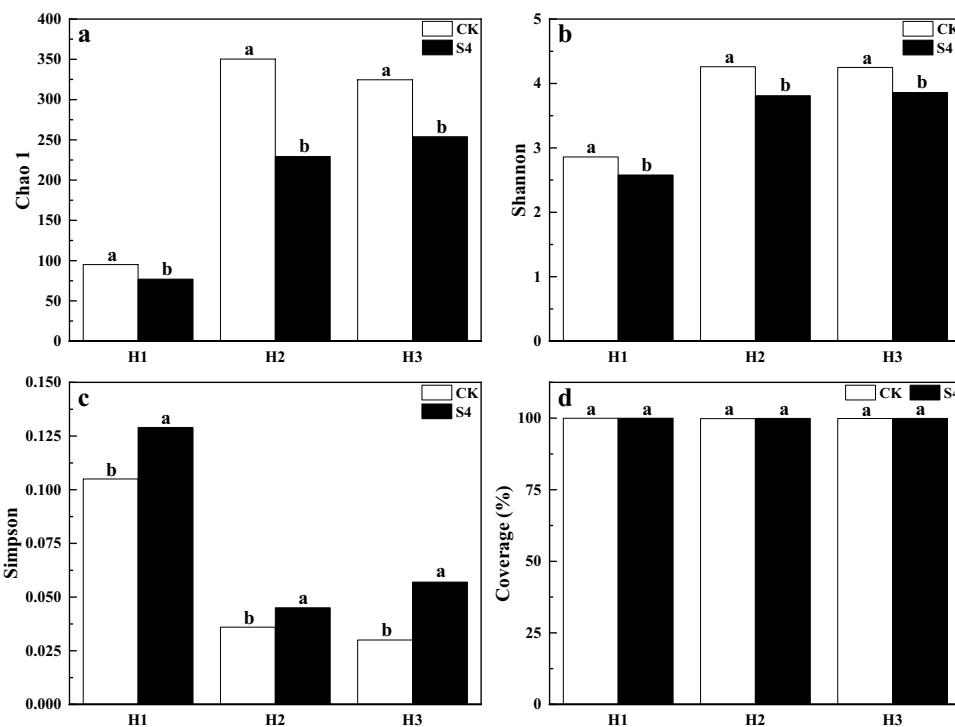


Fig. 3 Alpha-diversity of archaeal community measured as (a) Chao1, (b) Shannon, (c) Simpson and (d) Coverage indexes

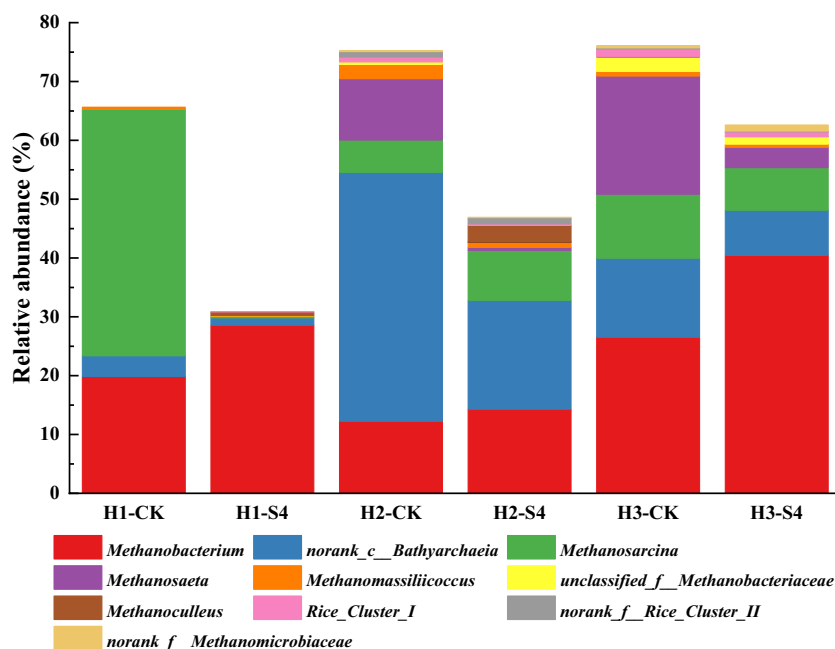


change, the Chao1 index decreased from 350.27 to 229.26. It is worth noting that the richness of archaeal communities in H1 soils was significantly lower than that in H2 and H3 soils, regardless of the addition of salt. Comparing the Shannon and Simpson indices, we found that increased salinity reduced the diversity of archaeal communities in soil (Fig. 3b and c).

