## A baseline soil survey of two peatlands associated with a lithium-rich salt flat in the Argentine Puna: physico-chemical characteristics, carbon storage and biota

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## SUMMARY

High-elevation peatlands are valuable because of their capacity to store high quantities of carbon (C) in their soils, but they are also vulnerable to disturbance caused by human activity. In the High Andean plateau, lithium mining is a growing industry that could have an unknown impact on peatlands associated with the lithium-rich salt flats under exploitation. We assessed different abiotic and biotic soil characteristics of the Laguna Negra (LN) and Laguna Tres Quebradas (LTQ) peatlands in the Argentine Puna, both of which may imminently be affected by lithium mining. We measured soil physico-chemical properties, soil C stocks, and soil microorganisms and mesofauna. Soils of the LN peatland stored four times more C and had significantly higher organic matter content, electrical conductivity and cation exchange capacity, but lower nitrate concentration, than soils of the LTQ peatland. The composition and abundance of soil microbial communities, determined by the phospholipid fatty acid (PLFA) method, were similar in the two peatlands. However, mesofauna communities were more abundant and diverse in LTQ than in LN. Our results show that each peatland has its own peculiar characteristics and could, therefore, differ from the other in function and response to disturbance originating from nearby lithium mining. This initial evaluation of peatland soils associated with salt flats in the Puna region represents a first step towards understanding the soil ecology of these ecosystems, and highlights the need for (and the importance of) baseline surveys where lithium extraction is in prospect.

KEY WORDS: Andean peatlands, lithium mining, soil carbon, soil mesofauna, soil microbial communities

## **INTRODUCTION**

Over the last few decades, carbon (C) reservoirs and particularly soils have become highly relevant in the context of global climate change mitigation (Smith 2012, IPCC 2014). Peatlands are very important in the global C cycle because they are thought to store as much as one-third of global soil C, although they cover only around 3 % of the land area (Gorham 1991). However, the increasing pressure on peatlands due to land use changes along with climate change effects could threaten the functioning of these ecosystems and their C stores (Davidson & Janssens 2006, Webster *et al.* 2013).

Peatland ecosystems occur where the rate at which organic matter accumulates exceeds the rate of decomposition, and over a long period of time this results in large quantities of C accumulating in their soils (Rydin & Jeglum 2006). Relatively high groundwater level provides an anoxic environment that results in slower anaerobic decomposition of organic matter by microorganisms (Bernal & Mitsch 2008). Consequently, waterlogged conditions contribute to the accumulation of organic matter forming long-term C deposits (Rydin & Jeglum 2006).

High-elevation peatlands are found in dry mountain ecosystems such as the Tibetan and peri-Himalayan plateaus, and also in the South American Andes. High Andean peatlands form in valley and basin bottoms when the ground surface becomes waterlogged, generally as a consequence of perennial groundwater flow from upland areas (Oyague & Cooper 2020). These systems are surrounded by a wider landscape of bare soil with some sparse grasses and shrubs (Squeo et al. 2006) and can be described as "islands" of biodiversity providing water and food for fauna and the human population (Izquierdo et al. 2018a). They also provide the main regional C store (Maldonado-Fonkén 2014). These characteristics determine their importance as natural and economic resources, mainly for local communities but also for the regional and global population (Izquierdo et al. 2018a, 2018b). This, in turn, makes the future preservation of these peatland ecosystems crucial.



Some High Andean peatlands are closely associated with salt flats, which have the potential to be exploited for their lithium deposits (Izquierdo et al. 2015, 2018b). The extraction of lithium is increasing as a consequence of international industrial demand, mainly related to next-generation technologies (Wanger 2011, Martin et al. 2017) including rechargeable lithium batteries for energy storage which are mostly branded as 'green' alternatives to conventional technologies (Izquierdo et al. 2015). Argentina, Chile and Bolivia together hold one of the largest lithium reserves in the world, and lithium mining is expected to increase in the coming decades (Martin et al. 2017). Lithium extraction affects local hydrology by removing large quantities of water from salt flats (Marazuela et al. 2019), and so could also affect soil processes in adjacent peatlands. At present, little is known about the effects of lithium mining on the ecosystem properties and processes of peatlands associated with the salt flats being exploited. In order to understand the possible consequences of lithium mining on peatlands, it is necessary first to collect a baseline dataset describing the different abiotic and biotic components of these ecosystems, as well as to perform ongoing monitoring to detect changes related to mining activities.

