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Comprehensive approach to coastal lagoon ecological health evaluation: Example of Petrel Lagoon, Central Chile

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ABSTRACT

Coastal aquatic ecosystems are highly dynamic and influenced by multiple natural and anthropogenic factors that can contribute to their degradation. In this study, we present an innovative and interdisciplinary approach that integrates the analysis of the physicochemical properties, metal concentrations, and microbial communities in the San Antonio River and Petrel Lagoon, located in Central Chile, to evaluate the ecological health of this coastal ecosystem. Nutrient concentrations, in particular NO_3^- (values ranged from 26.6 ± 0.0 and 37.6 ± 9.4 mg/L) and PO_4^{2-} (values > 8.0 mg/L), were elevated upstream, reflecting agricultural inputs, while microbial indicators (*E. coli*) surpassed regulatory thresholds (>1,000MPN/100 ml) in several sites. Metal analyses revealed high Na and Mg, concentrations (>2,000 mg/L and > 280 mg/L, respectively) in waters and in Al, Fe and Li in sediments (values ranged from 20,000 to 1.0x10^6 mg/kg). Microbial community analyses in waters identified that most of the sites were dominated by *Idiomarinaceae* (16 % of total community) and *Marinicella* (5 % of total community), and with enrichment in *Owenweeksia* (12 % of total community) in downstream sites. Potential pathogenic genera, including *Cutibacterium acnes*, were detected, suggesting risks to water quality and public health. Multivariate analyses revealed spatial structuring driven by salinity and nutrient inputs, emphasizing the influence of hydrodynamics and land use. These findings highlight significant environmental gradients and contamination hotspots, stressing the need for a comprehensive approach that integrates a similar approach to better understand microbial dynamics, metal behavior, and their combined impact on water quality. Future studies should aim to overcome the limitations of this work in order to identify relevant indicators and develop effective monitoring strategies that support the sustainability of ecosystem services.

1. Introduction

Coastal ecosystems encompass diverse habitats at the interface between land and sea, including estuaries, salt marshes, mangroves, and coastal lagoons. These environments are characterized by their dynamic nature, shaped by the ebb and flow of tides, providing a first line of defense against storm surges, mitigating the impact of rising sea or flood and aquifer salinization, controlling erosion and coast stabilization (Barbier et al., 2011; Nayak and Bhushan, 2022; Perillo et al., 2018; Russi et al., 2013). Coastal lagoons are critical zones of ecological significance because they play a multifaceted role in supporting biodiversity, serving as essential breeding and nursery grounds for various

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marine species (Cognetti and Maltagliati, 2010; Inácio et al., 2018). Additionally, they act as natural filters, improving water quality through sediment, nutrient, and pollutant trapping (Hupp et al., 1993; Tuladhar and Iqbal, 2020). These ecosystems such as coastal wetlands, often referred to as "blue carbon" ecosystems, also play a crucial role in sequestering carbon dioxide from the atmosphere at rates higher than many terrestrial forests (Macreadie et al., 2017; Siikamäki et al., 2013). As the world faces the challenge of mitigating climate change, recognizing and preserving the carbon sequestration capacity of coastal ecosystems becomes paramount. Moreover, these coastal ecosystems contribute significantly to local economies by providing livelihoods for fishing communities and supporting tourism (Eppink et al., 2014; Schuyt, 2005).

However, these delicate and multifunctional ecosystems are increasingly threatened by environmental alterations, including climate change, growing anthropogenic pressures, and habitat destruction, which can severely impair their ecological health. Key symptoms of degradations are eutrophication, hypoxia, harmful algal blooms, and biodiversity loss, and are often exacerbated by internal processes such as sediment resuspension and nutrient recycling under anoxic conditions (Bancon-Montigny et al., 2019; Lund-Hansen et al., 1999; Miyamoto et al., 2019; Ulloa et al., 2017; Wurtsbaugh et al., 2019). Among these threats, numerous studies have documented the widespread and multifaceted consequences of pollution stemming from urban, agricultural, and industrial runoff, as well as untreated sewage discharge, which introduce both biological and chemical contaminants into coastal lagoons (García-Pintado et al., 2007; Rivera et al., 2019; Robledo Ardila et al., 2024; Robledo et al., 2023; Velasco et al., 2006). Common contaminants include pathogens, excess nutrients (nitrogen and phosphorus), heavy metals (e.g., mercury, lead, and cadmium), hydrocarbons, pesticides, and emerging pollutants such as pharmaceuticals and microplastics. Once introduced, these substances can enter the water column, bind to suspended particles, and accumulate in sediments, where they may persist for long periods (Cunha et al., 2020; Egbueri et al., 2024; Egbueri and Mgbenu, 2020; Moreno-González et al., 2013).

Despite these concerns, most studies assessing lagoon ecological health focus on a single environmental compartment (e.g., typically water, sediment, biota, or surrounding soils) while comprehensive, multi-compartment assessments remain scarce (Arienzo et al., 2013; Barik et al., 2019; Bloundi et al., 2009; García-Pintado et al., 2007). Research incorporating two or more compartments is still limited, even though excluding the sediment compartment often constrains our understanding of ecological health (Boubonari et al., 2009; Nazneen et al., 2022; Carvalho et al., 2002; Rivera et al., 2019). Sediments act not only as long-term sinks for contaminants but also as potential secondary sources of pollution. Under specific environmental conditions, especially under shifts in redox conditions or physical disturbances, sediments may release accumulated contaminants back into the water column, complicating risk assessments through delayed and non-linear feedback mechanisms. Understanding water–sediment exchanges is therefore critical for accurate ecological risk evaluations and the development of effective management strategies, particularly under changing climatic and hydrological regimes. Additionally, bioaccumulation in aquatic organisms is often under-assessed, despite its value in providing critical information on trophic transfer and long-term ecosystem impacts. Biota can serve as effective bioindicators of contamination, offering insights that are not captured by water or sediment analyses alone (Baralla et al., 2021; Burns and Smith, 1981; Li et al., 2019; Waykar and Deshmukh, 2012). While existing studies acknowledge the cumulative impact of pollution on biodiversity, microbial communities, benthic fauna, fish populations, and overall ecosystem functioning, substantial knowledge gaps remain. Unraveling the complex interplay among nutrient cycling, sediment dynamics, and anthropogenic pressures requires a comprehensive interdisciplinary approach that bridges ecology, biogeochemistry, hydrology, microbiology, and socio-environmental sciences. Unfortunately, truly interdisciplinary studies on the ecological health of coastal lagoons remain rare. Integrating diverse disciplines can yield a more holistic understanding of these complex systems, revealing hidden feedback, enhancing predictive capabilities, and informing strategies for resilience in the face of climate change and unsustainable land use. This approach is essential not only for developing sustainable management practices and guiding conservation actions, but also for fostering meaningful community engagement and preserving the ecological and socio-economic services coastal lagoons provide (Camacho-Ibar and Rivera-Monroy, 2014; Cañedo-Argüelles et al., 2012).

In this study, our main goal was to present a comprehensive approach to evaluating ecological health, incorporating both chemical and microbial community assessments of a coastal lagoon (Fig. 1). By integrating multiple disciplines and analytical tools, we aimed to assess the ecological health of these complex ecosystems. To this end, we conducted the first study applying this interdisciplinary approach to the San Antonio River–Petrel Lagoon system in Central Chile, which serves as a natural laboratory. Our first objective was to characterize the system through chemical and biological indicators in both water and sediment. The second objective was to elucidate the site's dynamics, focusing on the interactions between water and sediment. Finally, our third objective was to test and critically discuss the effectiveness of our interdisciplinary methodology, highlighting its benefits and emphasizing the urgent need for similar approaches to generate meaningful insights into environmental quality and the potential stressors impacting coastal lagoons.

2. Material and methods

2.1. Study area

The study area is located in Pichilemu, O'Higgins region (Central Chile), Chile (Fig. 2). This area is relatively flat and includes the Petrel Lagoon and the lower part of the San Antonio River that discharges into it. Pichilemu is a growing coastal town of almost 13,000 permanent inhabitants that also hosts many tourists (IMP, 2021). Besides, land use in the San Antonio watershed is dominated by forest plantations principally and native forests and agricultural lands in minor proportions (CONAF, 2024).

The Petrel Lagoon is a coastal wetland of 45 ha. approximately. The lagoon is most of the time isolated from the sea by a sandbar that only opens during major rainfall events or storm surges. When this occurs, a two-way hydraulic connection between the lagoon and the sea can develop for some time. The Petrel Lagoon was the first wetland of the ÓHiggins region (Central Chile) to be officially declared under the legal category of 'urban wetland', thus serving as a model for other environmental conservation initiatives (Biblioteca del Congreso, 2020). This lagoon, nestled within the captivating landscapes of Central Chile, stands as a testament to the ecological richness that characterizes the region. Petrel Lagoon functions as a vital breeding ground for numerous marine species, playing a pivotal role in the life cycles of fish and invertebrates. Its marshes provide shelter and nesting sites for almost 30 migratory bird species, contributing to the regional and international avian biodiversity (Allendes-Muñoz et al., 2023). The significance of Petrel Lagoon extends beyond its ecological role as it holds cultural and recreational value for the surrounding communities. However, Petrel Lagoon is confronted with a multitude of challenges that collectively threaten their ecological health and resilience. One of the primary concerns is pollution generated by urban and industrial runoff, agricultural inputs, and/or untreated sewage discharge (Velasco et al., 2006).

