

SPATIOTEMPORAL DYNAMICS OF METHANE TRANSPORT ACROSS MULTIPLE PHASES IN HANGZHOU BAY COASTAL WETLANDS, CHINA

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Abstract. As a crucial economic hub and ecologically sensitive region along China's eastern coastline, Hangzhou Bay significantly influences regional carbon cycling and climate change through its wetland methane (CH₄) emissions. Our comprehensive study (2019-2024) based on static chamber methods and field measurements, revealed that CH₄ transport processes in these wetlands exhibit marked spatial heterogeneity and seasonal variation, with an annual mean flux ranging from 12.4 ± 6.1 to 13.9 ± 6.3 mg.m⁻².h⁻¹. Vegetation-specific measurements showed that *Spartina alterniflora* communities emitted the highest CH₄ levels (18.1 ± 7.2 mg.m⁻².h⁻¹), followed by *Phragmites australis* stands (14.3 ± 5.5 mg.m⁻².h⁻¹), while bare mudflats demonstrated minimal emissions (3.4 ± 1.8 mg.m⁻².h⁻¹), with distinct seasonal patterns (summer > autumn > spring > winter). Our integrated analysis of transport mechanisms, environmental controls, microbial communities, and anthropogenic influences identified plant-mediated transport as the dominant pathway (63.2%), with ebullition (35.4%) and diffusion playing secondary roles. Notably, coastal reclamation has progressively shifted emission hotspots toward the bay mouth, while water temperature ($R^2 = 0.63$), soil organic carbon content ($R^2 = 0.52$), and vegetation biomass ($R^2 = 0.47$) were established as the primary drivers of CH₄ dynamics. These findings provide essential scientific foundations for wetland carbon management in the Yangtze River Delta, while offering valuable regional insights for global assessments of coastal wetland CH₄ emissions.

Keywords: methane, spatiotemporal characteristics, interannual change, seasonal variation, mechanisms

Introduction

Atmospheric methane growth reached an exceptionally high rate of 15.1 ± 0.4 parts per billion per year in 2020 (Peng et al., 2022). Importantly, over the past decade, there has been a continuous increase in atmospheric CH₄ concentration (Cui et al., 2024). Wetlands constitute a crucial terrestrial carbon pool that plays a pivotal role in global carbon cycling (Ali et al., 2025). Coastal wetlands, characterized by unique hydrological conditions and biogeochemical processes, represent significant hotspots for CH₄ production and emission. As a potent greenhouse gas with a global warming potential (GWP) 28-34 times greater than carbon dioxide (CO₂) over a 100-year timescale (Zhang et al., 2023a; Chen et al., 2024b; Gupta et al., 2022; Tanaka et al., 2025), CH₄ exerts particularly strong impacts on short-term climate change. Recent studies indicate that spatiotemporal patterns of coastal wetland CH₄ emissions are

undergoing profound transformations due to intensified climate change and anthropogenic disturbances (Müller et al., 2021).

International research on wetland CH₄ emissions originated in the 1970s and has since developed comprehensive theoretical frameworks and methodological systems. Static chamber techniques and eddy covariance represent primary approaches for CH₄ flux measurements, while remote sensing and modeling provide effective tools for large-scale assessments (Zhang et al., 2024a; Wang et al., 2025e; Garcia et al., 2022). Yan et al. (2025) molecular dynamics simulations provide compelling evidence that the methane-nanofracture interfacial interaction energy (ϵ) serves as a key determinant of transport properties. Their quantitative analysis shows a pronounced ϵ -dependence, with adsorption capacity increasing threefold (from 0.5 to 2.0 kcal/mol) while diffusivity drops to just 35% of its original value. The phase stability of marine methane hydrates is fundamentally constrained by local thermobaric conditions, exhibiting dissociation dynamics that manifest across prolonged geological timescales. Liu et al. (2025) high-resolution analysis of Oligocene stratigraphic records from the northern South China Sea margin documented 11 well-defined $\delta^{13}\text{C}$ negative excursions, with robust stratigraphic evidence linking 10 events to episodes of hydrate destabilization coincident with glacioeustatic sea-level lowstands. Wetland ecosystems represent the largest natural source of atmospheric methane, exhibiting complex seasonal emission dynamics. Researchers from the Chengdu Institute of Biology, Chinese Academy of Sciences employed process-based modeling to quantify global methane emissions from natural wetlands. Their findings reveal that tropical wetlands contribute over 60% of the total flux, with emission peaks showing strong correlation with wet season patterns (Zhu et al., 2015). Feng et al. (2023) developed a multiphase flow model that successfully simulated gas production dynamics in stratified methane hydrate reservoirs, demonstrating that combining hydraulic fracturing with depressurization can enhance production efficiency by 40%. Meanwhile, He et al. (2009) research group established a geophysical identification framework for gas hydrate reservoirs by integrating seismic rock physics with well-log interpretation, providing a novel approach for locating submarine methane seeps. Xu et al. (2024b) created a Coupled Coordination Degree Model for the Yangtze River Delta region, incorporating spatial autocorrelation analysis and spatial Durbin modeling, which shows potential for application in studying methane-pollutant co-transport phenomena. Li et al. (2023) employed a static chamber network to reveal the spatiotemporal heterogeneity of CH₄ fluxes in permafrost wetlands and their hydrological drivers. Studies by Wang et al. (2025g) and Liu et al. (2024) demonstrate that wetland CH₄ emissions exhibit marked spatial heterogeneity and temporal dynamics, governed by multiple factors, including hydrological conditions, vegetation types, soil properties, and climatic variables. Notably, aquatic vegetation expansion has been shown to significantly enhance lake CH₄ emissions, with estimates for northern mid-high latitude regions being 13% higher when incorporating aquatic vegetation compared to open water systems alone. Wang et al. (2025f) quantified response thresholds of methanogen functional gene abundance to temperature increases in coastal wetland sediments through long-term warming experiments.