The present study sought to evaluate the baseline soil conditions of two peatlands located on the edge of a salt flat in the Argentine Puna, that is the focus of an imminent lithium mining project. The aims of the study were to analyse and compare between the two peatlands: (1) soil physico-chemical properties up to 15 cm depth; (2) soil organic C stocks up to 30 cm depth; and (3) the composition of soil microorganism and mesofauna communities, along with enzymatic microbial activity.

#### **METHODS**

#### Study area

The study was conducted in Catamarca Province, which is part of the Argentine Puna (Figure 1). This ecoregion is characterised by a dry and cold climate, intense solar radiation and marked daily temperature fluctuations. Mean annual temperature is 8 °C and mean annual precipitation ranges from 100 mm in the north-east to 400 mm in the south-west, with most rainfall concentrated in the summer months from December to March (Burkart et al. 1995). The study area is a high-altitude desert with isolated wetlands such as lakes, salt flats and peatlands surrounded by arid land (Izquierdo et al. 2015, 2016). Peatlands are the key ecosystem of the region, as they are the main provider of water along with other ecosystem services (Izquierdo et al. 2018a). In general, they are located in topographic depressions and commonly waterlogged due to the permanent groundwater supply from upland zones. The combination of high



Figure 1. Location of the study area in the Puna ecoregion, within Catamarca province in Argentina (left). Salt flat associated with lakes and peatlands selected for this study (right). Peatland associated with Laguna Tres Quebradas is located at the northern, and peatland associated with Laguna Negra is located at the southern edge of the salt flat.



soil moisture with low rates of decomposition (due to low air temperatures) results in a particular type of vegetation and soils (Izquierdo et al. 2015, 2018a). The vegetation of peatlands, mainly in stream floodplains and in well-drained areas, is dominated by cushion plants such as Distichia muscoides, Oxychloe andina and Plantago rigida. Cushion plants also include some species of Deyeuxia, Festuca sp., Amphiscirpus nevadensis, Carex sp. and Eleocharis sp. (Izquierdo et al. 2020). Peatland soils are classified as Typic Cryofolist, Typic Cryohemist or hydric Borohemist (Tchilinguirian & Olivera 2012). However, Histosols can be found associated with peatlands and small ravines (Pereyra & Fernández 2019). These types of soils consist of partially decomposed organic material in the surface layers and deeper layers with sand and gravel. They are usually moist and freeze deeply each winter, but have no permafrost (Soil Survey Staff 2014).

We selected two peatlands of about 40 ha, at an altitude of 4,100 m a.s.l. adjacent to a salt flat that is likely to be affected imminently by a lithium mining project. Each peatland was located at the edge of a (different) lake, one peatland being associated with Laguna Negra (hereafter, LN; 27° 39' S, 68° 33' W) and the other associated with Laguna Tres Quebradas (hereafter, LTQ; 27° 22' S, 68° 42' W) (Figure 1). The vegetation of the LN peatland is dominated by grass (Puccinellia sp.) combined with graminoids (e.g. Triglochin sp. and Scirpus sp.), whereas the vegetation cover of the LTQ peatland is dominated by Deyeuxia sp., Amphiscirpus sp. and Scirpus sp., mixed with Oxychloe andina (Phil.) cushions (Figure 2). Water table depth was measured using dipwells at the time of soil sampling, and the values found ranged from 0 to 15 cm below ground level in both peatlands.