The San Antonio River drains a 199 km^2 watershed that rises up to an elevation of 616 m a.s.l. in the Chilean Coastal Range. The climate is semiarid, with an average precipitation of 455 mm/yr (1990–2019 average at the Pichilemu weather station operated by the Chilean

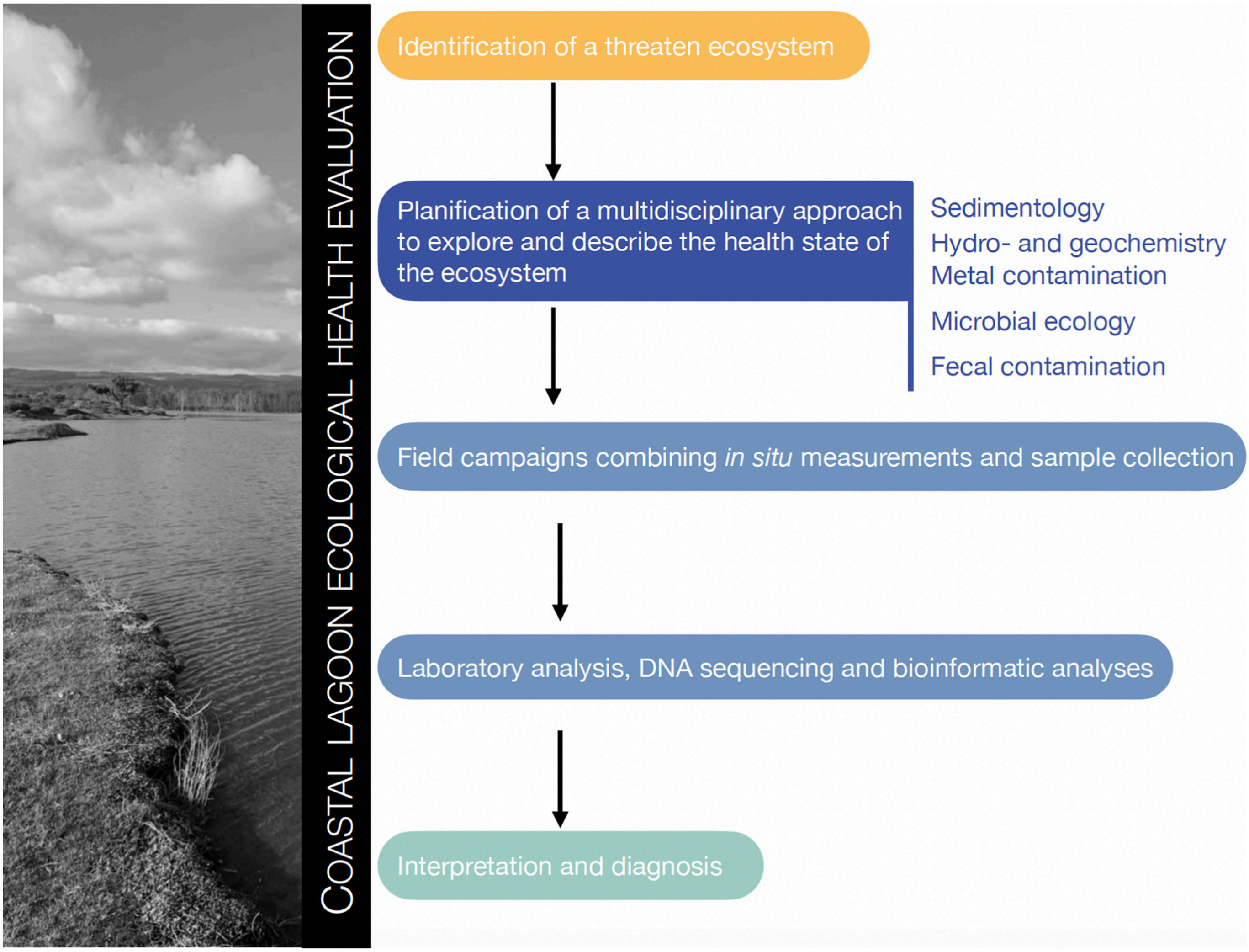

Fig. 1. Flowchart of the research steps for the ecological health evaluation of a coastal lagoon.

national water organization (Dirección General de Agua)). While the river is ungauged, qualitatively its discharge rate appears very low except for a few days following rainfall events.

The local geology consists of quaternary or older sediments of fluvial and potentially marine origin overlying Paleozoic metamorphic rocks (SENDOS, 1980). The thickness of the sediments can reach more than 70 m. The first 10 m (approximately) host an unconfined aquifer, and a confined aquifer has been recognized between about 40 m and 70 m deep. Groundwater quality is considered good in general (SENDOS, 1980).

Besides, land use in the San Antonio watershed is dominated by forest plantations principally and native forests and agricultural lands in minor proportions (CONAF, 2024).

2.2. Sampling strategy

Water (surface water – W and groundwater – GW) and sediment samples (S) were collected on January 17, 2023 (austral summer) (Table S1). In total, eight sites were selected including two in San Antonio River (i.e., the main tributary of the Petrel Lagoon), one in a small tributary of San Antonio River (Nuevo Reino River), one groundwater from a borehole (in connection with San Antonio River) and four in Petrel Lagoon (Fig. 2). Samples W1, S1 and W2, S2 are located upstream of the wastewater treatment plant (WWTP) of Pichilemu city whereas site W3, S3 is located 50 m downstream of the WWTP discharge. The fourth site (GW4) is on private land and corresponds to groundwater extracted from a borehole connected to the San Antonio River. Hence, no sediment was collected for this location. Site W5, S5 corresponds to the most downstream sampling site in the San Antonio River. The three last sampling sites (W6, S6; W7, S7; and W8, S8) are distributed along the lagoon shoreline. Overall, the Petrel Lagoon is potentially threatened by the presence of a horseback riding area without facilities to manage horse waste, eutrophication due to agricultural and urban runoff, as well as by the dynamics of the sandbar, which cause saline intrusion. The W6, S6 site is located at the discharge point of the San Antonio River while W7, S7 is located parallel to the coast. The W8, S8 sampling site presents the particularity to be located where discharge from another lagoon occurs during rainfall (i.e., drain outlet).

2.3. Surface water and groundwater analyses

2.3.1. Physicochemical parameters

Temperature, pH, electrical conductivity (EC), dissolved oxygen (DO), and oxidation–reduction potential (ORP) were measured *in situ* using an LAQUAact 100 sensor (Horiba, Japan). Additionally, conventional water quality parameters, including chemical oxygen demand (COD) and total suspended solids (TSS) were analyzed following standard methods (APHA, 2005). Ammonium (NH_4^+), nitrate (NO_3^-), nitrite (NO_2^-), and orthophosphate (PO_4^{3-}) were analyzed using Merck cell tests

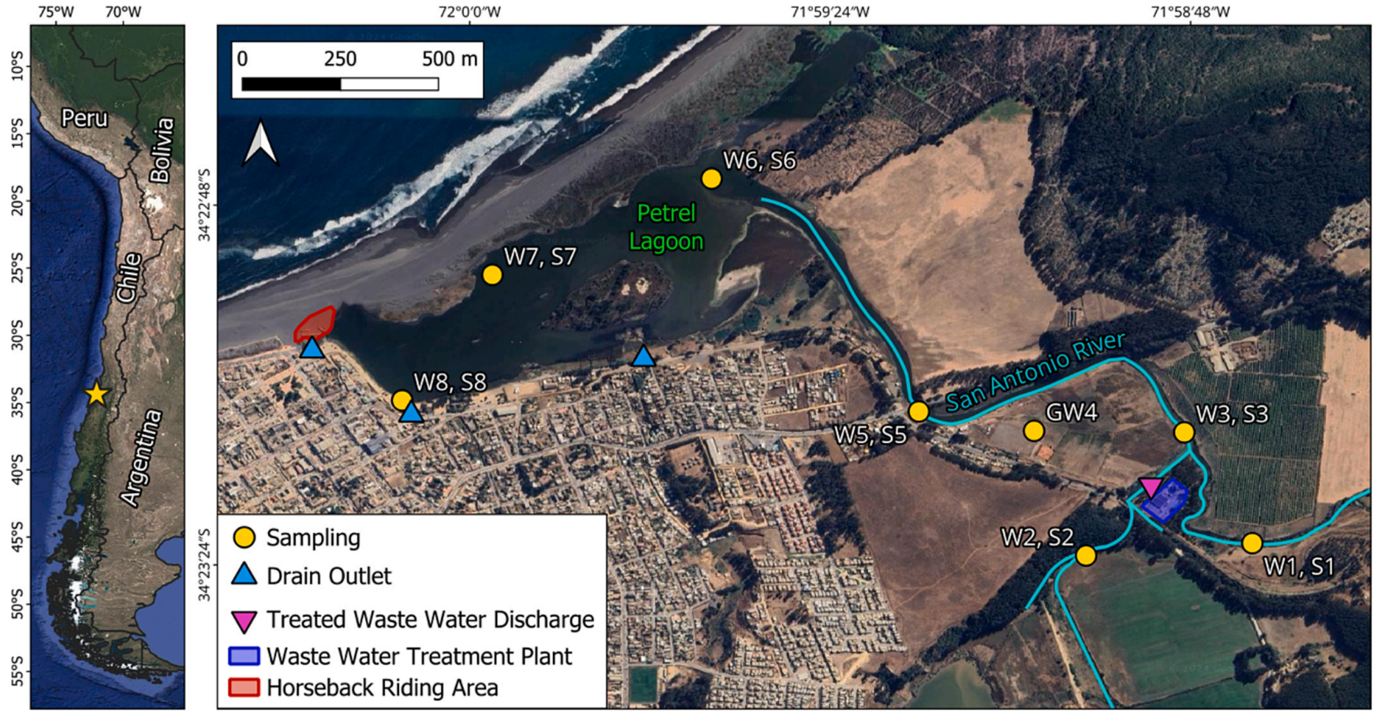

Fig. 2. Locations of the sampling sites in San Antonio River-Petrel Lagoon systems. W and GW indicate surface water and groundwater samples, respectively, while S indicates sediment samples.

on a Spectroquant Nova 60 spectrophotometer (Merck, Germany). Chlorophyll *a* concentration was determined following the US EPA Method 446.0 (Arar, 1997).