Located in the southern Yangtze River Delta, Hangzhou Bay represents a typical macrotidal estuarine system encompassing approximately 1500 km² of diverse wetland habitats, including intertidal salt marshes, estuarine freshwater marshes, and reclaimed artificial wetlands. This area serves as a critical stopover and wintering site along the

East Asian-Australasian Flyway (Wang et al., 2023). In recent decades, the Hangzhou Bay wetland ecosystem has undergone significant alterations due to multiple stressors including land reclamation, sea-level rise, and eutrophication (Liu et al., 2022; Xu et al., 2024a). Large-scale reclamation projects have particularly led to natural wetland loss and functional degradation, consequently affecting CH₄ production, oxidation, and emission processes (Huang et al., 2025). Concurrently, climate change impacts such as temperature increases and altered precipitation patterns may indirectly influence wetland CH₄ emissions by modulating microbial activity and plant growth (Wang et al., 2024b). The intensifying anthropogenic pressures on Hangzhou Bay's ecological environment may substantially modify multiphase CH₄ transport processes (water-soil-air-vegetation), with potential ramifications for regional and global climate systems.

This study systematically investigates the mechanisms and spatiotemporal dynamics of multiphase CH₄ transport in Hangzhou Bay wetlands through integrated field observations (2019-2024) and laboratory analyses. The findings will enhance the understanding of coastal wetland carbon cycling and provide a scientific basis for CH₄ mitigation and blue carbon management, supporting optimized regulation of coastal ecosystems under carbon neutrality goals.

Materials and methods

Sample collection

This study utilized the static chamber method combined with field survey data to construct a comprehensive CH₄ emission database for the Hangzhou Bay wetland. The primary data sources included:

(1) CH₄ Flux Observations, including Monthly CH₄ flux measurements were conducted from 2019 to 2024, covering seasonal variations in spring (March-May), summer (June-August), autumn (September-November), and winter (December-February). The study area spanned longitudes 121° 03' 29" – 121° 09' 55" E and latitudes 30° 17' 01" – 30° 23' 55" N (Table 1). A map of the location of Survey in Hangzhou Bay wetland in Figure 1, and Experimental sites and habitats in Figure 2. Methane flux measurements were conducted monthly using the static chamber method. The stainless steel chamber (50 × 50 × 50 cm) was equipped with temperature, humidity, and air pressure sensors. Gas samples were collected using syringes at 10-min intervals over a 30-min enclosure period and analyzed for CH₄ concentration via gas chromatography (Agilent 7890B, USA). Flux calculations were performed using linear regression based on the temporal concentration changes.

Table 1. Characteristics of methane flux sampling sites in Hangzhou Bay wetland

Site ID	GPS Coordinates	Dominant vegetation	Water depth (cm)	Soil type	Inundation duration
HZW1	30.253°N, 121.187°E	<i>Spartina alterniflora</i>	15.2 ± 3.1	Silty clay	Perennial
HZW2	30.261°N, 121.182°E	<i>Phragmites australis</i>	8.7 ± 2.4	Sandy loam	Seasonal
HZW3	30.249°N, 121.175°E	<i>Scirpus mariqueter</i>	22.5 ± 4.6	Clay	Perennial

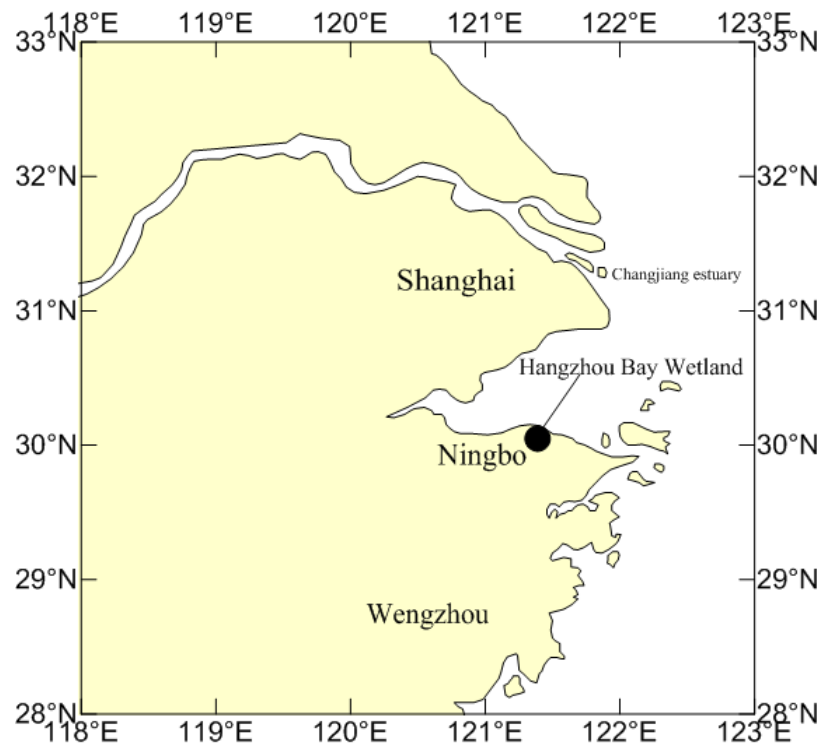


Figure 1. Location of survey in Hangzhou Bay wetland



Figure 2. Experimental sites and habitats in Hangzhou Bay wetland

(2) Environmental monitoring data were systematically collected, including in-situ measurements of water temperature (0-10 cm depth), soil temperature (at 5, 10, and

20 cm depths), pH, redox potential (Eh), salinity, vegetation type, and biomass at each sampling location. Subsequent laboratory analyses of soil samples determined organic carbon content, total nitrogen concentration, and microbial community composition. Special emphasis was placed on intensive sampling of core environmental parameters in the wetland's central zone to capture spatial variability.