#### Sample collection

In November 2017 we collected soil samples at five sampling plots of about 100 m<sup>2</sup>, separated by at least 25-30 m from each other, within each peatland. We collected all soil samples from within the top 30 cm. We decided to focus on the uppermost soil layers because it is at these depths where soil properties and the community of soil organisms are more likely to be seriously affected by land use changes or management practices (e.g. lithium mining). After collection, we stored all soil samples in sealed plastic bags and at 4 °C in the dark until processing. In the laboratory, we measured soil properties as detailed below.

#### Soil properties and C stocks

To describe soil physico-chemical properties, in each plot we collected a composite soil sample, formed by seven subsamples to 15 cm depth, using an open auger of diameter 1.8 cm. In the laboratory, first, we air-dried the soil samples, then we sieved them through a 2 mm mesh, and finally, we homogenised the soil samples using a mortar and pestle. We determined the following properties: pH using soil and distilled water at 1:2.5 ratio (Thomas 1996), electrical conductivity (EC) in a saturated paste of soil (Rhoades 1996) using a Hanna DIST 4 conductivity meter, cation exchange capacity (CEC) determined with 1 M ammonium acetate at pH 7 (Sumner & Miller 1996), organic matter (OM) content using the Walkley-Black technique (Nelson & Sommers 1996), total nitrogen (N) content using the Kjeldahl technique (Bremner 1996), total phosphorus (P) content using the Bray and Kurtz method (Bray & Kurtz 1945), ammonium (NH<sub>4</sub><sup>+</sup>) and nitrate (NO<sub>3</sub><sup>-</sup>) concentrations using the steam distillation method (Mulvaney 1996). We also



Figure 2. Representative images of the two different peatlands associated with Laguna Negra (left) and Laguna Tres Quebradas (right).



calculated the soil C/N quotient. Additionally, in a subsample of each composite soil sample, we estimated soil water content using the gravimetric method (Jarrell *et al.* 1999). We weighed all soil subsamples (fresh weight), dried them in an oven at 105 °C for 48 h, then re-weighed them (dry weight). We calculated soil water content (%) as the percentage weight lost.

To estimate soil organic C stocks, after removing all aboveground plant biomass from the surface, in each plot we collected a composite sample composed of three subsamples taken from discrete depth intervals at 0–5, 5–15 and 15–30 cm, following the same protocol as for the determination of soil physico-chemical properties. We oven dried the samples at 60 °C for at least 48 h. We measured C content (%) using a Perkin Elmer 2400 series II Autoanalyser. We did not correct the C values for inorganic C content because it has been reported that inorganic C concentration is very low in the soils of Andean peatlands (Ruthsatz 1993, Schittek *et al.* 2015). For this reason, the content of inorganic C is not generally considered in C stock calculations.

Due to logistical constraints, to obtain an intact soil sample without losing sampling precision, we estimated soil bulk density using the equation proposed by Post & Kwon (2000):

$$BD = \frac{100}{\frac{\% OM}{0.244} + \frac{100 - \% OM}{MBD}}$$
[1]

where %OM represents organic matter content and MBD is the mineral bulk density (1.64 g cm<sup>-3</sup>). Finally, we calculated the soil organic C stock (Mg ha<sup>-1</sup>) by multiplying the C content (mg g<sup>-1</sup>) by the bulk density (g cm<sup>-3</sup>) and the thickness of each depth interval (cm) for each soil sample (i.e., for each depth interval separately; 0–5, 5–15 and 15–30 cm), as well as for the total soil depth sampled (0–30 cm) by summing the values for all intervals.