2.3.2. Metals and total mercury

Bulk metal(oides) concentrations (not filtered samples) were measured directly in pre-acidified water samples using an inductively coupled plasma to optical emission spectrometry (ICP-OES, model iCAP Pro, ThermoScientific, USA). Certified reference material Trace Metals 1-WS (Sigma Aldrich, Merck) was used. Total mercury (tHg) was measured following an adaptation of the US EPA 1631 (EPA US, 2002) as described in (Torres-Rodriguez et al., 2024) allowing a detection limit of 0.2 pM.

2.3.3. E. coli

E. coli quantification was estimated using the most probable number (MPN) technique, based on Walker et al., (2018). Briefly, volumes of 10, 1, and 0.1 mL of each water sample were inoculated in a set of three tubes (9 tubes in total) with mineral-modified glutamate broth (MMGB) medium (Merk) and incubated at 37 °C for 24 h. From the tubes that turned yellow, 0.1 mL were plated on a Petri plate with tryptone bile X-glucuronide (TBX) agar (Merk) and incubated at 44 °C for 21 h. The corresponding tube was considered positive if blue-green colonies indicative of *E. coli* were observed on the plates. The MPN for the sample was determined by inspecting the combination of positive tubes per dilution and their associated MPN in the MPN table (USDA, 2014). MPNs were expressed in MPN/100 mL.

2.3.4. Microbial diversity

DNA extraction was performed from 20 μL water samples under a Biohazard Safety Cabinet (JSCB-900SB) using a microvolume physical lysis extraction (Bramucci et al., 2021). DNA samples were quantified with Qubit 1X dsDNA HS Assay kit (assay range between 0.1–120 ng) using QuBit 4 fluorometer, according to the manufacturer's instructions. DNA templates were sequenced with Illumina NovaSeq 6000 PE250 at Novogene using in-house primers 515F (GTGYCAGCMGCCGCGGTAA) and 806R (GGACTACNVGGGTWTCTAAT) for the V4 region of the 16S rDNA gene based on Caporaso et al. (2011).

2.4. Sediment analyses

2.4.1. Physicochemical parameters

Grain size analyses were carried out on sediment samples. After drying the sediments in the oven at 40–50 °C for up to 5 days, organic matter was eliminated in a portion of each of these samples (1000–500 gr) using 30 % H_20_2 at high temperature following the methodology of (Vaasma, 2008). This chemical treatment was applied 1–2 times in each sample until the reaction with organic matter was null. Finally, the processed sediment samples were dry-sieved using a ro-tap sieve shaker for 5 min. The weight (g) of each fraction retained in the sieves was measured and transformed to weight percent.

Soil pH and electrical conductivity (EC) were determined in water extract from a solid-to-liquid mixing ratio of 1:2.5 and 1:5 (w/v) respectively. The available phosphorus content (P-Olsen) was measured by extraction with sodium bicarbonate, and colorimetric determination was made using blue-molybdate. Total organic carbon (TOC) was determined by oxidation of organic matter with a mixture of dichromate and sulfuric acid and then by photometric determination by the chromium ion (Cr^{3+}) (Sadzawka R. et al., 2006).

2.4.2. Metals and total mercury

Frozen sediment samples were primarily freeze-dried 96 h at −50 °C and 1 atm (LGJ-18, Nanbei, China), then sieved through 0,25 mm, grounded in an agate pestle for homogenization, and finally digested following the US EPA 3052 method (US EPA, 2015) slightly modified. Briefly, 200 mg of grounded sediment were digested in a microwave (Metash, modelo MWD-700) in 6 mL of Ultrapure HNO_3 (EMSURE, Supelco, Sigma-Aldrich) and 2 mL of Ultrapure HCl (PanReac AppliChem ITW reagents, Spain). Digested samples were diluted in milli-Q water (10/40, vol/vol), and filtered through ash-free filter papers (390/W40, Sartorius, Germany) before being analyzed using an inductively coupled plasma to optical emission spectrometry (ICP-OES, model iCAP Pro, ThermoScientific, USA). Certified reference material BCR-277R (estuarine sediment, IRMM) was used. Total mercury (tHg) in grounded sediments was assessed directly through cold-vapor atomic fluorescence spectroscopy (CV-AFS, DMA-80, Milestone, USA) using the same reference material.

2.4.3. *E. coli*

For the quantification of *E. coli* in sediments, a procedure similar to that for the water samples was followed, except that the dilution series were made with 1, 0.1, and 0.01 g from each sample.

2.4.4. *Microbial diversity*

In sediment, gDNA was extracted from 250 to 300 mg of wet sample using DNeasy PowerSoil Pro Kit (QIAGEN, USA) following the manufacturer's recommendations. Between 60.32 and 200.99 ng/μL of gDNA was obtained as measured through spectrophotometry (EPOCH2, Bio-Tek). Metabarcoding of the V4 region of the 16S rRNA gene (515FB = GTGYCAGCMGCCGCGGTAA; 806RB = GGACTACNVGGGTWTCTAAT; (Walters et al., 2015)) was performed at the Integrated Microbiome Resource (IMR) of Dalhousie University (Halifax, Canada).

2.5. *Data analyses*

2.5.1. *Metals and total mercury*

The background values for every metal in waters and sediments corresponded to the mean of the values comprised between the mean +2 standard deviation (sd) and the mean – 2*sd (Akoto and Anning, 2021; Haris et al., 2017). Then, in water, the contamination factor (Cf) was assessed following the method of Akoto and Anning (2021) and the heavy metal evaluation index (HEI) was calculated according to Şener et al., (2023) were values <10 reveal low metal contamination, values from 10 to 20 reveal medium contamination and values >20 are considered as high metal contamination. In sediments, the contamination factor (Cf) and ecological risk index (ERI) were calculated following the method of Akoto and Anning (2021). Additionally, the Pollution load index (PLI) was estimated considering values of PLI ≤ 1 as only baseline levels of pollutants and PLI > 1 a progressive deterioration of the sediment quality (Şener et al., 2023).

The sediment samples were collected in triplicate, whereas water samples were not. Therefore, statistical analysis was performed only on the sediment sample results. Thus, a first principal component analysis (PCA) was performed using the *PCA* function of the 'FactoMineR' package (Husson et al., 2013) to identify patterns along the studied transect based on the major element composition. A first PCA with the sediment samples was performed using the 20 available elements. To enhance the robustness of the analysis, 10 elements were subsequently selected based on collinearity.

2.5.2. *Microbial diversity*

Sequencing data was processed using a customized pipeline largely based on 'VSEARCH' (Rognes et al., 2016). PhiX reads were removed with Bowtie2 (Langmead and Salzberg, 2012), and primer sequences were trimmed using Cutadapt (Martin, 2011). Paired-end reads were merged using the fastq_mergepairs algorithm in 'VSEARCH' and quality-filtered using a maximum expected error of one with the fastq_filter algorithm in 'VSEARCH' (Edgar and Flyvbjerg, 2015). The sequences were merged, quality-filtered, and then deduplicated using the derep_fulllength algorithm in 'VSEARCH'. The UNOISE algorithm, implemented in 'VSEARCH' (Edgar, 2016), was used to retrieve amplicon sequence variants (ASVs). The uchime algorithm, also implemented in 'VSEARCH' (Edgar et al., 2011), was used to detect and remove chimeric ASVs. The remaining ASVs were verified using Metaxa2 (16S rRNA) to evaluate the presence of non-target sequences for those genes (Bengtsson-Palme et al., 2015). The final ASV table was obtained by mapping the high-quality sequences against the verified ASV centroids using the usearch_global algorithm in 'VSEARCH'. Taxonomic classification was performed using the Naïve Bayesian classifier Sintax, implemented in 'VSEARCH' (Edgar, 2016), to classify the sequences against the non-redundant SILVA v132 database (Quast et al., 2013). ASVs assigned to mitochondria, chloroplasts, eukaryotes, rare ASVs with an abundance of <0.005 %, sparse ASVs that occurred in fewer than four samples (representing 1/6 of the sampling effort), and outlier ASVs, where the highest abundance was more than 0.15 times that of the second most abundant value across other samples, were removed. Finally, the filtered ASV table was normalized by iterative subsampling with 100 iterations using the *rrarefy* function in the 'vegan' package in R (Oksanen et al., 2020). Taxonomic composition and relative abundance plots were generated using 'ggplot2' v3.4.1 in R. Differences in β-diversity were assessed by calculating Bray–Curtis dissimilarities based on the iteratively rarefied ASV abundance tables, and PCoA analyses were performed to visualize changes in microbial community structure. A distance-based redundancy analysis (dbRDA) (Legendre and Anderson, 1999) was used to constrain β-diversity patterns in water and sediment physicochemical properties using the *dbrda* and *envfit* functions from the 'vegan' package. Finally, a differential ASV analysis was performed using the 'DESeq2′ package (Love et al., 2014) from non-normalized libraries, to determine the ASV that varied significantly (p-value adjusted < 0.01) between the clusters identified by multivariate analysis. Among the selected ASVs, those whose combined total abundance (across all samples) of more than 3 % of the total ASVs were displayed in a heatmap, using the *heatmap.2* function from the 'gplots' v3.1.3.1 package.