Community structure of methanogenic archaea at the genus level

As salinity increased, the relative abundance of methanogenic archaea was significantly reduced and the dominant archaea were changed (Fig. 4). The relative abundance of *Methanobacterium* increased by 8.71% (H1), 2.03% (H2),

Fig. 4 The distribution of methanogenic archaeal genera



and 13.90% (H3), respectively, with the increase of salinity. Affected by salinity, the relative abundance of methanogenic archaea decreased the most in H1 soil (from 65.65 to 30.87%). Among them, the genus of *Methanosarcina* changed most drastically, and its relative abundance decreased from 41.88 to 0.31%. In H2 soil, salinity reduced the relative abundance of methanogenic archaea from 75.25 to 46.93%. Among them, the relative abundance changes of *Bathyarchaeia* (decreased from 42.31 to 18.53%) and *Methanosaeta* (decreased from 10.43 to 0.48%) were the most significant (Fig. 4). However, salinity had the least effect on the relative abundance of methanogenic archaea in H3 soil, reducing the relative abundance of methanogenic archaea from 76.09 to 62.60% (Fig. 4). The relative abundance of *Methanosaeta* decreased most obviously, with a decrease of 16.62%.

The relationship between archaea and soil physicochemical properties and CH₄ emissions

Redundancy analysis (RDA) was used to evaluate the influences of soil physicochemical factors on archaeal communities at the order level (Fig. 5). The first and second axes explained 58.53% and 28.38% of the variance in archaeal community composition, respectively. The first axis was positively correlated with NH₄⁺ and NO₃⁻ concentrations and negatively correlated with pH, DOC, and Fe²⁺ contents. The second axis was positively correlated with pH and NO₃⁻ and negatively correlated with NH₄⁺, DOC, and Fe²⁺ concentrations (Fig. 5). DOC and NH₄⁺ contents had a significant effect on the archaeal community composition ($P < 0.05$). We observed the positive correlation between

Methanosarciniales and NO₃⁻ concentrations and *Bathyarchaeia* and NH₄⁺ concentrations. This suggests that archaeal community was influenced by soil inorganic nitrogen.

To reveal the impact of environmental variables on genus-level archaea, we plotted a heatmap of correlations between archaeal at the genera level (top 15) and physicochemical properties (Fig. 6). *Bathyarchaeia*, *Methanomassiliicoccus*, *Candidatus_Methanoperedens*, and NH₄⁺ concentrations were significantly positively correlated ($P < 0.05$). *Methanosarcina* and NO₃⁻ concentrations had a significant positive correlation ($P < 0.05$). *Methanocella* was negatively correlated with EC ($P < 0.05$) and DOC ($P < 0.01$) significantly. *Rice_Cluster_II* had a significant negative correlation with soil pH ($P < 0.01$). *Methanobacteriaceae* ($P < 0.01$) and *Rice_Cluster_I* ($P < 0.05$) were significantly negatively correlated with Fe²⁺ concentrations. There was a significant negative correlation between Fe³⁺ concentration and Shannon index ($P < 0.05$). *Methanomassiliicoccus* was significantly positively correlated with CH₄ emission ($P < 0.05$).

Discussion

Salinity reduced CH₄ emissions

Our study has shown that salinity suppressed CH₄ emissions, which is consistent with the findings in Cumberland Marsh Preserve (Dang et al. 2019). However, the inhibitory effects of salinity were different in the three habitat soils. Salinity reduced CH₄ emissions by 33.67–97.82% (H1), 0.48–89.26% (H2), and 18.68–87.97% (H3), respectively. This may be influenced by multiple effects of wetland

Fig. 5 Redundancy analysis (RDA) of soil physicochemical characteristics and archaeal communities at order level

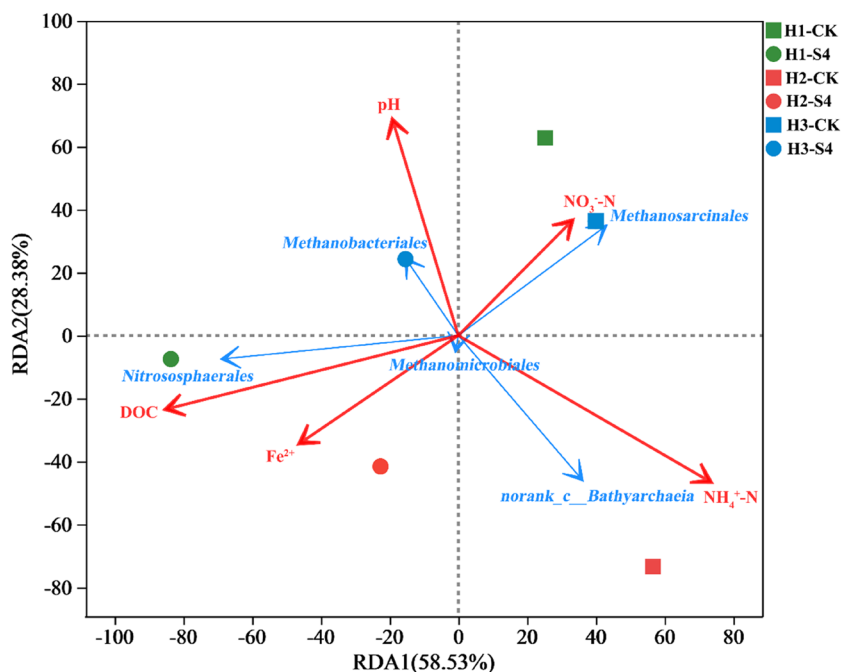
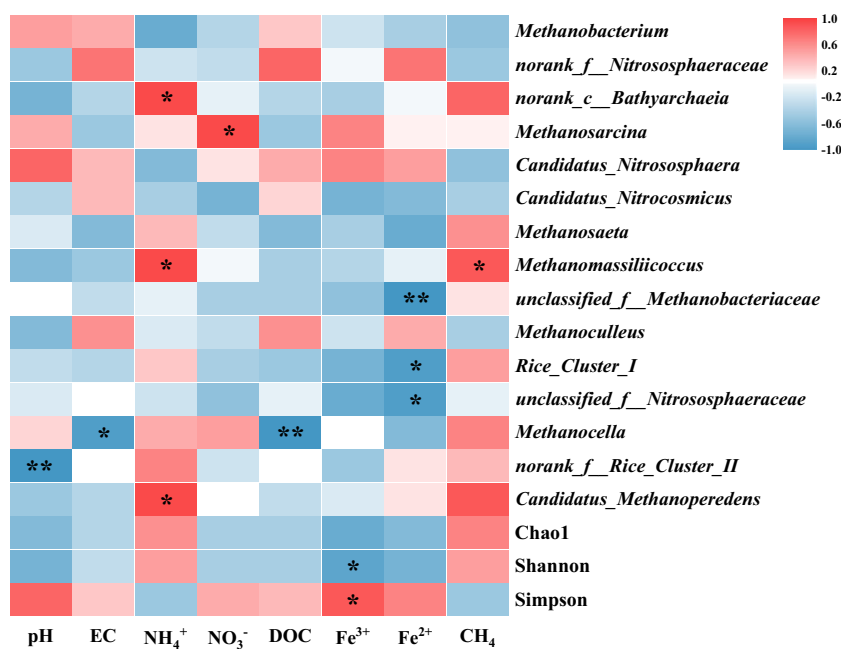