#### Microorganisms and mesofauna communities

To describe the composition and to quantify the biomass of active soil microbial communities we used phospholipid fatty acid (PLFA) analysis (Bossio & Scow 1998). In each sampling plot, we collected five subsamples of soil up to 5 cm depth and mixed them to obtain one composite soil sample. Then, we carefully separated roots and sieved them through a 2 mm mesh. We extracted PLFA from 7 g (dry weight) of soil using a mixture of chloroform, methanol and phosphate buffer (1:2:0.8 by volume) and extraction columns in solid phase (Silicic acid Sil350-Sigma). We used a gas chromatograph (Perkin-Elmer Clarus 500 equipped with an Elite-5

Crossbond 5 % diphenyl - 95 % dimethylpolysiloxane column) to identify phospholipid and we compared the results with an internal standard (19:0, methyl-nonadecanoate). As indicators of bacterial PLFA we used the sum of cy17:0, cy19:0, i15:0, a15:0, i16:0 and i17:0, and as fungal PLFA we used 18:2w6 fatty acid (Bossio et al. 2006, Ushio et al. 2008, Zechmeister-Boltenstern et al. 2011). Additionally, in a subsample of the same composite soil sample, we quantified microbial activity using fluorescein di-acetate (FDA) hydrolysis (Adam & Duncan 2001). We incubated 1 g of oven-dry soil with phosphate buffer (0.5 mM, pH=7.6) and 100  $\mu$ L FDA (2 mg mL<sup>-1</sup>) for 30 minutes at 30 °C. After the incubation, we quantified the concentration of fluorescein (which is a coloured end-product released by the action of microbial enzymes) using a spectrophotometer at a wavelength of 490 nm. Microbial activity was expressed as the mass of fluorescein released by microbes present in the 1 g of dry soil per hour, in units of  $\mu g g^{-1} h^{-1}$ .

To analyse the composition of soil mesofauna communities, i.e., invertebrates whose body lengths range from 0.2 to 2 mm, we collected in each plot a composite soil sample up to 5 cm depth. We extracted invertebrates over the course of 5 days with Berlese-Tullgren funnels (1 mm mesh) using 25 W light bulbs, and preserved them in 70 % alcohol. They were subsequently identified at order/suborder level.

#### **Statistical analysis**

To describe soil physico-chemical characteristics of the peatlands across plots, we performed a Principal Component Analysis (PCA). Through this analysis, we obtained two main axes (Axis 1, Axis 2) that summarise the variation in soil properties. To analyse the differences in soil properties and C stocks between peatlands across the total range of soil depth sampled (0–30 cm) and for each depth interval (0–5, 5–15 and 15–30 cm) separately, we used linear models. When necessary, data were log-transformed to meet statistical assumptions.

We also performed PCA to explore the composition of soil microbial communities across plots. To compare the abundance of different microbial groups (PLFA Gram-positive, Gram-negative and Fungi), fungi to bacteria (F/B) quotient and microbial enzymatic activity between peatlands, we produced linear models. Finally, we described the composition and relative abundance of the soil mesofauna communities of the peatlands by constructing rank-abundance curves. All data analyses were performed with the open-source software package R, version 3.5.1 2018 (R Core Team 2019).



#### RESULTS

# Soil physico-chemical properties and soil organic C stocks

PCA showed that plots were clearly separated in the ordination space (Figure 3) with plots of LN peatland located at the positive extreme of Axis 1 (explained 38 % of variance), which was associated with high values of OM, total N, EC and CEC. The plots of LTQ peatland were located at the negative extreme of Axis 1, which was associated with high NO3<sup>-</sup> concentration. Axis 2 (explained 23 % of variance) separated plots with higher soil pH and C/N quotient at the positive extreme from those with higher total K and  $NH_4^+$  at the negative extreme (Figure 3). Both peatlands were found to have alkaline and salty soils (Table 1). LN peatland had significantly higher OM content, organic C concentration, EC and CEC, but lower NO<sub>3</sub><sup>-</sup> concentration, than LTQ peatland. There were no significant differences in C/N, P, K, total N and NH<sub>4</sub><sup>+</sup> content between peatlands (Table 1).