3. Results

3.1. *Water samples*

Temperature ranged from 17.8 to 25.1 °C with consistent values (21.3–25.1 °C) for surface water (W1-3 and W5-8), while groundwater (GW4) recorded the lowest at 17.8 °C (Table 1). The pH values varied between 8.0 and 8.7, slightly alkaline and consistent with the local geology dominated by metamorphic rocks (Sernageomin, 2002). Electrical conductivity ranged from 3,380 to 18,410 μS/cm with the highest values at the Petrel Lagoon points W6-8. The lowest value (3380 μS/cm) was observed for the groundwater samples (GW4). These greatly exceed the Chilean standard for irrigation water NCh1333 (Norma Chilena oficial 1333 de 1978) (e.g., < 750 μS/cm; (INI, 1987)), likely due to saline intrusion. ORP ranged from -83 to -120 mV, indicating a reducing environment in both the San Antonio River and the Petrel Lagoon. DO levels varied from 5.76 to 18.6 mg/L in surface waters and 0.6 mg/L in borehole water. Chlorophyll *a* concentrations were notably higher upstream (W1-3 and W5-6) with values ranging from 109.6 ± 9.5 μg/L to 196.5 ± 8.5 μg/L, indicating the presence of eutrophic conditions. In contrast, downstream sites W7 and W8 have moderate concentrations (< 26.0 μg/L). The lowest value (e.g., 1.4 ± 0.6 μg/L) was found in the groundwater sample. TSS concentrations ranged from 7.0 ± 0.7 to 26.1 ± 11.4 mg/L, with the lowest value measured at GW4. COD values were also lowest at W4 (5.5 mg/L), while the remaining samples showed values between 8.0 and 35.0 mg/L, with the highest values (> 30 mg/L) observed at W1 and W5. NO_3^- concentrations ranged from 4.0 ± 0.0 to 37.6 ± 6.3 mg/L with the lowest value in groundwater (GW4) and the highest values at W1, W2, W3, and W5 sampling points upstream of the Petrel Lagoon, reflecting areas potentially impacted by agricultural activities. NH_4^+ concentrations ranged from 0.3 ± 0.0 to 9.6 ± 0.3 mg/L, following a similar trend to NO_3^-. NO_2^- concentrations showed consistent concentration (e.g. from 6.1 ± 0.2 to 8.2 ± 1.9 mg/L). PO_4^{2-} values ranged from 2.5 ± 1.3 to 9.5 ± 0.4 mg/L, with the lowest at GW4 and consistently higher values (>8.4 mg/L) in surface waters across all sampling sites. The highest *E. coli* concentrations were observed upstream and furthest from the Petrel Lagoon, with values exceeding 2,400 MPN/100 mL at W2 and W3, and 1100 MPN/100 mL at W1. From W5 onward, lower concentrations were observed, ranging from 13 to 160 MPN/100 mL. These latter values are below the Chilean standard for

Table 1
Physical, chemical, and microbiological parameters of water samples from the study area.

Sample ID	Temp (°C)	pH	EC (µS/cm)	ORP (mV)	DO(mg/L)	Chl-a (µg/L)	TSS(mg/L)	COD (mg/L)	NH_4^+ (mg/L)	NO_3^-(mg/L)	NO_2^- (mg/L)	PO_4^{2-}(mg/L)	E. coli MPN/100 ml
W1	24.2	8.6	13,930	−118	18.6	162.3 ± 3.8	22.1 ± 2.8	37.1	8.7 ± 1.9	26.6 ± 0.0	7.2 ± 4.7	8.9 ± 0.4	1,100
W2	22.9	8.6	13,260	−118	16.5	196.5 ± 8.5	16.8 ± 4.0	13.4	9.6 ± 0.3	35.4 ± 6.3	8.1 ± 0.2	8,7 ± 0.2	>2,400
W3	25.1	8.5	14,710	−115	18.4	130.4 ± 6.5	16.8 ± 1.2	26.9	6.1 ± 0.2	35.4 ± 6.3	6.1 ± 2.1	9.1 ± 0.7	>2,400
GW4	17.8	8.7	3,380	−120	0.3	1.4 ± 0.6	7.0 ± 0.7	5.5	0.3 ± 0.0	4.0 ± 0.0	6.6 ± 1.4	2.5 ± 1.3	0
W5	23.4	8.6	14,710	−119	n.d[a]	121.1 ± 37.5	19.9 ± 1.2	35.0	5.4 ± 0.4	37.6 ± 9.4	8.2 ± 1.9	8.4 ± 0.7	240
W6	22.3	8.3	18,410	−108	18.4	109.6 ± 9.5	26.1 ± 11.4	23.6	2.7 ± 0.9	22.1 ± 6.3	6.1 ± 0.2	8.4 ± 0.2	240
W7	21.3	8.2	17,550	−117	5.8	15.1 ± 2.4	17.1 ± 0.4	8.0	1.0 ± 0.1	17.7 ± 6.3	6.7 ± 0.7	9.5 ± 0.4	460
W8	21.5	8.0	17,710	−83	6.9	25.8 ± 0.6	8.4 ± 0.3	29.8	1.0 ± 0.1	24.3 ± 15.7	7.6 ± 0.5	9.2 ± 0.4	43

[a] Not determined.

recreation activities.

A total of 21 different metals, including total mercury, were detected in the water samples (Table 2). In surface water, the elements with the highest concentrations were Na and Mg, with values ranging from 2,443.8 to 3,396.9 mg/L and 280.4 to 387.6 mg/L, respectively, which is consistent with the high conductivity reported. Ca, K and S were also present in high concentration with values ranging from 103.8 mg/L and 261.9 mg/L. Also, P and B were detected with concentrations ranging from 0.890 to 2.7 mg/L. The rest of the elements presented a concentration below 1 mg/L. Additionally, it is noteworthy that a concentration of 2.1 ng/L of total mercury was detected at the most upstream sampling point, while this value does not exceed the Chilean standard 1333. In groundwater (GW4), the same metals were detected, but in almost all cases, the concentrations were much lower than in surface water, except for Ca, where the concentration was similar (109.7 mg/L), and for Fe, where the concentration was the highest among all water samples, with a value of 2.7 mg/L and probably related to the local geology (Fuentes et al., 2024). The HEI values ranged between 1.7 and 4.2 revealing low metal contamination in the studied waters (Table S4).

The microbial community consisted of over 99 % Bacteria, while Archaea were detected only in W7 and W8 at a relative abundance < 0.05 % of the total community. The main phyla were Proteobacteria (48.8 %), Bacteroidetes (17.5 %), Actinobacteria (15.6 %) and Cyanobacteria (14.1 %). Interestingly, the *Idiomarinaceae* family represented on average 15.7 % in each sample. The groundwater GW4 exhibited a very different assemblage dominated by the Proteobacteria *Burkholderiaceae* affiliated to Ralstonia. The PCoA explained 78.9 % of the variation of the microbial diversity in superficial water samples (e.g.,

Table 2
Metal concentrations in water samples from the study area.

Metal (mg/L)	ID sample W1	W2	W3	GW4	W5	W6	W7	W8
tHg[a]	2.111	0.643	0.670	0.331	0.312	0.385	0.348	0.537
Ag	0.028	0.024	0.023	0.068	0.025	0.024	0.034	0.028
Al	0.257	0.319	0.143	0.085	0.179	0.620	0.437	0.154
As	0.009	0.008	0.009	0.013	0.010	0.009	0.012	0.011
B	0.944	0.890	1.001	0.175	1.014	1.080	1.183	1.183
Ba	0.039	0.037	0.038	0.006	0.038	0.043	0.041	0.038
Be	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL
Ca	114.6	111.3	121.5	109.7	123.0	126.0	135.3	136.7
Cd	BDL[b]	BDL	BDL	BDL	BDL	BDL	BDL	BDL
Co	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL
Cr	0.007	0.007	0.007	0.009	0.007	0.008	0.007	0.006
Cu	0.004	0.004	0.003	0.003	0.003	0.003	0.003	0.003
Fe	0.239	0.293	0.186	2.700	0.229	0.739	0.527	0.211
K	108.9	103.8	116.7	28.9	117.7	122.5	133.9	136.0
Li	0.017	0.016	0.017	0.023	0.017	0.018	0.019	0.019
Mg	302.3	280.4	316.8	137.5	319.3	338.8	380.1	387.6
Mn	0.212	0.175	0.183	0.536	0.178	0.190	0.241	0.216
Mo	0.002	0.002	0.002	0.003	0.002	0.002	0.002	0.002
Na	2,616.8	2,443.8	2,765.0	417.6	2,769.7	2,946.0	3,299.5	3,396.9
Ni	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL
P	2.7	2.6	2.7	0.272	2.7	2.5	2.7	2.7
Pb	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL
S	208.7	197.4	220.7	49.3	220.4	231.8	257. 2	261.9
Se	BDL	BDL	BDL	BDL	0.0013	BDL	0.0014	BDL
V	0.007	0.007	0.007	BDL	0.007	0.009	0.008	0.007
Zn	0.008	0.006	0.005	0.004	0.005	0.005	0.004	0.002

[a] Total Mercury is expressed in ng/L.
[b] Below detection limit.