Fig. 6 Heatmap of archaeal at genus level (top 15) and environmental variables based on Pearson’s correlation. * $P < 0.05$; ** $P < 0.01$



habitat, soil characteristics, and microbial community (Alves et al. 2022; Luo et al. 2022; Zhang et al. 2023). It is worth noting that a low concentration of salinity (1%) did not have a significant effect on CH₄ emissions in H2 soil, which is consistent with the results in mangrove wetlands (Konnerup et al. 2014). In H1 soil, the four salinity treatments all significantly inhibited CH₄ emissions. This may be related to lower microbial community richness and

diversity in H1 soils (Table 1). In this study, CH₄ emission was significantly correlated with soil DOC and NH₄⁺ contents ($P < 0.01$). This is because CH₄ emission is controlled by substrate availability (Yuan et al. 2018a). In addition, CH₄ flux was mainly produced by methanogenic archaea and consumed by methanotrophs. The metabolic activity of these microorganisms is an important factor affecting CH₄ emission (Lai 2009).

The effect of salinity on archaeal community

The alpha diversity of methanogenic archaea decreased significantly with the increase in salinity (Zhang et al. 2019). Similarly, salinity reduced the richness and diversity of soil archaeal communities in this study. Characterization of archaeal communities using high-throughput analysis revealed that elevated salinity significantly reduced the relative abundance of *Methanosarciniales* and *Bathyarchaeia*, which indicated that these two methanogens were less resistant to high salinity. They were found and dominated in paddy soils and marine sediments (Yuan et al. 2018b; Romano et al. 2021).

The relative abundance of *Methanobacterium* belonging to the order of *Methanobacteriales* increased under the salinity treatment. *Methanobacterium* is a salt-tolerant methanogen that is found to live in estuarine wetlands even in areas of high salinity (Mori and Harayama 2011; Chen et al. 2020b). The genus of *Methanosarcina* can use H_2/CO_2 , acetic acid, and methyl substances as substrates to produce CH_4 (Youngblut et al. 2015; Lyu et al. 2018). The inhibitory response of this genus to salinity was most evident in H1 soil. However, the genus of *Methanosaeta* belonging to the same order of *Methanosarciniales* was not detected in H1 soil. In H2 and H3 soils, salinity decreased the relative abundance of *Methanosaeta*. Different from *Methanosarcina*, *Methanosaeta* can only use acetate to produce CH_4 (Mori et al. 2012). Previous research results have shown that the relative abundance of *Bathyarchaeia* was positively correlated with CH_4 emissions (Cui et al. 2019). The relative abundance of *Bathyarchaeia* in H2 soil (18.53–42.31%) was higher than that in H3 soil (7.68–13.44%) and H1 soil (1.25–3.52%). This could partly explain why CH_4 emissions were higher in H2 soil than those in H3 and H1 soils. *Bathyarchaeia* is widespread in deep-sea sediments and peatlands (Cui et al. 2019; L. Bräuer et al. 2020). However, high salinity inhibits the growth of *Bathyarchaeia* (Kallistova et al. 2020). The same result was obtained in this study. This may be because salinity affects the energy costs (associated with osmoregulation) and metabolic pathways of *Bathyarchaeia*, acting as an evolutionary barrier leading to the diversification of *Bathyarchaeota* (Fillol et al. 2016). *Methanomassiliicoccus* is a methylotrophic methanogen that is widely distributed in rice fields and wetlands (Söllinger et al. 2016; Lyu et al. 2018; Lu et al. 2022). In the present study, *Methanomassiliicoccus* was significantly positively correlated with CH_4 emissions ($P < 0.05$), which was consistent with the results in rice fields in Hunan Province, China (Lu et al. 2022). However, there were inconsistent results in other regions (Jiang et al. 2022; Luo et al. 2022).