We found that total SOC stored up to 30 cm depth and its distribution across the different depth intervals varied between peatlands. Across the total soil depth sampled (0–30 cm), LN peatland stored four times more SOC (281.8  $\pm$  21.6 Mg ha<sup>-1</sup>, mean  $\pm$ SD) than LTQ peatland (71.5  $\pm$  42.0 Mg ha<sup>-1</sup>) (*F* = 15.99, *p* < 0.0001). The vertical distribution of SOC stored in the different depth intervals also varied between peatlands. At depth 0–5 cm, we did not find significant differences in SOC stored in the two peatlands (Figure 4). However, at 5–15 cm (*F*=9.04, p=0.017) and 15–30 cm (*F*=139.72, p < 0.0001) the C stored in LN peat was significantly higher than in LTQ peat (Figure 4).

# Composition and activity of microbial communities

We found that the composition of soil microbial communities of both peatlands was similar (i.e., there were no significant differences between peatlands in the abundance of PLFA markers measured; Table 2). More than 50 % of PLFA markers were represented by i15:0, a15:0 and cy19:0 (related to bacterial groups, particularly Gram-positive bacteria; Table 2). The results of PCA (Axis 1 and Axis 2 explained 78 % and 11 % of the total variance, respectively) showed that plots with a higher abundance of bacterial PLFA markers were located at the negative extreme of Axis 2, and plots with a higher abundance of fungal PLFA markers were located at the positive extreme of Axis 2 (Figure 5). Finally, we found that enzymatic microbial activity was also similar between peatlands (Table 2).



Figure 3. a) Distribution of the 10 plots in the first two axes of the principal component analysis (PCA) based on the soil physico-chemical properties. b) Contribution of soil variables to the ordination. LN: peatland associated with Laguna Negra, LTQ: peatland associated with Laguna Tres Quebradas. EC: electrical conductivity, OM: organic matter, CEC: cation exchange capacity, C/N: carbon to nitrogen quotient. The percent variability explained by each axis of the PCA is shown (between brackets).



Soil properties	LN	LTQ	F	<i>p</i> -value
рН	8.2 (± 0.22)	8.1 (± 0.32)	1.13	0.320
EC ( $\mu$ S cm <sup>-1</sup> )	7,160 (±2,452)	1,325 (± 798)	26.2	0.001
OM (%)	19.8 (± 4.37)	8.0 (± 3.82)	9.97	0.013
Organic C (mg g <sup>-1</sup> )	115 (± 25.4)	46.5 (± 22.1)	20.8	0.002
Total N (mg g <sup>-1</sup> )	6.1 (± 1.40)	3.1 (± 1.92)	3.35	0.105
Total P (mg g <sup>-1</sup> )	5.4 (± 2.08)	5.9 (± 0.28)	0.36	0.567
Total K (mg g <sup>-1</sup> )	1.6 (± 0.99)	1.8 (± 0.54)	0.08	0.789
C/N quotient	18.9 (± 2.66)	21.2 (± 14.53)	0.00	0.946
CEC (cmol <sub>c</sub> kg <sup>-1</sup> )	27.9 (± 9.43)	15.5 (± 2.36)	6.04	0.039
NH <sub>4</sub> <sup>+</sup> (ppm)	78.5 (± 22.71)	72.5 (± 99.47)	1.53	0.252
NO <sub>3</sub> <sup>-</sup> (ppm)	7.2 (± 2.89)	66.3 (± 39.55)	30.1	0.001

Table 1. Soil physico-chemical properties (mean value  $\pm$  standard deviation, n=5) at depth 0–15 cm in the peatlands associated with Laguna Negra (LN) and Laguna Tres Quebradas (LTQ). EC: electrical conductivity, OM: organic matter, C: carbon; CEC: cation exchange capacity. For soil properties that differ significantly (p<0.05) between peatlands, the values of *F* and *p* are shown in **bold**.



Figure 4. Soil organic carbon (SOC) stocks up to 30 cm depth, comparing three discrete depth intervals between peatlands. LN: peatland associated with Laguna Negra, LTQ: peatland associated with Laguna Tres Quebradas. Different letters indicate significant differences (p < 0.05) in SOC stocks between peatlands for each depth interval separately.