excluding the groundwater sample GW4; Fig. 4a and Fig. S1 with GW4). This multivariate analysis revealed two distinct clusters along the studied transect: the first group included sites W1 to W6, while the second group comprised sites W7 and W8. Using a differential analysis, we identified 100 discriminant ASVs affiliated to 55 genera responsible for the significant differences between the clusters (DESeq2, $p < 0.01$; Fig. 3c). On one hand, the upstream sites (i.e., from W1 to W6) were characterized by five ASVs with higher relative abundance compared to W7-W8: ASV12 *Alcanivoracaceae Marinicella* (mean relative abundance of the genus in this cluster = 4.8 %), ASV19 *Idiomarinaceae Idiomarina* (mean relative abundance of the genus in this cluster = 20.4 %), ASV40 *Rhodobacteraceae* (mean relative abundance of the genus in this cluster = 7.4 %), ASV44 *Cyclobacteriaceae Algoriphagus* (mean relative abundance of the genus in this cluster = 2.7 %) and ASV48 *Cyclobacteriaceae*

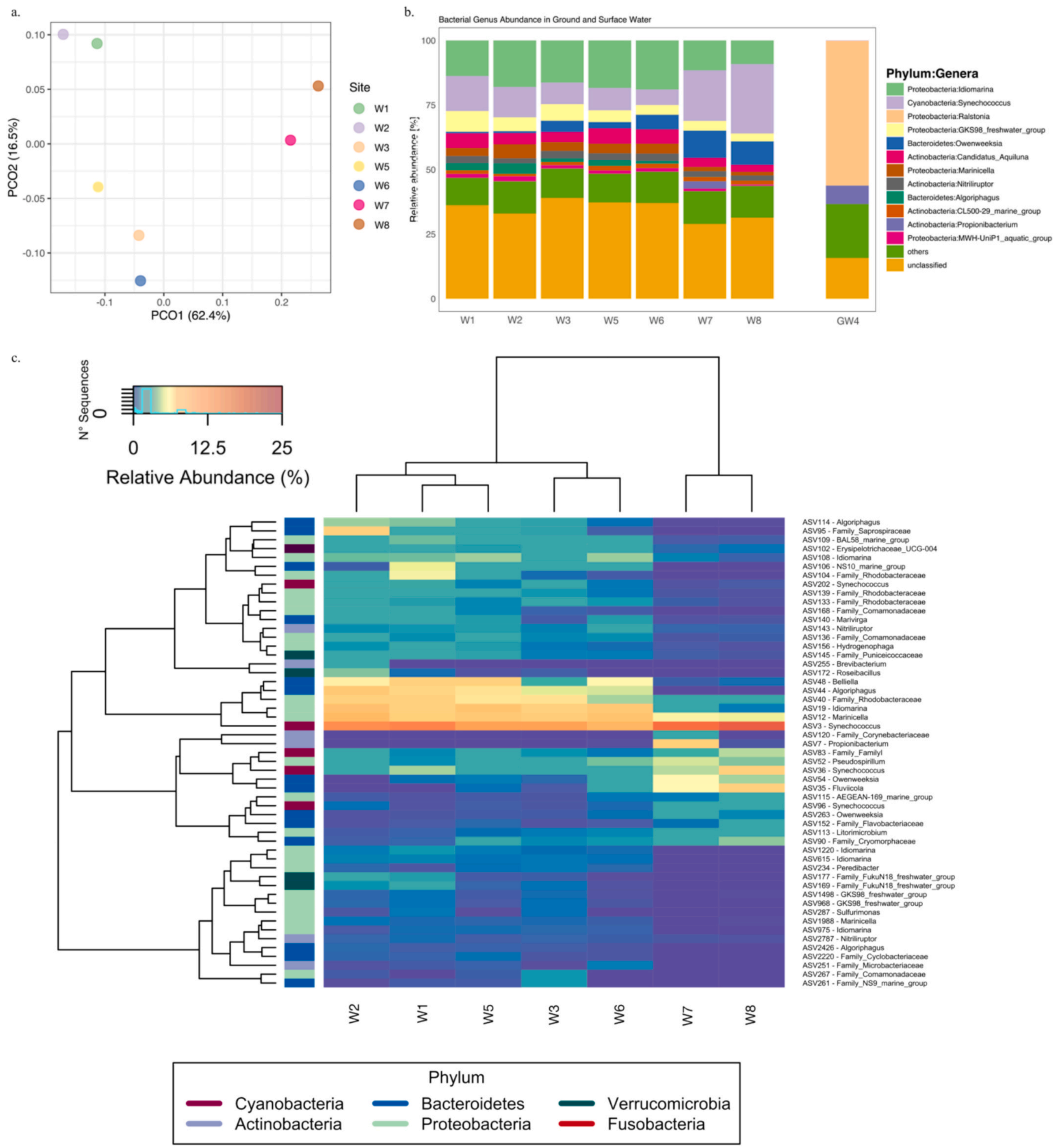

Fig. 3. Microbial communities in water. a) PCoA based on the Bray-Curtis distance at the ASV level clustering the 7 superficial water samples (excluding the groundwater W4); b) Relative abundance of the main 12 genera from all the superficial and subterranean water samples; c) Heatmap built with the relative abundance of the discriminant ASVs significantly different among the two clusters (i.e., identified by DESEq2 differential analysis with $p < 0.01$, see method section) with a total relative abundance (sum in all 7 samples) > 30 %.

Belliella (mean relative abundance of the genus in this cluster = 1.8 %). On the other hand, the coastal water samples W7 and W8 were characterized by a progressive enrichment in ASVs affiliated to *Flavobacteriales Owenweeksia* reaching 11.9 % of the total microbial community in W7 (Fig. 3b). The search for potential pathogens in water revealed the presence of 18 genus over 109 listed in the literature (https://www.hartmann-science-center.com/en/hygiene-knowledge/pathogens-a-z). Among them, in superficial water, site W7 was dominated by Propionibacterium, with 3.1 % of total relative abundance affiliated to *Cutibacterium acnes*. In the groundwater GW4, we also retrieved a high relative abundance of Propionibacterium in addition to the Proteobacteria *Burkholderiaceae* affiliated to *Ralstonia*.

3.2. Sediment samples

In the sediment samples S2, S3, S5, and S6, the sandy fraction predominates, with a medium to fine sand distribution ranging from 56.3 % (e.g., P5) to 94.0 % (e.g., P6) (Table S2). In contrast, S1, S7, and S8 exhibit a more heterogeneous distribution characterized by a higher proportion of coarse materials (granules, very coarse and coarse sand), with the combined sum of these grain sizes ranging from 41.7 % (e.g., P7) to 53.6 % (e.g., P8). The predominance of sandy fractions in S2, S3, S5, and S6 suggests relatively high-energy depositional environments, whereas the heterogeneous distribution of coarse materials in S1, S7, and S8 implies localized hydrodynamic variability and potential sediment transport from upstream sources. It is noteworthy that sample S8 shows the highest percentage of granules, with a value of 30.4 %.

In all sediment samples, pH values were homogeneous, ranging from 6.9 ± 0.1 to 7.2 ± 0.2 (Table 3). Electrical conductivities range from 1.3 ± 0.1 to 2.0 ± 0.0 µS/cm, except for sediment S1, which shows a higher value of 6.9 ± 0.0 µS/cm. The total organic carbon content follows a similar pattern to electrical conductivity, with an average value of 1.3 % for all sediments except S1, which exhibits a value of 6.3 ± 0.8 %. However, this trend is reversed for P-Olsen concentration, where the lowest concentration (4.1 ± 0.2 mg/kg) was observed in S1. At the same time, the rest of the sediment samples show concentrations between 9.6 ± 0.7 and 13.7 ± 0.2 mg/kg.

E. coli was also detected in sediment samples. The lowest concentrations were observed in Petrel Lagoon (S6, S7, and S8) and at the most upstream sampling point (S1), with values ranging between 92 and 430 MPN/100 mL. A concentration of 1,100 MPN/100 mL was found at S2, the second point most upstream to the lagoon. The sediment samples located downstream of the treated wastewater discharge (e.g., S3 and S5) presented the highest concentration (>11,000 NPM/100 mL), indicating the presence of fecal contamination in sediments.

A total of 22 different metals, including total mercury, were detected (Table 4). The concentration ranges were wide, with the highest levels observed for Li, Al, Fe, and S. The PCA performed with the 10 selected elements explained 80.48 % of the dataset variation (Fig. S2). The first dimension explained 52.82 % of the dataset variation and was mainly characterized by Hg, Zn, Mo, S, and Ca. The second dimension explained 27.66 % of the dataset variation and represented Mg and Pb. The combination of these two dimensions allowed for the separation of the sediment samples into 4 clusters. Interestingly, the two tributaries to the mainstream exhibited different metal compositions. S1 presented the highest content of Cu, Hg, S, Mo, Zn (i.e., 28.5, 38.8, 21,832.3, 73.2, 119.5 mg/kg, respectively; note that Hg is expressed in ng/g), whereas S2 shows highest content of Fe, Mn, and V (i.e., 45,380.5, 567.6, 252.3 mg/kg, respectively). Notably, S2 clusters with S5 and S3, the following sampling sites in the stream flow. Within the coastal lagoon, the S6 and S7 sampling sites cluster together, characterized by a high content of Ca and Mg and a specificity for S6 which was found moderately enriched in Pb. Surprisingly, the closest sediments to the sea in S8, cluster separately in the middle of the ordination, with high concentrations in Cd and Hg. Accordingly, to the PCA ordination, the highest PLI values were retrieved in S1 and S8 (PLI > 1.2) suggesting a progressive degradation of the sediment quality (Table S6).