The relationship between archaea and soil characteristics

In this study, archaeal community characteristics were affected by soil pH, DOC, NH_4^+ , NO_3^- , and Fe^{2+} concentrations. In general, a slightly alkaline environment is suitable for the growth of methane archaea (Malyan et al. 2016). The relative abundance of *Methanobacteriales* was positively correlated with pH, which was consistent with studies at pH 9.0 and 9.5 (Rao et al. 2018). However, *Rice_Cluster_II*, which prefers low hydrogen, was significantly negatively correlated with pH ($P < 0.01$). *Rice_Cluster_II* is widely present in acidic rice fields, and the change of pH could affect its metabolic pathways (Luo et al. 2022). In this study, *Methanocella* was significantly negatively correlated with DOC concentration and some other methanogenic archaea had weaker negative correlations with DOC, which might be due to the development of methanogenic archaea in tight association with the consumption of DOC. After the incubation, soil DOC content decreased by 20.87–97.71%. In addition, DOC can promote the utilization of CH_4 by methanotrophs (Lew and Glińska-Lewczuk 2018). The same result was also found in rice fields (Luo et al. 2022). NH_4^+ was significantly positively correlated with *Bathyarchaeia*, *Methanomassiliicoccus*, and *Candidatus_Methanoperedens* ($P < 0.05$). Among them, *Candidatus_Methanoperedens* was only detected in H2 soil. This genus is an anaerobic methanotrophic archaeon that oxidizes CH_4 to CO_2 and reduces NO_3^- to N_2 and NH_4^+ (Ettwig et al. 2016; Chen et al. 2021). In this study, Fe^{3+} decreased the archaeal community diversity. Besides, Fe^{2+} concentration was significantly negatively correlated with the relative abundance of *Methanobacteriaceae* and *Rice_Cluster_II* ($P < 0.05$). This revealed a correlation between iron and CH_4 emissions. Under anaerobic conditions, iron can promote decomposition of soil organic matter (Chen et al. 2020a). Methanogenic archaea can anaerobically degrade microscopic organic matter into CH_4 (Cai et al. 2019).

Conclusion

High salinity (5%) significantly suppressed CH_4 emission, reducing by 97.82% (H1), 89.20% (H2), and 87.97% (H3), respectively. Low salinity (1%) showed a significant inhibitory effect on CH_4 emission in H1 and H3 soils, but had no significant effect in H2 soils. In addition, salinity decreased the relative abundance of methanogenic archaea and changed the community structure. Specifically, salinity had a promoting effect on *Methanobacterium*, while an inhibitory effect on *Bathyarchaeia* and *Methanosaeta*. The

response of *Methanosarcina* to salinity was different in the three habitat soils. Soil pH, EC, DOC, and NH_4^+ concentrations were significantly correlated with the characteristics of the methanogenic archaeal community, thereby affecting CH_4 emissions.

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Data availability All data are mentioned in the body of manuscript, tables, and figure.

Declarations

Ethical approval Not applicable.

Consent to participate Not applicable.