### Composition of invertebrate communities

We collected a total of 533 individuals which were classified into eight taxonomic groups (Table 3). The collections at the LTQ peatland produced three times more individuals and a greater diversity of taxonomic groups than those at the LN peatland (Table 3, Figure 6). The invertebrate communities of both peatlands were dominated by mites (specifically, members of the order Oribatida), which accounted for 96 % of mesofauna abundance in the LN peatland and 67 % in the LTQ peatland (Table 3). In the LTQ peatland, the second most abundant group was Collembola (19%), while insects accounted for 7% (Figure 6). In the LN peatland, the only group present apart from mites (insects) was poorly represented (1.5% of total individuals) (Table 3, Figure 6). Taxonomic richness was unevenly distributed between the studied plots. Both peatlands showed great variability between plots in the number of individuals as well as in the number of taxonomic groups (Table 3), which could indicate high spatial variability in the communities of soil invertebrates.



Table 2. PLFA (phospholipid fatty acid) profiles and concentrations, F/B (fungi/bacteria) quotients and FDA (fluorescein di-acetate) hydrolysis rates (mean values  $\pm$  standard deviation, n = 5; dry soil basis) in soil from two peatlands in the Argentinean Puna, as indicators for the active microbial communities. LN: peatland associated with Laguna Negra, LTQ: peatland associated with Laguna Tres Quebradas. None of the microbial variables differed significantly between peatlands (p > 0.05).

Indiactor	Group of	PLFA	Peatland			
Indicator	microorganisms	markers	LN	LTQ		
PLFA concentration (µg g <sup>-1</sup> )	Gram-positive bacteria	<i>i</i> 15:0	14.3 (± 2.60)	9.2 (± 7.34)		
		<i>a</i> 15:0	17.5 (± 1.45)	13.1 (± 8.23)		
		<i>i</i> 16:0	4.9 (± 1.10)	3.5 (± 2.23)		
		<i>i</i> 17:0	4.1 (± 0.61)	2.9 (± 2.07)		
	Gram-negative bacteria	<i>cy</i> 17:0	5.3 (± 0.94)	3.6 (± 2.14)		
		<i>cy</i> 19:0	17.6 (± 4.30)	10.3 (± 8.12)		
	Fungi	18:2ω6	5.5 (± 1.20)	6.3 (± 2.00)		
F/B quotient			0.1 (± 0.03)	0.4 (± 0.51)		
FDA hydrolysis rate ( $\mu g g^{-1} h^{-1}$ ) 12.3 (:			12.3 (± 1.42)	9.9 (± 3.94)		



Figure 5. a) Distribution of the 10 plots in the first two axes of the principal component analysis (PCA) based on the composition and activity of active microbial communities. b) Ordination of bacterial and fungal phospholipid fatty acid (PLFA) markers (as indicators of microbial biomass and community composition), fungi/bacteria quotient (F/B), and fluorescein di-acetate (FDA) hydrolysis rate (as indicator of microbial activity). LN: peatland associated with Laguna Negra, LTQ: peatland associated with Laguna Tres Quebradas. The percent variability explained by each axis of the PCA is shown (between brackets).



Taxon	LN peatland plots			LTQ peatland plots						
	А	В	С	D	Е	F	G	Н	Ι	J
Oribatida	5 (0.72)	4 (1)	116 (0.97)	1 (1)	1 (1)	0	12 (0.67)	50 (0.33)	19 (1)	189 (0.89)
Hemiptera	0	0	0	0	0	0	2 (0.11)	0	0	2 (0.01)
Diptera (L.)	1 (0.14)	0	0	0	0	0	0	4 (0.03)	0	0
Collembola	0	0	0	0	0	0	1 (0.06)	76 (0.50)	0	1 (0.01)
Prostigmata	1 (0.14)	0	2 (0.02)	0	0	0	2 (0.11)	8 (0.05)	0	14 (0.07)
Coleoptera (L.)	0	0	0	0	0	0	1 (0.06)	8 (0.05)	0	1 (0.01)
Coleoptera	0	0	0	0	0	0	0	1 (0.01)	0	0
Thysanoptera	0	0	1 (0.01)	0	0	0	0	4 (0.03)	0	4 (0.02)
Psocoptera	0	0	0	0	0	0	0	1 (0.01)	0	1 (0.01)
Total individuals (N)	7	4	119	1	1	0	18	152	19	212
Total taxa (S)	3	1	3	1	1	0	5	8	1	7