Considering the microbial community, no archaeal sequences were detected. Among Bacteria, the three dominant phyla were Proteobacteria (63.1 % relative abundance of the total community), Bacteroidetes (10.7 %), and Planctomycetes (8.6 %). Within Proteobacteria, Gammaproteobacteria predominated, followed by Betaproteobacteria and Deltaproteobacteria, with mean relative abundances of 33.4 %, 15.1 %, and 7.7 %, respectively. The PCoA ordination (explaining 72.8 % of dataset variation; Fig. 4a) revealed two samples with distinct microbial assemblages. First, sample S1 was dominated by 100 ASVs affiliated with the sulfur-oxidizing Halothiobacillus (57.3 %; Fig. 4b), consistent with the high sulfur concentration at this site. Second, sample S6 exhibited a microbial community comprising 60.6 % unidentified representatives of the mostly mesophilic *Comamonadaceae* family (Willems, 2014), followed by the Firmicutes *Bacillus* (26.8 %). Interestingly, the predominant Bacillus-related ASV 1507, was not related to potentially pathogenic strains but to environmental uncultured bacterium associated with dust (99.3 % homology with JF832327; Chuvochina et al., 2011) and close to *Niallia circulans* strain NBRC 13626 (accession number NR_112632) with 98.6 % homology. Within the remaining group of samples, S7 and S8 exhibited a slightly different assemblage, as revealed by the DESeq2 analysis (Fig. 3c), with 6.4 % and 7.2 % of aerobic chemoorganotroph Planctomycetota Pirellula-affiliated ASVs.

Table 3
Physical, chemical, and biological parameters of sediment samples from the study area.

Sample ID	pH	EC (µS/cm)	TOC (%)	P-Olsen (mg/kg)	E. coli MPN/100 ml
S1	7.2 ± 0.2	6.9 ± 0.0	6.3 ± 0.8	4.1 ± 0.2	230
S2	7.1 ± 0.0	1.4 ± 0.0	1.3 ± 0.1	13.2 ± 0.2	1,100
S3	6.9 ± 0.1	1.9 ± 0.0	1.9 ± 0.1	10.6 ± 0.1	>11,000
S5	7.0 ± 0.1	1.5 ± 0.0	1.0 ± 0.2	14.5 ± 0.1	>11,000
S6	7.1 ± 0.1	1.3 ± 0.1	0.8 ± 0.2	9.6 ± 0.7	230
S7	7.2 ± 0.0	1.7 ± 0.0	0.9 ± 0.0	10.2 ± 0.0	92
S8	7.0 ± 0.1	2.0 ± 0.0	1.7 ± 0.0	13.7 ± 0.2	430

4. Discussion

4.1. Ecological health of the Petrel Lagoon

The water quality analysis of the San Antonio River and the Petrel Lagoon reveals significant spatial variations in physicochemical parameters, microbial communities, and contamination levels, reflecting multiple environmental influences along the transect of the study area. Upstream (W1-W5) San Antonio River samples are characterized by levels of DO (>16 mg/L) and nutrients (NO_3^-: 4.0–37.6 mg/L, NH_4^+: 0.3–9.6 mg/L and PO_4^{2-} > 8.4 mg/L) that highlight eutrophication concerns. Additionally, the elevated chlorophyll-a levels (109.6–196.5 µg/L) suggest algal proliferation, likely fueled by agricultural runoff from the surrounding areas. Fecal contamination is also evident, with *E. coli* concentrations exceeding 2,400 MPN/100 mL at W1–W3, likely originating from surrounding agricultural activities or illegal discharges or from the treated wastewater discharge upstream of W3 (Fig. 2). Metal contamination was not identified when compared to the Chilean standards (NCh1333). However, W1 exhibited the highest Hg concentration confirmed by the contamination factor (Cf) with a value of 4.8 and highest levels also in S1 (with a Cf of 3.1) (Table S3). The microbial communities were first composed by *Alteromonadales Idiomarinaceae*. As

Table 4
Metal concentrations in sediment samples from the study area.

Metal (ng/g)	ID sample S1	S2	S3	S5	S6	S7	S8
tHg[a]	38.8 ± 1.4	13.1 ± 0.8	9.9 ± 1.8	15.6 ± 1.4	6.1 ± 0.8	6.4 ± 0.1	29.8 ± 10.8
Metal (mg/kg)	ID sample S1	S2	S3	S5	S6	S7	S8
Ag	11.0 ± 17.9	65.2 ± 15.4	32.4 ± 8.3	20.2 ± 4. 1	310.2 ± 79.7	141.1 ± 27.0	83.1 ± 16.4
Al	384,054.5 ± 105,122.9	260,239.6 ± 53,903.3	279,670.7 ± 28,475.3	323,245.5 ± 15,635.8	300,207.3 ± 8,941.7	281,010.6 ± 9,684.0	300,937.6 ± 8,873.6
B	78.3 ± 6.0	74.4 ± 1.0	56.1 ± 4.5	59.6 ± 18.2	50.7 ± 0.9	40.0 ± 0.7	73.0 ± 2.9
Ba	365.3 ± 33.9	265.3 ± 2.6	183.8 ± 21.6	260.1 ± 34.3	301.4 ± 66.9	168.6 ± 24.2	198.6 ± 13.0
Be	0.297 ± 0.085	BDL[b]	BDL	BDL	BDL	BDL	BDL
Ca	3,868.4 ± 104.9	4,338.3 ± 7.8	4,506.5 ± 113.2	4,334.1 ± 68.8	5,246.6 ± 65.5	5,205.3 ± 88.3	4,858.9 ± 83.0
Cd	3.3 ± 0.6	4.1 ± 0.1	3.0 ± 0.2	3.8 ± 0.0	2.7 ± 0.2	2.1 ± 0.0	4.0 ± 0.2
Co	21.0 ± 1.1	21.8 ± 0.4	18.1 ± 1.3	23.3 ± 0.1	16.2 ± 0.8	14.6 ± 0.1	21.1 ± 0.5
Cr	112.2 ± 7.1	126.4 ± 2.2	87.9 ± 6.2	115.4 ± 2.8	74.0 ± 3.9	54.6 ± 1.4	122.0 ± 8.4
Cu	28.5 ± 2.5	12.8 ± 0.5	13.7 ± 1.2	15.4 ± 0.1	16.1 ± 0.4	17.6 ± 0.1	24.0 ± 1.4
Fe	42,124.0 ± 1,582.0	45,380.5 ± 626.8	35,714.0 ± 2,322.6	42,589.3 ± 222.0	31,898.6 ± 1,210.8	26,660.1 ± 326.9	43,047.0 ± 968.7
K	6316.7 ± 590.1	3,327.6 ± 251.0	2,962.1 ± 260.7	3,338.1 ± 145.1	2,180.9 ± 39.8	2,347.7 ± 215.7	3,679.1 ± 208.1
Li	2,382,686.5 ± 216,984.9	1,185,474.5 ± 50,330.1	1,364,237.5 ± 133,510.4	1,386,480.3 ± 63,599.0	1,326,129.2 ± 62,440.0	1,469,765.7 ± 77,926.0	1,860,430.1 ± 78,294.9
Mg	6,164.2 ± 528.4	4,280.0 ± 495.8	4,704.4 ± 295.2	4,411.9 ± 38.5	7,791.1 ± 399.8	7,570.6 ± 291.4	8,408.5 ± 130.4
Mn	567.6 ± 23.2	741.8 ± 39.2	452.9 ± 24.3	549.5 ± 15.9	564.1 ± 23.4	491.6 ± 17.0	611.0 ± 10.4
Mo	73.2 ± 8.1	44.5 ± 2.44	42.6 ± 5.5	50.3 ± 3.0	48.2 ± 3.0	44.2 ± 1.3	47.5 ± 1.9
Na	14,469.6 ± 4,449.6	1,859.2 ± 198.8	2,166.4 ± 190.7	2,199.3 ± 46.3	3,034.5 ± 141.5	4,203.4 ± 1,064.3	4,290.5 ± 289.7
Ni	BDL	BDL	BDL	BDL	BDL	BDL	BDL
Pb	61.5 ± 15.5	33.7 ± 6.4	17.4 ± 3.0	17.6 ± 0.3	85.8 ± 33.6	26.1 ± 6.8	39.9 ± 2.5
S	21,831.3 ± 2,215.7	1,124.7 ± 191.6	1,387.0 ± 249.4	1,032.9 ± 23.8	1,468.2 ± 61.6	2,230.7 ± 319.2	4,094.4 ± 287.3
V	185.6 ± 4.5	252.3 ± 8.0	188.0 ± 11.2	225.0 ± 1.5	156.8 ± 8.1	123.8 ± 0.3	234.5 ± 9.1
Zn	119.5 ± 9.6	98.9 ± 2.3	73.6 ± 2.5	96.3 ± 0.8	64.6 ± 3.5	53.6 ± 1.3	110.9 ± 3.2

[a] Total mercury.
[b] Below detection limit.

halophilic microorganisms, the representants of *Idiomarinaceae* exhibit a diverse set of metal resistance genes conferring them the ability to grow in contaminated areas or areas with a high background in metals (Kaur and Kaur, 2024). Their presence along the studied stream suggests that this metal resistance capacity makes them among the most competitive taxa in the studied ecosystem.