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References

- Alves KJ, Pylro VS, Nakayama CR, Vital VG, Taketani RG, Santos DG, Rodrigues JLM, Tsai SM, Andreote FD (2022) Methanogenic communities and methane emissions from enrichments of Brazilian Amazonia soils under land-use change. *Microbiol Res* 265:127178. <https://doi.org/10.1016/j.micres.2022.127178>
- Borrel G, Parisot N, Harris H, Peyretailade E, Gaci N, Tottey W, Bardot O, Raymann K, Gribaldo S, Peyret P (2014) Comparative genomics highlights the unique biology of Methanomassiliicoccales, a Thermoplasmatales-related seventh order of methanogenic archaea that encodes pyrrolysine. *BMC genomics* 15(1):1–24. <https://doi.org/10.1186/1471-2164-15-679>
- Bräuer SL, Basiliko N, Siljanen HMP, Zinder SH (2020) Methanogenic archaea in peatlands. *FEMS Microbiol Lett* 367(20):fnaa172. <https://doi.org/10.1093/femsle/fnaa172>
- Bridgman SD, Cadillo Quiroz H, Keller JK, Zhuang Q (2013) Methane emissions from wetlands: biogeochemical, microbial, and modeling perspectives from local to global scales. *Glob Change Biol* 19(5):1325–1346. <https://doi.org/10.1111/gcb.12131>
- Cai P, Ning Z, Zhang N, Zhang M, Guo C, Niu M, Shi J (2019) Insights into biodegradation related metabolism in an abnormally low dissolved inorganic carbon (DIC) petroleum-contaminated aquifer by metagenomics analysis. *Microorganisms* 7(10):412. <https://doi.org/10.3390/microorganisms7100412>
- Chambers LG, Guevara R, Boyer JN, Troxler TG, Davis SE (2016) Effects of salinity and inundation on microbial community structure and function in a mangrove peat soil. *Wetlands* 36:361–371. <https://doi.org/10.1007/s13157-016-0745-8>
- Chambers LG, Reddy KR, Osborne TZ (2011) Short-term response of carbon cycling to salinity pulses in a freshwater wetland. *Soil Sci Soc Am J* 75(5):2000–2007. <https://doi.org/10.2136/sssaj2011.0026>
- Chen C, Hall SJ, Coward E, Thompson A (2020a) Iron-mediated organic matter decomposition in humid soils can counteract protection. *Nat Commun* 11(1):2255. <https://doi.org/10.1038/s41467-020-16071-5>
- Chen F, Zheng Y, Hou L, Niu Y, Gao D, An Z, Zhou J, Yin G, Dong H, Han P (2021) Microbial abundance and activity of nitrite/nitrate-dependent anaerobic methane oxidizers in estuarine and intertidal wetlands: heterogeneity and driving factors. *Water Res* 190:116737. <https://doi.org/10.1016/j.watres.2020.116737>
- Chen L, Li L, Zhang S, Zhang W, Xue K, Wang Y, Dong X (2022) Anaerobic methane oxidation linked to Fe(III) reduction in a Candidatus Methanoperedens-enriched consortium from the cold Zoige wetland at Tibetan Plateau. *Environ Microbiol* 24(2):614–625. <https://doi.org/10.1111/1462-2920.15848>
- Chen S, Wang P, Liu H, Xie W, Wan XS, Kao SJ, Phelps TJ, Zhang C (2020b) Population dynamics of methanogens and methanotrophs along the salinity gradient in Pearl River Estuary: implications for methane metabolism. *Appl Microbiol Biotechnol* 104:1331–1346. <https://doi.org/10.1007/s00253-019-10221-6>
- Chen S, Zhou Y, Chen Y, Gu J (2018) fastp: an ultra-fast all-in-one FASTQ preprocessor. *Bioinformatics* 34(17):i884–i890. <https://doi.org/10.1093/bioinformatics/bty560>
- Cui H, Su X, Chen F, Holland M, Yang S, Liang J, Su P, Dong H, Hou W (2019) Microbial diversity of two cold seep systems in gas hydrate-bearing sediments in the South China Sea. *Mar Environ Res* 144:230–239. <https://doi.org/10.1016/j.marenvres.2019.01.009>
- Dang C, Morrissey EM, Neubauer SC, Franklin RB (2019) Novel microbial community composition and carbon biogeochemistry emerge over time following saltwater intrusion in wetlands. *Glob Change Biol* 25(2):549–561. <https://doi.org/10.1111/gcb.14486>
- Duan B, Cai T, Man X, Xiao R, Gao M, Ge Z, Mencuccini M (2022) Different variations in soil CO_2 , CH_4 , and N_2O fluxes and their responses to edaphic factors along a boreal secondary forest successional trajectory. *Sci Total Environ* 838:155983. <https://doi.org/10.1016/j.scitotenv.2022.155983>
- Edgar RC (2013) UPARSE: highly accurate OTU sequences from microbial amplicon reads. *Nat Methods* 10(10):996–998. <https://doi.org/10.1038/nmeth.2604>
- Ettwig KF, Zhu B, Speth D, Keltjens JT, Jetten MSM, Kartal B (2016) Archaea catalyze iron-dependent anaerobic oxidation of methane. *Proc Natl Acad Sci* 113(45):12792–12796. <https://doi.org/10.1073/pnas.1609534113>
- Fan L, Schneider D, Dippold MA, Poehlein A, Wu W, Gui H, Ge T, Wu J, Thiel V, Kuzyakov Y (2021) Active metabolic pathways of anaerobic methane oxidation in paddy soils. *Soil Biol Biochem* 156:108215. <https://doi.org/10.1016/j.soilbio.2021.108215>
- Feng L, Zhang Z, Yang G, Wu G, Yang Q, Chen Q (2023) Microbial communities and sediment nitrogen cycle in a coastal eutrophic lake with salinity and nutrients shifted by seawater intrusion. *Environ Res* 225:115590. <https://doi.org/10.1016/j.envres.2023.115590>
- Filloil M, Auguet JC, Casamayor EO, Borrego CM (2016) Insights in the ecology and evolutionary history of the Miscellaneous Crenarchaeotic Group lineage. *ISME J* 10(3):665–677. <https://doi.org/10.1038/ismej.2015.143>
- Gao D, Liu F, Xie Y, Liang H (2018) Temporal and spatial distribution of ammonia-oxidizing organisms of two types of wetlands in Northeast China. *Appl Microbiol Biotechnol* 102(16):7195–7205. <https://doi.org/10.1007/s00253-018-9152-9>
- Gütlein A, Gerschlauser F, Kikoti I, Kiese R (2018) Impacts of climate and land use on N_2O and CH_4 fluxes from tropical ecosystems in the Mt. Kilimanjaro region, Tanzania. *Glob Change Biol* 24(3):1239–1255. <https://doi.org/10.1111/gcb.13944>