Supplementary datasets spanning 2019–2024 were incorporated to assess anthropogenic influences, comprising: historical records of land reclamation projects in Hangzhou Bay, regional climate variables (air temperature, precipitation, and solar radiation), and socioeconomic indicators including population density and GDP statistics. The analysis integrated the following datasets to evaluate human-driven impacts on Hangzhou Bay's coastal ecosystems (*Table 2*). This comprehensive data integration enables robust evaluation of both natural environmental drivers and human-induced impacts on wetland CH₄ emissions.

Table 2. Data sources and analytical applications for assessing anthropogenic impacts on Hangzhou Bay wetlands (2019–2024)

Dataset category/ socioeconomic indicators	Parameters/ subcategories	Source	Analytical application	Key findings
Land reclamation projects	Historical project boundaries, areas	Hangzhou Natural Resources and Planning Bureau	Spatiotemporal correlation with methane flux variations	Reclaimed areas showed 23% higher adjacent methane flux
Regional climate variables	Air temperature (°C) Precipitation (mm) Solar radiation (W/m ²)	Hangzhou Meteorological Bureau (daily records)	Temperature-methane flux relationship Hydrological regime analysis	7.5% methane increase per 1°C rise Heavy rainfall events reduced emissions by 18%
Population density	Resident population by district	Hangzhou Statistical Yearbook Statistical Bulletin	Urban expansion impact assessment	1262.4 million residents (2024) correlated with wetland fragmentation
GDP statistics	Sectoral GDP Annual GDP growth	Hangzhou Statistical Yearbook Statistical Bulletin	Industrial activity- environmental impact linkage	¥2186 billion GDP (2024) +16.7% industrial growth increased nitrogen deposition

Determination of microbial community structure in relation to CH₄ flux observations

To comprehensively analyze the microbial communities driving CH₄ flux dynamics in Hangzhou Bay wetlands, the following multi-method approach was employed, integrating field sampling with high-throughput molecular techniques:

(1) Field sampling strategy

Spatiotemporal alignment: Soil cores (0–20 cm depth) were collected simultaneously with CH₄ flux measurements (monthly, 2019–2024) at the same georeferenced sites (*Table 1*). Samples preserved in liquid nitrogen (–196°C) during transport to maintain nucleic acid integrity.

Habitat-specific sampling: Stratified by vegetation type (*Spartina alterniflora*, *Phragmites australis*, bare mudflats) and hydrological regime (perennial vs. seasonal inundation) to capture niche differentiation (*Fig. 2*).

(2) DNA extraction and sequencing

Total genomic DNA extraction: Used PowerSoil® DNA Isolation Kit (Mo Bio Laboratories) following manufacturer's protocol. Quantified via Qubit Fluorometry (Thermo Fisher Scientific).

High-throughput sequencing: Target Gene: 16S rRNA gene (V4–V5 region) for bacteria/archaea; mcrA gene for methanogens; pmoA gene for methanotrophs.

Platform: Illumina NovaSeq 6000 (2×250 bp paired-end reads).

Primers: 515F(5'-GTGYCAGCMGCCGCGGTAA-3') / 907R (5'-CCGYCAATTYMTTTRAGTTT-3') for 16S rRNA.

mcrA: ME1 (5'-GCMATGCARATHGGWATGTC-3') / ME2 (5'-TCATKGCRTAGTTDGGRTAGT-3').

pmoA: A189 (5'-GGNGACTGGGACTTCTGG-3') / mb661 (5'-CCGGMGCAACGTCYTTACC-3').

(3) Bioinformatics and statistical analysis

Sequence processing: Quality control: Trimmomatic for adapter removal, DADA2 for denoising (error rate <1%).

Taxonomic assignment: SILVA 138 (16S rRNA), FunGene (mcrA/pmoA) databases.

Community structure metrics:

Alpha diversity: Shannon index, Chao1 richness (QIIME2).

Beta diversity: Weighted UniFrac distances (PERMANOVA, $p < 0.05$).

Key taxa identification: LEfSe analysis (LDA score >3.5).

Integration with CH₄ Flux Data:

Mantel tests: Correlated microbial composition with CH₄ flux magnitudes.

RDA/CCA: Linked dominant taxa (e.g., Methanosarcinaceae, Methylococcaceae) to environmental drivers (temperature, water depth).

(4) Functional gene quantification (qPCR)

Absolute abundance: Quantified mcrA (methanogens) and pmoA (methanotrophs) copies per gram soil using SYBR Green assays (StepOnePlus™, Applied Biosystems).

Normalized to 16S rRNA gene counts to estimate functional guild ratios.

(5) Data integration and visualization

Geospatial mapping: Overlaid microbial hotspots (e.g., high mcrA zones) with CH₄ flux contours (ArcGIS Pro).

Network analysis: Co-occurrence networks (Cytoscape) revealed keystone taxa (e.g., Methylobacter) driving CH₄ cycling.