Then, a shift in water quality is observed downstream in Petrel Lagoon. Electrical conductivity values (3,380–18,410 µS/cm) indicate potential saline intrusion, where levels exceed Chilean irrigation standards (NCh1333). Microbial composition also changes with an increasing proportion of cyanobacteria *Synechococcus* and *Flavobacteria Owenweeksia*. *Owenweeksia* is not well studied but commonly found in the sea water bacterioplankton as an aerobic heterotrophic bacteria (Zheng et al., 2023) capable for degradation of degrading high-molecular-mass organic matter (Lau et al., 2005). Also reported as a new microorganism useable for bioremediation of the pesticide fenvalerate, this organism may dominate in the coastal lagoon as one of the main actors for organic matter recycling and remediation of potential pesticide contamination from agricultural effluents. The occurrence of *Cutibacterium acnes* dominating W7 (3.1 %), suggests different contamination sources and environmental conditions. However, coastal influence seems to mitigate contamination through dilution and tidal flushing, a pattern observed in other estuarine and lagoon systems (Mitchell et al., 2017; Portnoy and Allen, 2006). By contrast, groundwater sample (GW4) exhibited the lowest nutrient concentrations, confirming that surface runoff is the primary pollution source rather than contamination through lateral and vertical water soil movement. However, metal analysis indicates high Fe contamination (Cf = 7.83) and moderate contamination for Ag and Mn, suggesting alternative contamination pathways (Table S3 and 4), possibly from geological leaching or industrial effluents (Gonzalez et al., 2004). The microbial community was also distinct, with *Ralstonia* being the most prominent genus, probably due to the specific geochemical composition of the water and suggesting potential risk for human health since this genus has been reported as a human pathogen (Ryan and Adley, 2014) and potentially resistant to a large set of antibiotics (Falcone-Dias et al., 2012).

The sediment composition of the San Antonio River–Petrel Lagoon system varies significantly along the transect, reflecting differences in depositional environments, contamination sources, and ecological risks. The S1 sample exhibits distinct physicochemical characteristics compared to other sites. It has the highest organic carbon content (6.3 %), likely due to organic matter accumulation from upstream sources (e. g. agricultural activities). Despite this, its low P-Olsen concentration (4.1 mg/kg) suggests limited bioavailable phosphorus, possibly due to mineral adsorption or complexation. Conductivity is notably higher at this site (6.9 µS/cm), indicating localized enrichment of dissolved ions. Microbial community analysis also reveals a specific and unique assemblage. Additionally, metal analysis shows that S1 has the highest concentrations of Cu, Hg, S, Mo, and Zn, suggesting a distinct contamination source. This pattern may be associated with anthropogenic activities such as industrial processes, although no specific information confirming the presence of such activities was reported. Alternatively, it may reflect the sités historical use as dumping area. This hypothesis is strengthened by the highest relative abundance of *Halothiobacillus* purple bacteria in this site since these sulfur-oxidizing bacteria are known to be present in mine tailing discharges (Kimura et al., 2011). Interestingly, the dominance of *Halothiobacillus* with high concentration of oxygen and high conductivity suggest a complete sulfur oxidation and hence low risk for acidification events due to mining effluents (Liu et al., 2025). Midstream, S2, S3 and S5 share similar geochemical fingerprint, with elevated Fe, Mn, and V concentrations suggesting common sedimentary inputs. The predominance of sandy fractions at this section of the transect indicates relatively high-energy depositional environments. *E. coli* concentrations exceed 11,000 MPN/100 mL at S3 and S5, pointing to significant fecal contamination likely originating from treated wastewater discharge upstream of S3. These contamination patterns resemble those observed in other anthropogenically impacted wetland sediments (Walton et al., 2024). Additionally, Cd poses a high ecological risk at S2 and S5 with values of ERI > 20 (Table S7) (Akoto and Anning, 2021). By contrast, the sediment composition and biogeochemical fingerprint in Petrel Lagoon reflects a transition mainly influenced by

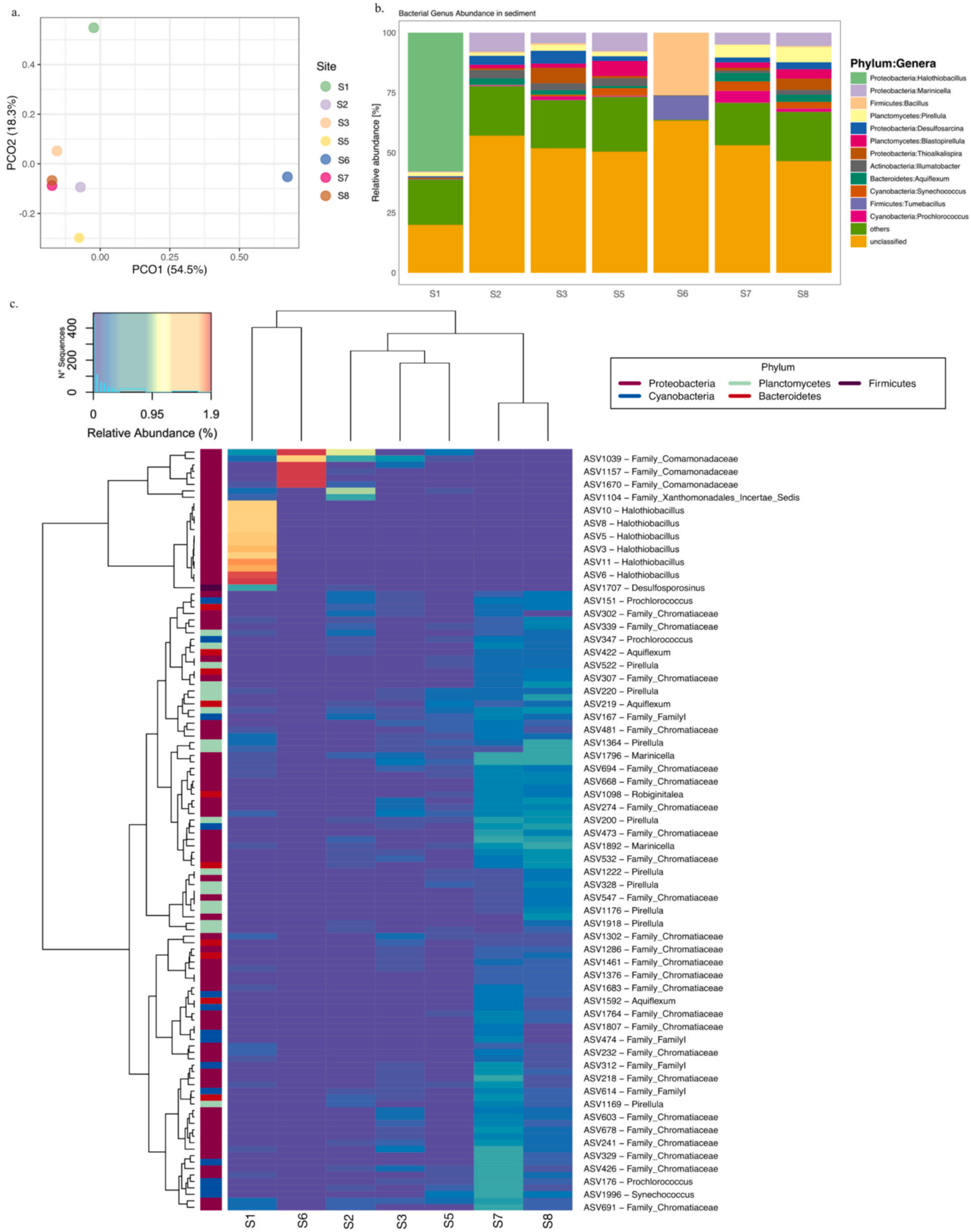

Fig. 4. Microbial communities in sediments. a) PCoA based on the Bray-Curtis distance at the ASV level clustering the 7 sediment samples; b) Relative abundance of the main 12 genera from all the sediment samples; c) Heatmap built with the relative abundance of the discriminant ASVs significantly different among the two clusters (i.e., identified by DESEq2 differential analysis with $p < 0.01$, see method section) with a total relative abundance (sum in all 7 samples) > 30 %.

marine processes. S6 and S7 cluster together due to high Ca and Mg concentrations, while S8, which contains 30.4 % granules, stands out with high Cd and Hg levels, suggesting unique sedimentary dynamics (Fig. S2). *E. coli* concentrations are lower at S6–S8, likely due to dilution and reduced direct pollution sources as it was observed in surface water samples. Regarding metal contamination, it remains a concern as Ag and Pb exhibit elevated Cf values at S6, and Pb contamination at this site is classified as a moderate ecological risk (Table S5). S8 presents one of the highest ecological risks due to elevated Cd levels, as also confirmed with PLI values > 1 (Table S6). The substantial ERI and PLI values at these locations suggest long-term contamination, likely from upstream sources or historical anthropogenic activities (Table S6-7). Along the transect, microbial community analyses revealed assemblages typical of coastal aquatic sedimentary environments, particularly with the occurrence of Gammaproteobacteria dominant taxa in Chilean coastal wetlands (Pozo-Solar et al., 2022) but also suggesting anthropogenic influences.

The findings highlight varying degrees of contamination and ecological risk across the San Antonio River–Petrel Lagoon system. While some areas exhibit low contamination levels, hotspots of concern include elevated nutrient and microbial contamination in W1–W3 due to agricultural runoff and treated wastewater discharge, as well as metal pollution, particularly cadmium, in S1, S2, S5, and S8. The detection of E. coli and distinct microbial assemblages further underscores potential health risks associated with fecal contamination.

Compared to similar ecosystems in a Mediterranean-climate region, such as El Yali wetland, our study similarly identified significant anthropogenic pressures (Rivera et al., 2019). However, while El Yali was characterized by high concentrations of heavy metals (e.g., particularly in copper and lead) in the water column and an overall poor ecological status as determined by oxygen demand-based indices, our findings point to a different contaminant profile and potentially distinct sources of environmental stress. For example, the sediment quality and microbial community observed in our study may reflect chronic contamination patterns or greater internal biogeochemical buffering capacities. Methodologically, while (Rivera et al., 2019) applied classical water quality and multivariate analysis approaches, our approach integrates microbiological, geochemical, and hydrodynamic perspectives. This interdisciplinary framework enables a more holistic understanding of ecosystem dynamics and may capture complex interactions often overlooked in more narrowly focused evaluations.