Table 3. Results of the analysis of mesofauna communities: number (and proportion) of individuals belonging to each taxonomic group, total number of individuals (N) and total number of taxonomic groups (S) registered per plot in Laguna Negra (LN) peatland (A–E) and Laguna Tres Quebradas (LTQ) peatland (F–J). L. = larvae.





Figure 6. Rank abundance curves of mesofauna taxonomic groups collected in the peatlands associated with Laguna Negra (LN) and Laguna Tres Quebradas (LTQ). Values are plotted on a logarithmic scale of the proportional abundance  $(p_i)$  for each taxonomic group. Key to abbreviations: Orib. = Oribatida mites; Coleop. = Coleoptera; L. Coleop. = Coleoptera larvae; Thys. = Thysanoptera; L. Dipt. = Diptera larvae; Prost. = Prostigmata mites.

#### DISCUSSION

To our knowledge, this is one of the first studies to integrate baseline information of soil physicochemical properties, C stocks and the composition of soil microbial and mesofauna communities in highaltitude peatlands in the Andean Puna. The two peatlands are located at similar altitudes and in the same hydrological catchment, and are associated with the same salt flat. However, our results for the soil variables evaluated showed marked differences between the sites as well as intra-site variability, which could indicate spatial differences in soil functioning. This pattern of dissimilarity between peatlands has been found in other studies focusing on different taxonomic groups (Izquierdo et al. 2020), vegetation (Ruthsatz et al. 2020) and macroinvertebrates (Nieto et al. 2016, 2017), which showed the pattern to be closely related to variations in salinity. We emphasise that the information obtained here could be relevant not only to mining companies, but also to the policymakers who regulate the lithium industry in the region.

In the two peatlands of the present study, our data showed a site-specific variation in soil C stored. We evaluated the C store across plots in the LN peatland at 253–311 Mg ha<sup>-1</sup>; these values are comparable to

those reported by Segnini et al. (2010) for similar systems in Peru (230–300 Mg ha<sup>-1</sup>). However, soil C storage in the LTQ peatland is four times lower, ranging from 13 to 128 Mg ha<sup>-1</sup> across plots. These values are more similar to published data for the Colombian Páramos (38–51 Mg ha<sup>-1</sup>, Benavides et al. 2013). In addition, the distribution of C content (%) with depth differed between the LN and LTO peatlands. In the LN peatland we quantified an average of 14.0, 14.3 and 12.6 % C, while in the LTQ peatland we found 6.1, 2.8 and 0.6 % C at 0-5, 5-15 and 15-30 cm depth, respectively. These results suggest that these systems can vary significantly in their capacity to store C in their soils. This variability could be related to plant community and soil mineral composition (Hribljan et al. 2016, Polk et al. 2019). Vegetation generates organic material not only from aboveground tissues but also from root biomass, and it has been observed that soil organic matter content is strongly related to the composition of plant communities (Benavides 2013, Polk et al. 2019). In the peatlands studied, we observed that the vegetation communities are dominated by different species of graminoids and grasses, which could influence plant litter input to the soil, soil structure and rhizosphere processes, thus determining the differences in soil C content observed (Rasse et al. 2005, Cotrufo et al. 2013, Wilson et al. 2018). Furthermore, differences in wind-blown mineral input from higher topographic areas of the catchments could influence the accumulation of sediment in the soil profile (Hribljan et al. 2016, Polk et al. 2019, Favaro et al. 2020), differentially affecting the soil mineral and organic composition of each peatland. Future studies are needed to understand the relationship between the composition of plant communities, soil mineral characteristics, and the down-profile variation in soil C in these peatlands.