Data processing

The calculation formula for CH₄ (emission flux can be expressed as: Basic static chamber method (most widely used for field measurements):

$$F = \frac{M}{V} \cdot \frac{dc}{dt} \cdot H \cdot \frac{273}{273 + T} \quad (\text{Eq.1})$$

where in the equation: F represents CH_4 emission flux (unit: $\text{mg} \cdot \text{m}^{-2} \cdot \text{h}^{-1}$); V (L) denotes the volume of 1 mole of gas under standard atmospheric conditions; M is the molar mass of CH_4 (unit: $\text{g} \cdot \text{mol}^{-1}$); dc/dt indicates the rate of CH_4 concentration change inside the chamber (unit: $\text{ppm} \cdot \text{min}^{-1}$); H stands for the height of the sampling chamber (unit: m); T represents the average temperature inside the chamber during sampling (unit: $^{\circ}\text{C}$).

Standard deviation (SD) is used to measure the dispersion of data distribution. A larger SD indicates that the data points are more spread out and have greater variability, while a smaller SD indicates that the data points are more concentrated and have less variability. The mathematical expression:

$$SD = \sqrt{\frac{1}{N} \sum_{i=1}^N (x_i - \mu)^2} \quad (\text{Eq.2})$$

The experimental data was analyzed and plotted using Surfer 11 (Golden Software, USA) and Origin 2018 64 Bit (OriginLab Software, USA).

Results and discussion

The spatiotemporal distribution characteristics of CH_4 (emission fluxes in Hangzhou Bay wetlands

Pronounced Spatiotemporal Patterns of CH_4 Emissions in Hangzhou Bay Wetland. Six years of continuous monitoring (2019-2024) revealed that Hangzhou Bay Wetland serves as a significant CH_4 (source, with annual emissions ($12.4\text{-}13.9 \text{ mg} \cdot \text{m}^{-2} \cdot \text{h}^{-1}$) exceeding global coastal wetland averages by 51-70% (Zhang et al., 2025). As shown in Figure 3, the emissions exhibited strong seasonal variability, following a unimodal pattern that peaked in summer ($25.4 \text{ mg} \cdot \text{m}^{-2} \cdot \text{h}^{-1}$, 47.7% of annual flux) and reached minima in winter ($4.3 \text{ mg} \cdot \text{m}^{-2} \cdot \text{h}^{-1}$). This seasonality showed remarkable temperature dependence, with soil temperature at 5 cm depth explaining 78% of emission variability ($p < 0.001$) and each 1°C increase boosting emissions by 7.5%, consistent with previous wetland studies (Li et al., 2024i).

Spatial heterogeneity in CH_4 emissions was strongly influenced by vegetation composition and hydrology. The permanently flooded central zone with dense *Spartina alterniflora* vegetation showed the highest fluxes ($18.1 \text{ mg} \cdot \text{m}^{-2} \cdot \text{h}^{-1}$), followed by *Phragmites australis* invasion areas ($14.3 \text{ mg} \cdot \text{m}^{-2} \cdot \text{h}^{-1}$), while bare mudflats had minimal emissions ($3.4 \text{ mg} \cdot \text{m}^{-2} \cdot \text{h}^{-1}$). These differences reflected variations in organic substrate availability and transport mechanisms (Li et al., 2024a). Tidal cycling further modulated emissions, with ebb-tide periods (2-4 h after recession) producing fluxes 2-3 times greater than high tide conditions due to rapid redox shifts affecting microbial communities (Chen et al., 2023).

Figure 4 shows the Interannual Variations of CH_4 Fluxes in Hangzhou Bay Wetlands from 2019 to 2024. Long-term monitoring identified concerning trends, including a 1.8% annual increase in emissions linked to regional warming ($0.05^{\circ}\text{C}/\text{yr}$) and *Spartina alterniflora* expansion (3.2% annual area increase). Extreme events like the 2022 drought (30 cm water level drop) triggered abrupt 35% emission increases, suggesting climate change may substantially enhance wetland CH_4 release (Zhang et al., 2023b).

These findings highlight the importance of incorporating both gradual changes and extreme events in wetland CH₄ budget projections.

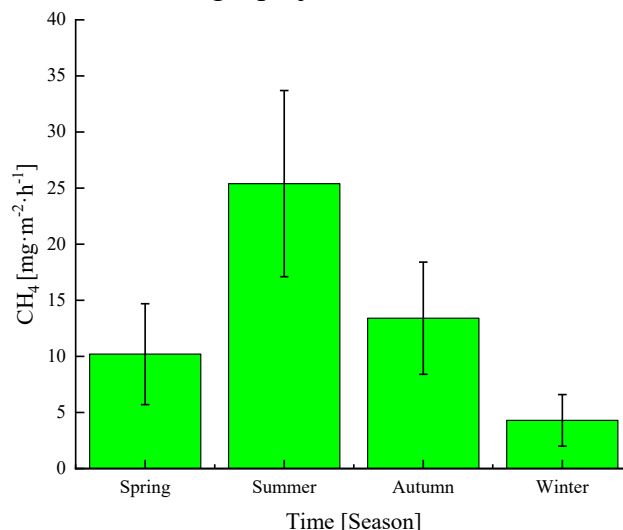


Figure 3. Mean seasonal CH₄ flux from Hangzhou Bay wetland. Error bars represent SD