4.2. Dynamics of the water column

As previously observed, the study area exhibits significant heterogeneity in its physicochemical and microbiological characteristics. In aquatic ecosystems, the interactions between the water column and sediments are inherently complex, driven by physical, chemical, and biological processes that regulate contaminant distribution and persistence (Cappellen and Gaillard, 2018; Taylor and Owens, 2009). Understanding these dynamics is crucial for assessing ecosystem health and developing effective management and conservation strategies. In the San Antonio River-Petrel Lagoon system, the results clearly demonstrate how these interactions play a key role in modulating contamination patterns. Similar to findings in other Chilean coastal lagoons, such as El Yali (Rivera et al., 2019), the results indicate that while water quality is strongly influenced by agricultural runoff, wastewater inputs, and coastal dynamics, sediments function as both a sink and a potential secondary source of contaminants (Carvalho et al., 2002). The accumulation of contaminants, such as metals or fecal bacteria, in sediments poses a significant ecological risk, as sediments act not only as long-term sinks but also as potential secondary sources of pollution (through their remobilization) under changing environmental conditions. Hydrodynamic shifts, redox fluctuations, microbial activity, and organic matter degradation, all influence the chemical speciation and mobility of contaminants stored in sediments, potentially leading to episodic releases that can generates unexpected water quality deterioration. Such dynamics have been reported in Central Chile (Gana et al., 2024) and globally (Brito et al., 2009; Gikas et al., 2006; León et al., 2017). Hence, given these complex interactions, it is essential that both water and sediment compartments are considered in monitoring programs, especially in the context of conservation efforts and regulatory frameworks. Water samples offer only a temporal snapshot, reflecting conditions at a specific moment and heavily influenced by hydrological variability. In contrast, sediments function as a latent reservoir of contaminants like a "pollution time bomb" with the potential to release harmful substances gradually or abruptly, depending on environmental triggers. This unpredictability limits the ability of stakeholders to anticipate contamination events and respond effectively. Indeed, key questions remain regarding the extent to which sediment-bound contaminants contribute to water quality degradation over time and how seasonal variations influence these processes. Further research and continuous long-term monitoring are essential to understanding these dynamics and designing effective strategies for the conservation of this protected ecosystem.

4.3. Limitations of the study

Although our study presents a promising and comprehensive approach to evaluating the ecological health of a coastal lagoon, several limitations must be acknowledged. First, the findings should be considered preliminary, providing an initial assessment of the ecological condition of Petrel Lagoon. While our interdisciplinary and integrative methodology offers valuable insights, the absence of sample replicates and the restriction to a single sampling event during the summer of 2023 limit the representativeness of our results. In particular, water quality assessments reflect a temporal snapshot and are highly sensitive to seasonal and hydrological variability. Thus, our data captures only the state of the lagoon at a specific point in time, rather than its overall or long-term ecological status.

To more accurately evaluate the lagoon's ecological health and to assess the effects of potential management or regulatory interventions, future efforts must incorporate replicated sampling and a long-term, seasonally resolved monitoring framework. Additionally, it is essential to include targeted sampling during specific environmental events that are likely to influence ecosystem dynamics. In the case of Petrel Lagoon, such events include the natural or artificial (mechanically induced) opening of the sandbar, upstream dam releases, as well as extreme flooding or drought conditions. These events may significantly alter hydrological connectivity, water chemistry, and sediment dynamics, and therefore should be prioritized for additional monitoring.

Despite the strengths of our interdisciplinary approach and the simultaneous analysis of both water and sediment compartments, an effective monitoring program must be adaptive and comprehensive. This includes incorporating regular assessments of persistent and emerging chemical contaminants, such as pharmaceuticals, personal care products, and microplastics. Only through such a robust, multi-compartmental, and event-responsive monitoring strategy can we generate the information necessary to guide evidence-based ecosystem management.

Ultimately, ensuring the ecological health of Petrel Lagoon is not only critical for maintaining biodiversity and ecosystem function, but also for protecting public health by preventing harmful algal blooms or microbial contamination and sustaining key economic activities such as tourism, which is vital to the local economy of Central Chile. A healthy lagoon will contribute to both ecological integrity and the long-term well-being and prosperity of the surrounding human population.

4.4. Implications for the conservation of shallow coastal Petrel Lagoon

Petrel Lagoon has been designated as a protected area under the national legislation Ley de Humedales Urbanos N° 21.202 (Biblioteca del Congreso Nacional | Ley Chile, 2021), which aims to safeguard urban wetlands in Chile. This law acknowledges the ecological significance of wetlands and strives to protect these vital environments from degradation caused by urban development. It mandates that local municipalities identify and classify these wetlands, develop management plans with conservation strategies and regulations, and assess the potential environmental impacts of any development projects affecting them. Additionally, the law requires regular monitoring of wetland conditions and the enforcement of regulations to prevent illegal activities and mitigate environmental harm. To support the success of this legal framework for the conservation of Petrel Lagoon, studies such as those presented here are essential, as they provide comprehensive tools to identify ecological threats and facilitate the effective implementation of conservation solutions.

Concerns about contamination and the conservation of Petrel Lagoon are not new and were raised decades before it became a protected area under the Ley de Humedales Urbanos legislation. Pichilemu is a coastal city and the most popular tourist destination in the O'Higgins Region during the summer. However, despite its importance, the city lacked a WWTP for many years, even as it was recognized as an area severely affected by multiple sources of contamination, including agricultural runoff and the direct discharge of untreated wastewater. After years of advocacy and struggle, a WWTP was finally implemented near W3 in 2010. However, despite its installation, our results show that contamination persists in both the water column and sediments, raising concerns about the long-term ecological health of the system. The accumulation of metals in sediments and the high nutrient concentrations in the water should serve as a strong warning to policymakers, as agricultural runoff and potential leaks from wastewater infrastructure continue to degrade water quality in the San Antonio River–Petrel Lagoon system. This underscores the need for an integrated management approach that extends beyond wastewater treatment, incorporating stricter regulations, improved monitoring, and targeted strategies to address sediment contamination, diffuse pollution sources, and groundwater-surface water interactions. Without such efforts, the ongoing contamination threatens the ecological integrity of Petrel Lagoon, necessitating a multifaceted conservation strategy to preserve biodiversity including aquatic microorganisms, improve water quality, and ensure the long-term sustainability of this aquatic system.

5. Conclusion

The comprehensive analysis of water quality, sediment composition, and microbial communities in the San Antonio River and Petrel Lagoon highlights significant spatial variability and multiple contamination sources impacting the ecosystem. Upstream sections (W1–W5) are characterized by high nutrient concentrations, algal proliferation, and fecal contamination, largely driven by agricultural runoff and wastewater discharges. In contrast, coastal influence in the lagoon (W7–W8) appears to modulate contamination levels, altering microbial assemblages and reducing nutrient loads. Groundwater (GW4) exhibits distinct geochemical signatures, suggesting an alternative contamination pathway, possibly linked to industrial or geological inputs. Sediment analyses reveal heterogeneous depositional environments and localized contamination hotspots, with high cadmium (Cd) concentrations at S1, S2, S5, and S8 posing potential ecological risks. The presence of *E. coli* and the occurrence of metals in sediments further underscores contamination concerns and potential consequent public health issues. These preliminary results demonstrated a fragile ecosystem subject to recurrent contamination and vulnerable to episodic pollution peaks, particularly triggered by natural environmental events that may lead to the remobilization of contaminants stored in sediments. Hence, this first study on San Antonio River-Petrel Lagoon systems underscores the need for targeted management strategies to mitigate contamination sources and safeguard its ecological integrity. Long-term monitoring efforts should focus on nutrient fluxes, microbial indicators, and heavy metal accumulation to assess the effectiveness of mitigation measures. Additionally, improving wastewater treatment and promoting best agricultural practices are essential to reducing chemical and microbial pollution. Wastewater treatment can be enhanced by adopting new technologies and optimizing existing systems, including the use of bioremediation and the implementation of buffer zones. Similarly, agricultural practices can be improved through more effective nutrient management, water conservation strategies, and measures to enhance soil health. Implementing these solutions will significantly improve water quality, strengthen ecosystem resilience, and positively impact the sustainability of ecosystem services, particularly those related to tourism and local economic activities associated with the lagoon.

CRediT authorship contribution statement

Morgane Derrien: Writing – review & editing, Writing – original draft, Visualization, Validation, Supervision, Methodology, Investigation, Formal analysis, Data curation, Conceptualization. **Céline Lavergne:** Writing – review & editing, Writing – original draft, Visualization, Validation, Supervision, Methodology, Investigation, Formal analysis, Data curation, Conceptualization. **Polette Aguilar-Muñoz:** Writing – review & editing, Visualization, Formal analysis, Data curation. **Yoelvis Sulbaran:** Writing – review & editing, Visualization, Validation, Data curation. **María Soledad Pavlov:** Writing – review & editing, Data curation. **Macarena Pérez:** Writing – review & editing, Visualization, Data curation. **Carolina Reyes:** Writing – review & editing, Methodology, Data curation. **Gabriel Arriagada:** Writing – review & editing, Validation, Data curation. **Etienne Bresciani:** Writing – review & editing, Visualization, Data curation, Conceptualization. **Ismael Maldonado:** Writing – review & editing, Methodology, Data curation. **Tania Villaseñor:** Writing – review & editing, Validation, Data curation. **Verónica Molina:** Writing – review & editing, Resources, Project administration, Funding acquisition. **Claudia Rojas:** Writing – review & editing, Project administration, Investigation, Funding acquisition.

Declaration of competing interest

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

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Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.ecolind.2025.113679.

Data availability

Data will be made available on request.

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