- Haese RR, Wallmann K, Dahmke A, Kretzmann U, Müller PJ, Schulz HD (1997) Iron species determination to investigate early diagenetic reactivity in marine sediments. *Geochim Cosmochim Acta* 61(1):63–72. [https://doi.org/10.1016/S0016-7037\(96\)00312-2](https://doi.org/10.1016/S0016-7037(96)00312-2)
- Herbert ER, Boon P, Burgin AJ, Neubauer SC, Franklin RB, Ardón M, Hopfensperger KN, Lamers LPM, Gell P (2015) A global perspective on wetland salinization: ecological consequences of a growing threat to freshwater wetlands. *Ecosphere* 6(10):1–43. <https://doi.org/10.1890/es14-00534.1>
- Hofmann K, Praeg N, Mutschlechner M, Wagner AO, Illmer P (2016) Abundance and potential metabolic activity of methanogens in well-aerated forest and grassland soils of an alpine region. *FEMS Microbiol Ecol* 92(2):fiv171. <https://doi.org/10.1093/femsec/fiv171>
- IPCC (2021) Summary for Policymakers. In: Masson-Delmotte V, Zhai P, Pirani A, Connors SL (eds) *The physical science basis. Contribution of working group I to the sixth assessment report of the intergovernmental panel on climate change*. Cambridge University Press. <https://www.ipcc.ch/report/sixth-assessment-report-working-group-i/>. Accessed 20 March 2023
- Jeppesen E, Beklioglu M, Özkan K, Akyürek Z (2020) Salinization increase due to climate change will have substantial negative effects on inland waters: a call for multifaceted research at the local and global scale. *The Innovation* 1(2):100030. <https://doi.org/10.1016/j.xinn.2020.100030>
- Jiang M, Xu P, Wu L, Zhao J, Wu H, Lin S, Yang T, Tu J, Hu R (2022) Methane emission, methanogenic and methanotrophic communities during rice-growing seasons differ in diversified rice rotation systems. *Sci Total Environ* 842:156781. <https://doi.org/10.1016/j.scitotenv.2022.156781>
- Kallistova A, Merkel A, Kanapatskiy T, Boltanskaya Y, Tarnovetskii I, Perevalova A, Kevbrin V, Samylina O, Pimenov N (2020) Methanogenesis in the Lake Elton saline aquatic system. *Extremophiles* 24:657–672. <https://doi.org/10.1007/s00792-020-01185-x>
- Kirschke S, Bousquet P, Ciais P, Saunoy M, Canadell JG, Dlugokencky EJ, Bergamaschi P, Bergmann D, Blake DR, Bruhwiler L (2013) Three decades of global methane sources and sinks. *Nat Geosci* 6(10):813–823. <https://doi.org/10.1038/ngeo1955>
- Kong D, Li S, Jin Y, Wu S, Chen J, Hu T, Wang H, Liu S, Zou J (2019) Linking methane emissions to methanogenic and methanotrophic communities under different fertilization strategies in rice paddies. *Geoderma* 347:233–243. <https://doi.org/10.1016/j.geoderma.2019.04.008>
- Konnerup D, Betancourt Portela JM, Villamil C, Parra JP (2014) Nitrous oxide and methane emissions from the restored mangrove ecosystem of the Ciénaga Grande de Santa Marta, Colombia. *Estuar Coastal Shelf Sci* 140:43–51. <https://doi.org/10.1016/j.ecss.2014.01.006>
- Krauss KW, Whitbeck JL (2012) Soil greenhouse gas fluxes during wetland forest retreat along the lower Savannah River, Georgia (USA). *Wetlands* 32:73–81. <https://doi.org/10.1007/s13157-011-0246-8>
- Lai DYF (2009) Methane dynamics in northern peatlands: a review. *Pedosphere* 19(4):409–421. [https://doi.org/10.1016/s1002-0160\(09\)00003-4](https://doi.org/10.1016/s1002-0160(09)00003-4)
- Lan X, KW Thoning, EJ Dlugokencky (2022) Trends in globally-averaged CH₄, N₂O, and SF₆ determined from NOAA Global Monitoring Laboratory measurements. Version 2023-03. Global Monitoring Laboratory. <https://doi.org/10.15138/P8XG-AA10>
- Lew S, Glińska Lewczuk K (2018) Environmental controls on the abundance of methanotrophs and methanogens in peat bog lakes. *Sci Total Environ* 645:1201–1211. <https://doi.org/10.1016/j.scitotenv.2018.07.141>
- Liu C, Li H, Zhang Y, Si D, Chen Q (2016) Evolution of microbial community along with increasing solid concentration during high-solids anaerobic digestion of sewage sludge. *Bioresour Technol* 216:87–94. <https://doi.org/10.1016/j.biortech.2016.05.048>