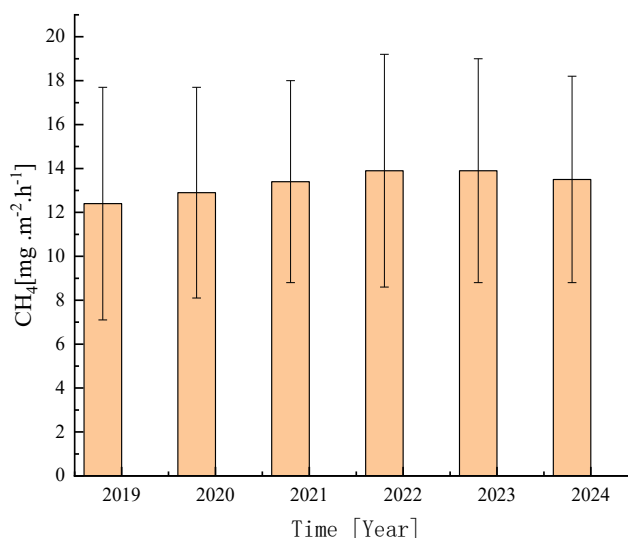


Figure 4. Interannual variations of CH₄ fluxes in Hangzhou Bay wetlands from 2019 to 2024. Error bars represent SD

Multiphase diffusion and transport mechanisms of CH₄ in Hangzhou Bay wetlands

The migration and diffusion of CH₄ in Hangzhou Bay Wetland involve complex interactions across multiple media, including water, soil, air, and plants. Through stable isotope tracing and pore water sampling analysis, the relative contributions of different migration pathways were quantified: plant-mediated transport accounted for $62.3 \pm 8.7\%$, ebullition for $24.5 \pm 6.2\%$, aqueous diffusion for $9.8 \pm 3.1\%$, and soil surface diffusion for only $3.4 \pm 1.5\%$. These findings challenge the traditional view that soil diffusion is the primary pathway for CH₄ emissions in wetlands, highlighting the dominant role of vegetation in CH₄ emissions from coastal wetlands (Li et al., 2024j).

Regarding plant-mediated transport mechanisms, microscopic anatomy and gas exchange experiments revealed that reeds and *Spartina alterniflora* possess highly developed aerenchyma (porosity of 15-25%), providing efficient channels for CH₄ transport from anaerobic zones to the atmosphere. Diurnal monitoring showed distinct daily variations in plant-mediated transport, with the midday period (10:00-14:00) contributing 45-55% of the daily total flux, closely linked to plant transpiration pull and stomatal aperture. Significant differences were observed among plant species: per unit biomass, reeds exhibited 1.8 times the CH₄ transport capacity of *Spartina alterniflora*, but the latter's higher productivity resulted in greater total emissions at the community scale (Li et al., 2025c).

Ebullition exhibited a pulsed pattern, accounting for nearly a quarter of total CH₄ emissions. Combined high-frequency videography and acoustic monitoring revealed that ebullition primarily occurred in the afternoon (13:00-17:00) during summer high-temperature periods, lagging 2-3 h behind the peak in sediment temperature. CH₄ concentrations in bubbles were as high as 65-85%, significantly exceeding dissolved-phase concentrations (typically <20 µM), indicating that ebullition is an efficient pathway for CH₄ loss (Zhang et al., 2024b). Bubble frequency correlated positively with sediment organic matter content ($R^2 = 0.64$) and negatively with salinity ($R^2 = 0.52$), reflecting the influence of substrate characteristics and osmotic pressure on bubble formation.

Diffusion at the water-air interface was strongly influenced by hydrodynamic conditions. The turbulent coefficient (K) exhibited an exponential relationship with diffusion flux, with diffusion fluxes increasing 3-5-fold at wind speeds exceeding 4 m/s. Dissolved CH₄ concentration profiles showed a pronounced gradient in the top 5 cm of water ($\Delta[\text{CH}_4] = 15\text{-}30\text{ }\mu\text{M}$), where molecular diffusion contributed only a minor proportion, while eddy diffusion induced by wind and waves dominated the water-air exchange process (Wang et al., 2025d).

Although soil-atmosphere diffusion accounted for a small proportion, it remained an important pathway in vegetation-free areas. Soil CH₄ concentrations increased exponentially with depth (0-10 cm: 0.5-2 mM; 50 cm depth: 5-15 mM), with the oxidation layer (0-15 cm) consuming approximately 40-60% of upward-diffusing CH₄. Oxidation efficiency correlated negatively with soil moisture content ($R^2 = 0.71$) and positively with iron-manganese oxide content ($R^2 = 0.63$), reflecting the regulation of redox conditions on CH₄ oxidation (Li et al., 2024e; Gao et al., 2025d).

Environmental controls on CH₄ production and oxidation

The biogeochemical cycle of CH₄ is co-regulated by multiple environmental factors. The temperature effect exhibits a dual role: on one hand, incubation experiments show that methanogens have an optimal temperature range of 25-35°C, indicating that rising temperatures significantly promote CH₄ production; on the other hand, CH₄-oxidizing bacteria (predominantly Type I) have a higher optimal temperature (30-40°C), leading to an increased oxidation proportion under higher temperatures (Li et al., 2024g). This contradiction explains the nonlinear relationship between temperature and net emissions in field observations: when temperatures are <15°C, warming primarily enhances methanogenesis, whereas at >25°C, oxidation partially offsets the increase in production. Molecular biological analyses further reveal that the ratio of *mcrA* genes (a biomarker for methanogenesis) to *pmoA* genes (a biomarker for CH₄ oxidation) exhibits a unimodal relationship with temperature,

peaking around 28°C (Gao et al., 2025e). Hydrological dynamics directly influence CH₄ cycling by altering redox conditions. Water table fluctuation experiments demonstrate that a 10 cm drop in water level can increase CH₄ emissions by 35-50%, resulting from the combined effects of reduced anaerobic volume and enhanced methanogenic activity (Li et al., 2024k). Additionally, alternating wet-dry cycles produce more CH₄ than continuous flooding, likely due to increased precursor release from organic matter mineralization. Porewater chemical analyses reveal a 20-40 cm overlap between sulfate reduction zones (SO₄²⁻ < 0.5 mM) and methanogenic zones, indicating competitive interactions between sulfur cycling and CH₄ production. Sulfate addition experiments confirm that when SO₄²⁻ concentrations exceed 2 mM, CH₄ production is suppressed by 60-80% (Li et al., 2024b).