Bacteria and fungi contribute more than 90 % of the microbial biomass in soils and are the main agents driving the decomposition of organic matter and soil C accumulation (Soares & Rousk 2019). Different studies have shown that the composition of microbial communities is strongly associated with the rate of nutrient cycling, thus they have been proposed as indicators of ecosystem functioning (Van Der Heijden et al. 2008, De Vries et al. 2012). In this study we found that, although both peatlands had a similar composition of microorganisms, the LN peatland tended to have a higher microbial biomass than the LTO peatland, mainly composed of bacterial groups. Although fungi accounted for less than 5 % of the total microbial biomass in both peatland soils, we found a trend towards higher fungal biomass and F/B quotient in LTQ peatland than in LN peatland.



The small dissimilarities could imply differences in nutrient cycling between the peatlands. It has been shown that fungal-dominated communities are associated with highly conservative nutrient cycling because they immobilise available N more efficiently than bacteria (De Vries *et al.* 2011). The higher soil nitrate concentration in the LTQ peatland could be related to the dominance of fungi in the microbial community; however, specific studies are needed to elucidate this relationship. Shifts in soil microbial communities should be permanently monitored due to their sensitivity to soil disturbance, possibly affecting not only the processes in which they are involved but also higher trophic levels in the food web (De Vries *et al.* 2013, George *et al.* 2017).

In the present study we also characterised soil mesofauna as an indicator of soil functioning since they are involved in the decomposition process, nutrient cycling and soil formation (Lavelle et al. 2006, Barrios 2007). Our results showed that the communities of mesofauna were more abundant and diverse in LTQ peatland than in LN peatland. In both peatlands Acari (mainly order Oribatida) was the most abundant group, as found previously in peatlands from Chile (Covarrubias 2009), indicating the importance of organic matter content and humidity conditions for these organisms (George et al. 2017). Although Collembola is another abundant group within the soil mesofauna (Briones 2014), this taxon was found only in the LTQ peatland. Collembolans are important consumers of biofilms and fungal hyphae (Rusek 1998), thus the higher abundance of these organisms in the LTQ peatland is probably related to a higher proportion of fungal biomass. Moreover, the higher diversity of Insecta in the LTQ peatland could be associated with lower soil salinity when compared to soil from the LN peatland. Additionally, our results showed substantial intra-site variability with differences in the abundance and diversity of mesofauna between plots in each peatland. This pattern highlights the complexity of edaphic processes and their relationship with different ecosystem properties such as vegetation, hydrology and soil salinity. Nevertheless, the relationships between the abundance of different mesofauna groups and microbial composition, as well as soil properties, need further analysis. Soil mesofauna make up a large component of soil biological communities, and are involved in important soil processes. This group of organisms could be used as indicators of edaphic habitat quality (Gerlach et al. 2013). In particular, oribatid mites which have slow growth rates, low reproductive rates and limited mobility - are highly sensitive to environmental disturbance (Hansen 2000).

Therefore, they could be used as bioindicators of the stability of soil communities (George *et al.* 2017).

In summary, this work represents a first step towards understanding soil abiotic and biotic relationships in peatlands of the Puna region. Our results show that each peatland could function as a separate unit with its own peculiar characteristics and processes, and could therefore respond differently to human disturbance. Mining activities could significantly affect soil properties in these peatlands, and could affect soil C storage in the shortand long-term as shown by other authors (Glina et al. 2019). Although further studies are needed to identify particular relationships and mechanisms between the soil biota and soil processes related to C cycling, the present study adopted an integrated approach to the study of soil ecology in these ecosystems. The findings of this study emphasise the need for monitoring of soil communities and C stocks in order to understand the ecological consequences of changes in soil properties under different land use change scenarios.

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#### **AUTHOR CONTRIBUTIONS**

MFC, MVV and AEI: conceptualisation, sampling design, methodology and investigation; MFC and MVV: processing of soil samples and data analysis; AEI: funding acquisition; MFC, MVV and AEI: writing original drafts, reviewing and editing subsequent versions of the manuscript.

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