- Liu F, Zhang Y, Liang H, Gao D (2019) Long-term harvesting of reeds affects greenhouse gas emissions and microbial functional genes in alkaline wetlands. *Water Res* 164:114936. <https://doi.org/10.1016/j.watres.2019.114936>
- Lu Y, Liu Q, Fu L, Hu Y, Zhong L, Zhang S, Liu Q, Xie Q (2022) The effect of modified biochar on methane emission and succession of methanogenic archaeal community in paddy soil. *Chemosphere* 304:135288. <https://doi.org/10.1016/j.chemosphere.2022.135288>
- Luo D, Li Y, Yao H, Chapman SJ (2022) Effects of different carbon sources on methane production and the methanogenic communities in iron rich flooded paddy soil. *Sci Total Environ* 823:153636. <https://doi.org/10.1016/j.scitotenv.2022.153636>
- Lyu Z, Shao N, Akinyemi T, Whitman WB (2018) Methanogenesis. *Curr Biol* 28(13):R727–R732. <https://doi.org/10.1016/j.cub.2018.05.021>
- Magoč T, Salzberg SL (2011) FLASH: fast length adjustment of short reads to improve genome assemblies. *Bioinformatics* 27(21):2957–2963. <https://doi.org/10.1093/bioinformatics/btr507>
- Malyan SK, Bhatia A, Kumar A, Gupta DK, Singh R, Kumar SS, Tomer R, Kumar O, Jain N (2016) Methane production, oxidation and mitigation: a mechanistic understanding and comprehensive evaluation of influencing factors. *Sci Total Environ* 572:874–896. <https://doi.org/10.1016/j.scitotenv.2016.07.182>
- Moberly JG, Bernards MT, Waynant KV (2018) Key features and updates for origin 2018. *J Cheminform* 10:1–2. <https://doi.org/10.1186/s13321-018-0259-x>
- Morgan GA, Barrett KC, Leech NL, Gloeckner GW (2019) *IBM SPSS for introductory statistics: use and interpretation*. Routledge Press, UK
- Mori K, Harayama S (2011) *Methanobacterium petrolearium* sp. nov. and *Methanobacterium ferruginis* sp. nov., mesophilic methanogens isolated from salty environments. *International Journal of Systematic and Evolutionary Microbiology* 61(1):138–143. <https://doi.org/10.1099/ijs.0.022723-0>
- Mori K, Iino T, Suzuki KI, Yamaguchi K, Kamagata Y (2012) Acetate and NaCl-requiring methanogen “*Methanosaeta pelagica*” sp. nov., isolated from marine tidal flat sediment. *Appl Environ Microb* 78(9):3416–3423. <https://doi.org/10.1128/aem.07484-11>
- Nakagawa F, Yoshida N, Nojiri Y, Makarov V (2002) Production of methane from allasses in eastern Siberia: implications from its ¹⁴C and stable isotopic compositions. *Glob Biogeochem Cycles* 16(3):14–11. <https://doi.org/10.1029/2000gb001384>
- Pattnaik P, Mishra SR, Bharati K, Mohanty SR, Sethunathan N, Adhya TK (2000) Influence of salinity on methanogenesis and associated microflora in tropical rice soils. *Microbiol Res* 155(3):215–220. [https://doi.org/10.1016/s0944-5013\(00\)80035-x](https://doi.org/10.1016/s0944-5013(00)80035-x)
- Qiu S, Zhang X, Xia W, Li Z, Wang L, Chen Z, Ge S (2023) Effect of extreme pH conditions on methanogenesis: methanogen metabolism and community structure. *Sci Total Environ* 877:162702. <https://doi.org/10.1016/j.scitotenv.2023.162702>
- Quast C, Pruesse E, Yilmaz P, Gerken J, Schweer T, Yarza P, Peplies J, Glöckner FO (2012) The SILVA ribosomal RNA gene database project: improved data processing and web-based tools. *Nucleic Acids Res* 41(D1):D590–D596. <https://doi.org/10.1093/nar/gks1219>
- Rao Y, Wan J, Liu Y, Angelidaki I, Zhang S, Zhang Y, Luo G (2018) A novel process for volatile fatty acids production from syngas by integrating with mesophilic alkaline fermentation of waste activated sludge. *Water Res* 139:372–380. <https://doi.org/10.1016/j.watres.2018.04.026>
- Rath KM, Fierer N, Murphy DV, Rousk J (2019) Linking bacterial community composition to soil salinity along environmental gradients. *ISME J* 13(3):836–846. <https://doi.org/10.1038/s41396-018-0313-8>
- Romano RG, Bendia AG, Moreira JCF, Franco DC, Signori CN, Yu T, Wang F, Jovane L, Pellizari VH (2021) Bathyarchaeia occurrence in rich methane sediments from a Brazilian ría. *Estuar Coastal Shelf Sci* 263:107631. <https://doi.org/10.1016/j.ecss.2021.107631>