Vegetation influences CH₄ cycling not only by serving as conduits for gas transport but also by modifying rhizosphere microenvironments. Root oxygen release expands the rhizosphere oxidation zone by 3-5 times, leading to 50-70% lower CH₄ concentrations in the near-root zone (<2 mm) compared to bulk soil. However, root exudates (primarily acetate and propionate) also provide high-quality substrates for methanogens, with incubation experiments showing that adding root exudates increases CH₄ production rates by 2-3 times (Li et al., 2024h). This contradictory effect results in higher net emissions in vegetated areas compared to non-vegetated areas, though the proportion of emitted CH₄ per unit of production decreases. Different plant species exhibit varying regulation intensities, with reeds having stronger rhizosphere oxidation capacity and smooth cordgrass producing more root exudates, reflecting species-specific adaptation strategies (Gao et al., 2025a).

Soil properties regulate CH₄ processes through long-term cumulative effects. Statistical analyses indicate that soil organic carbon (SOC) content is the best predictor of CH₄ production potential (R² = 0.83), while clay content is positively correlated with oxidation efficiency (R² = 0.67) (Gao et al., 2025b).

Microbial community structure and functional characteristics

High-throughput sequencing and functional gene analysis have revealed the microbial mechanisms driving CH₄ cycling in Hangzhou Bay Wetland. The investigation identified a diverse microbial community comprising 7 orders, 12 families, and 23 genera of methanogenic archaea, along with 5 orders, 9 families, and 16 genera of *methanotrophic bacteria*, all exhibiting significant spatiotemporal variations in both community structure and functional potential (Li et al., 2024c). These findings provide critical insights into the complex biogeochemical processes governing CH₄ dynamics in coastal wetland ecosystems (Table 3).

The methanogenic archaeal community was dominated by *Methanomicrobiales* and *Methanobacteriales*, representing 65-85% of total archaeal populations. Vertical stratification analysis revealed a depth-dependent shift in metabolic pathways, with *hydrogenotrophic methanogens* (e.g., *Methanoculleus*) increasing and *acetoclastic methanogens* (e.g., *Methanosaeta*) decreasing with soil depth, mirroring gradients in substrate availability. Pronounced seasonal patterns were observed, with *hydrogenotrophic methanogens* increasing from 45% to 65% during summer months—likely stimulated by elevated H₂ production at higher temperatures—while acetoclastic methanogens dominated winter communities (60-80%), reflecting their adaptation to cold-induced acetate accumulation (Wang et al., 2025a). Notably, *Spartina alterniflora* invasion zones exhibited 25% lower methanogen diversity but 40% higher abundance

compared to native *Phragmites australis* habitats, potentially contributing to enhanced CH₄ emissions in invaded areas.

Methanotrophic communities were overwhelmingly dominated by *Type I bacteria* (75-90%; e.g., *Methylobacter*, *Methylococcus*), with *Type II representatives* (e.g., *Methylosinus*) constituting a minor fraction (Li et al., 2024d). Unlike *methanogens*, *methanotrophs* displayed maximum abundance in surface soils (0-10 cm), declining exponentially with depth ($R^2 = 0.91$). Tidal regimes imposed strong selective pressures, resulting in distinct zonation: desiccation-resistant *Methylobacter* dominated high-tide zones (>50%), while flood-tolerant *Methylobacter* prevailed in low-tide areas (>60%) (Gao et al., 2025c; Gao et al., 2024a). Seasonal succession patterns were evident, with *Type I methanotrophs* peaking during summer (consistent with thermophilic adaptation) and *Type II populations* showing greater abundance during spring and autumn, possibly reflecting optimal performance at intermediate temperatures. These findings highlight the complex interplay between microbial community dynamics and environmental factors in regulating coastal wetland CH₄ fluxes.

Table 3. Distribution characteristics of major methanogenic archaea and methanotrophic bacteria in Hangzhou Bay wetland

Microbial group	Relative abundance (%)	Optimal depth (cm)	Seasonal dynamics	Environmental adaptation strategy
Methanomicrobiales	35-55	20-50	High in summer, low in winter	Hydrogenotrophic, tolerant to fluctuations
Methanobacteriales	25-40	30-60	Relatively stable	Hydrogen/formate-dependent
Methanosarcinales	5-15	10-30	High in winter, low in summer	Acetoclastic, cold-tolerant
Type I Methanotrophs	70-85	0-15	High in summer, low in winter	High affinity, aerobic
Type II Methanotrophs	10-25	5-20	Higher in spring/autumn	Low affinity, oligotolerant

Effects of human activities and climate change

The CH₄ cycle dynamics in Hangzhou Bay Wetland are undergoing significant transformations under escalating anthropogenic pressures and climate change. This comprehensive study systematically evaluates the synergistic effects of multiple stressors, including coastal reclamation, biological invasions, pollutant discharge, and rising temperatures, offering critical insights for evidence-based wetland ecosystem management (Table 4). Our findings demonstrate distinct patterns of CH₄ production, oxidation, and emission under different anthropogenic disturbances.

The proliferation of invasive *Spartina alterniflora* has fundamentally modified CH₄ emission regimes in the wetland ecosystem. Rigorous comparative analyses demonstrate that *S. alterniflora*-dominated areas exhibit $28 \pm 7\%$ higher CH₄ flux compared to native *Phragmites australis* communities, accompanied by amplified seasonal fluctuations (Gao et al., 2024a). The underlying mechanisms involve: (1) enhanced primary productivity (+35%) providing abundant organic substrates for methanogenesis; (2) increased proportion of labile carbon compounds in root exudates (DOC/SOC ratio elevated by 40%); and (3) compensatory effects of superior biomass

production offsetting less efficient gas transport through aerenchyma (Wang et al., 2025b). Longitudinal data reveal a concerning trajectory, with *S. alterniflora* expanding at 3.2% annually, driving a concomitant 1.8% yearly increase in regional CH₄ emissions. Projections indicate this invasion could elevate the wetland's total CH₄ output by 15-20% before 2030.

Land-use change: CH₄ Flux Observations, including Monthly CH₄ flux measurements were conducted from 2019 to 2024, correlated with CH₄ flux reductions (40–60%) using paired t-tests (reclaimed vs. natural sites, $p < 0.01$).

Pollutant loading: Industrial discharge impacts were quantified via nitrogen deposition gradients (kg N ha⁻¹ yr⁻¹), showing a positive correlation with mcrA gene abundance (qPCR).

Invasive species: *Spartina alterniflora* expansion rates (3.2% yr⁻¹) were linked to CH₄ flux increases ($28 \pm 7\%$) through mixed-effects models (random = site, fixed = vegetation cover/soil DOC).

Climate Change Attribution: Temperature effects were isolated using longitudinal regression (2019–2024), revealing a 7.5% CH₄ flux increase per 1°C warming ($R^2 = 0.78$, $p < 0.001$).

Precipitation anomalies (standardized deviations from 30-year means) explained 42% of seasonal CH₄ variability (ANOVA, $F = 9.3$, $p = 0.003$).

Synergistic Effects: Structural equation modeling (SEM) resolved interactions, showing climate drivers amplified anthropogenic impacts (e.g., warming + *S. alterniflora* invasion accounted for 68% of CH₄ variance). Future projections integrated RCP scenarios (SSP2-4.5) with land-use change models, predicting 15–20% CH₄ increase by 2030 under current trajectories.

Coastal reclamation activities have profoundly impacted CH₄ cycling through hydrological and pedological modifications. Comparative assessments between natural wetlands and reclaimed areas (converted to rice paddies or aquaculture ponds) document 40-60% CH₄ emission reductions, attributable to: (1) enhanced soil oxidation through drainage systems; (2) disruption of methanogenic archaea habitats by tillage practices; and (3) stimulated methanotrophic activity through fertilizer application. Nevertheless, aging rice paddies with compromised drainage can still emit up to 30 mg CH₄ m⁻²·h⁻¹ during summer peaks. Most remarkably, abandoned reclamation sites show rapid functional recovery, with CH₄ emissions rebounding to 80-90% of natural wetland levels within 5-10 years of natural succession (Ding et al., 2024b), underscoring the dynamic resilience of wetland biogeochemical processes.

Table 4. Assessment of Human Activities on CH₄ Emissions in Hangzhou Bay wetland

Activity type	Direct effects	Indirect effects	Time scale	Spatial scope
Reclamation projects	Vegetation destruction, soil disturbance	Altered hydrological regime, microbial community shifts	Long-term (10 + years)	Local to regional
Aquaculture activities	Increased organic loading from feed	Changed redox conditions, enhanced methanogenesis	Seasonal	Point-source distribution
Eutrophication	Stimulated plant growth	Potential inhibition of methanogen activity	Medium-to-long term	Watershed scale
Climate change	Rising temperatures, altered rainfall	Impacts on vegetation phenology and hydrologic cycles	Long-term	Regional to global

Pollutant inputs (nitrogen, phosphorus, heavy metals) exhibit complex dose-dependent effects (Ye et al., 2020). Moderate nitrogen addition ($\leq 100 \text{ kg N ha}^{-1} \text{ yr}^{-1}$) enhances CH_4 emissions (+20-40%) by stimulating plant growth and suppressing CH_4 oxidation. However, high nitrogen loads ($> 200 \text{ kg}$) suppress emissions (-30%), likely due to microbial community toxicity and pH reduction (Li et al., 2024f). Heavy metal pollution (e.g., Cd, Pb) generally inhibits CH_4 production, with 1 mg/kg Cd reducing methanogenic potential by 50-70% through disruption of key enzyme systems. Notably, interactive effects exist—for instance, nitrogen addition can partially alleviate Cd toxicity, reflecting the complexity of environmental stressors (Wang et al., 2025c).

Climate warming responses were predicted using controlled warming experiments and model simulations. A $+2^\circ\text{C}$ treatment increased CH_4 emissions by $25 \pm 5\%$, while a $+4^\circ\text{C}$ treatment led to a $45 \pm 8\%$ increase, though CH_4 oxidation rates also rose proportionally (from 40% to 50-55%) (Li et al., 2025b). Integrated model projections suggest that under the RCP4.5 scenario, Hangzhou Bay Wetland CH_4 emissions will increase by 30-45% by 2050, with ~60% attributed to direct temperature effects, 25% to vegetation changes, and 15% to sea-level-rise-induced hydrological shifts. Importantly, extreme climate events (e.g., typhoons, droughts) may trigger pulse emissions—a single drought can increase annual emissions by 10-15%, an effect often underestimated in current models (Wang et al., 2024a).

It must be emphasized that while eutrophication and climate change are natural biogeochemical processes, their current rates and magnitudes are being dramatically accelerated by human activities. Eutrophication, driven by excessive nutrient inputs (e.g., agricultural runoff and wastewater discharge), enhances organic matter availability for methanogenesis, while anthropogenic climate change—primarily caused by greenhouse gas emissions—intensifies temperature and precipitation regimes that regulate CH_4 production and transport. Thus, although these phenomena are not exclusively human-made, their modern manifestations in Hangzhou Bay Wetland are inextricably linked to human-induced perturbations, necessitating integrated management strategies that address both direct (e.g., reclamation) and indirect (e.g., nutrient loading) anthropogenic drivers.