- Šmilauer P, Lepš J (2014) *Multivariate analysis of ecological data using CANOCO 5*. Cambridge university press, UK
- Söllinger A, Schwab C, Weinmaier T, Loy A, Tveit AT, Schleper C, Urlich T (2016) Phylogenetic and genomic analysis of Methanomassiliococcales in wetlands and animal intestinal tracts reveals clade-specific habitat preferences. *FEMS Microbiol Ecol* 92(1):fiv149. <https://doi.org/10.1093/femsec/fiv149>
- Ström L, Ekberg A, Mastepanov M, Røjle Christensen T (2003) The effect of vascular plants on carbon turnover and methane emissions from a tundra wetland. *Glob Change Biol* 9(8):1185–1192. <https://doi.org/10.1046/j.1365-2486.2003.00655.x>
- Sun Z, Jiang H, Wang L, Mou X, Sun W (2013) Seasonal and spatial variations of methane emissions from coastal marshes in the northern Yellow River estuary, China. *Plant Soil* 369:317–333. <https://doi.org/10.1007/s11104-012-1564-1>
- Venturini AM, Dias NMS, Gontijo JB, Yoshiura CA, Paula FS, Meyer KM, Nakamura FM, da França AG, Borges CD, Barlow J (2022) Increased soil moisture intensifies the impacts of forest-to-pasture conversion on methane emissions and methane-cycling communities in the Eastern Amazon. *Environ Res* 212:113139. <https://doi.org/10.1016/j.envres.2022.113139>
- Wallmann K, Hennies K, König I, Petersen Wand Knauth HD (1993) New procedure for determining reactive Fe(III) and Fe(II) minerals in sediments. *Limnol Oceanogr* 38(8):1803–1812. <https://doi.org/10.4319/lo.1993.38.8.1803>
- Wang J, Cai C, Li Y, Hua M, Wang J, Yang H, Zheng P, Hu B (2018) Denitrifying anaerobic methane oxidation: a previously overlooked methane sink in intertidal zone. *Environ Sci Technol* 53(1):203–212. <https://doi.org/10.1021/acs.est.8b05742>
- Wang W, Liang H, Li F, Su H, Li H, Gao D (2023) Water level of inland saline wetlands with implications for CO₂ and CH₄ fluxes during the autumn freeze–thaw period in Northeast China. *Environ Sci Pollut Res* 30(17):50125–50133. <https://doi.org/10.1007/s11356-023-25862-4>
- Wang Y, Hu Z, Shen L, Liu C, Islam ARMT, Wu Z, Dang H, Chen S (2021) The process of methanogenesis in paddy fields under different elevated CO₂ concentrations. *Sci Total Environ* 773:145629. <https://doi.org/10.1016/j.scitotenv.2021.145629>
- Weston NB, Vile MA, Neubauer SC, Velinsky DJ (2011) Accelerated microbial organic matter mineralization following salt-water intrusion into tidal freshwater marsh soils. *Biogeochemistry* 102:135–151. <https://doi.org/10.1007/s10533-010-9427-4>
- Wilson BJ, Mortazavi B, Kiene RP (2015) Spatial and temporal variability in carbon dioxide and methane exchange at three coastal marshes along a salinity gradient in a northern Gulf of Mexico estuary. *Biogeochemistry* 123:329–347. <https://doi.org/10.1007/s10533-015-0085-4>
- Wu J, Wang M, Li P, Shen L, Ma M, Xu B, Zhang S, Sha C, Ye C, Xiong L (2022) Effects of pig manure and its organic fertilizer application on archaea and methane emission in paddy fields. *Land* 11(4):499. <https://doi.org/10.3390/land11040499>
- Youngblut ND, Wirth JS, Henriksen JR, Smith M, Simon H, Metcalf WW, Whitaker RJ (2015) Genomic and phenotypic differentiation among *Methanosarcina mazei* populations from Columbia River sediment. *ISME J* 9(10):2191–2205. <https://doi.org/10.1038/ismej.2015.31>
- Yuan J, Yuan Y, Zhu Y, Cao L (2018a) Effects of different fertilizers on methane emissions and methanogenic community structures in paddy rhizosphere soil. *Sci Total Environ* 627:770–781. <https://doi.org/10.1016/j.scitotenv.2018.01.233>
- Yuan Q, Hernández M, Dumont MG, Rui J, Scavino AF, Conrad R (2018b) Soil bacterial community mediates the effect of plant material on methanogenic decomposition of soil organic matter. *Soil Biol Biochem* 116:99–109. <https://doi.org/10.1016/j.soilbio.2017.10.004>
- Zhang K, Shi Y, Cui X, Yue P, Li K, Liu X, Tripathi Binu M, Chu H (2019) Salinity is a key determinant for soil microbial communities in a desert ecosystem. *mSystems* 4(1):e00225. <https://doi.org/10.1128/mSystems.00225-18>
- Zhang W, Sheng R, Zhang M, Xiong G, Hou H, Li S, Wei W (2018) Effects of continuous manure application on methanogenic and methanotrophic communities and methane production potentials in rice paddy soil. *Agric Ecosyst Environ* 258:121–128. <https://doi.org/10.1016/j.agee.2018.02.018>
- Zhang Z, Yang Z, Yue H, Xiao M, Ge T, Li Y, Yu Y, Yao H (2023) Discrepant impact of polyethylene microplastics on methane emissions from different paddy soils. *Appl Soil Ecol* 181:104650. <https://doi.org/10.1016/j.apsoil.2022.104650>

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