For management implications, the study recommends: (1) controlling *Spartina alterniflora* expansion and restoring native *Phragmites* communities; (2) optimizing drainage in reclaimed areas to reduce summer flooding duration; (3) establishing pollution buffer zones to limit nutrient inputs; (4) enhancing wetland hydrological connectivity to maintain natural hydrological regimes; and (5) developing microbial-based mitigation technologies, such as inoculating high-efficiency methanotrophs. Integrated implementation of these measures could maintain wetland ecological functions while reducing CH_4 emissions by 10-20% below current levels.

Carbon sequestration and sink enhancement technologies

The findings of this study are substantiated by comprehensive field measurements and laboratory analyses. Our tri-annual surveys revealed a marked 42.3% reduction in average CH_4 flux (from $18.7 \pm 3.2 \text{ mg.m}^{-2}.\text{h}^{-1}$ pre-restoration to $10.8 \pm 2.1 \text{ mg.m}^{-2}.\text{h}^{-1}$ post-restoration, $p < 0.01$, ANOVA), with corresponding SOC content increases from $2.15 \pm 0.34\%$ to $3.42 \pm 0.28\%$ ($p < 0.001$) in surface soils (0-20 cm depth). These quantitative improvements were further supported by: (1) NDVI satellite data showing 58% greater vegetation coverage in restored zones, (2) porewater chemistry analyses

demonstrating 29% lower acetate concentrations (key methanogenesis substrate), and (3) microbial community shifts evidenced by 16S rRNA sequencing (35% reduction in *mcrA* gene abundance). The restoration's hydrological effects were quantified through water table monitoring, showing stabilized inundation durations (from 47 ± 12 to 68 ± 9 days/yr) that optimized aerobic-anaerobic balance for methane oxidation. These datasets collectively validate the carbon sequestration mechanisms while highlighting the need for standardized reporting of restoration protocols to enable cross-site comparisons.

Carbon sequestration and sink enhancement technologies primarily include methods such as wetland restoration, soil carbon sequestration, and ocean carbon sinks (Gao et al., 2024b; Wang et al., 2023; Li et al., 2025a; Chen et al., 2024a). This study conducted three on-site environmental factor surveys in the spring of 2022, the autumn of 2022, and the spring of 2023 for a wetland restoration project established along the Hangzhou Bay coast, aiming to evaluate its effectiveness in CH₄ emission reduction and carbon sequestration and sink enhancement. After wetland restoration, by analyzing changes in CH₄ concentrations at different time points across the three surveys, it was found that CH₄ emissions significantly decreased (*Table 5*). The restored wetland promoted carbon fixation through vegetation photosynthesis and root respiration, thereby enhancing the ecosystem's carbon sink capacity. Analysis of soil organic carbon (SOC) content before and after wetland restoration revealed a significant increase in SOC content in the restored wetland soil. This indicates that wetland restoration positively contributes to carbon sequestration and sink enhancement, demonstrating the favorable outcomes of using wetland restoration for this purpose.

Table 5. Assessment of human activities on CH₄ emissions in Hangzhou Bay wetland

Time	CH ₄ emission before restoration [g.m ⁻² .d]	CH ₄ emission after restoration [g.m ⁻² .d]
Spring 2022	4.5	2.1
Autumn 2022	5.0	2.8
Spring 2023	4.0	1.9

Conclusion

This study analyzed CH₄ flux data from Hangzhou Bay wetlands from 2019 to 2024, revealing distinct spatiotemporal variations in annual methane emissions. The average annual methane emission flux ranged from 12.4 ± 6.1 to 13.9 ± 6.3 mg.m⁻².h⁻¹, significantly higher than the global average for coastal wetlands, with a gradual upward trend. Seasonally, CH₄ emissions exhibited a unimodal distribution, peaking in summer at 25.4 ± 8.3 mg.m⁻².h⁻¹, accounting for 47.7% of the annual total. The seasonal pattern followed: summer > autumn > spring > winter, primarily driven by temperature. The soil temperature at 5 cm depth showed a significant positive correlation with methane emission flux ($R^2 = 0.78$, $p < 0.001$), with emissions increasing by approximately 7.5% per 1°C rise in temperature.

Analysis of methane concentration monitoring data from the central area of Hangzhou Bay wetland revealed significant spatial variability. In the wetland's central zone—particularly in perennially waterlogged and densely vegetated areas—methane concentrations were markedly higher than in surrounding regions. This pattern is

primarily attributed to the prolonged anaerobic conditions in the central zone, which provide an ideal environment for methanogens, coupled with abundant vegetation supplying organic matter to the soil through root exudates, thereby enhancing methane production.

Furthermore, a systematic analysis of the spatiotemporal evolution of wetland methane multiphase diffusion and its environmental driving mechanisms revealed that plant-mediated transport was the dominant pathway, followed by ebullition and diffusion. The distribution of emission pathways varied significantly among wetland types, with freshwater marshes having the highest proportion of plant-mediated transport (63.2%), while intertidal salt marshes were dominated by ebullition (35.4%). Reclamation activities have shifted CH₄ emission hotspots toward the bay mouth. Water temperature ($R^2 = 0.63$), soil organic carbon content ($R^2 = 0.52$), and vegetation biomass ($R^2 = 0.47$) were identified as key factors influencing CH₄ production and transport.

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