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# The microbial communities and potential greenhouse gas production in boreal acid sulphate, non-acid sulphate, and reedy sulphidic soils

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## HIGHLIGHTS

- Boreal acid sulphate soils contain large amounts of organic C and N in subsoils.
- Microbial communities throughout the acid sulphate soil profile were analysed.
- Rich microbial communities in the deepest Cg2 horizon were found.
- High emissions of GHG could occur when high water table is lowered because of arable farming.

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## ABSTRACT

Acid sulphate (AS) soils along the Baltic coasts contain significant amounts of organic carbon and nitrogen in their subsoils. The abundance, composition, and activity of microbial communities throughout the AS soil profile were analysed. The data from a drained AS soil were compared with those from a drained non-AS soil and a pristine wetland soil from the same region. Moreover, the potential production of methane, carbon dioxide, and nitrous oxide from the soils was determined under laboratory conditions. Direct microscopic counting, glucose-induced respiration (GIR), whole cell hybridisation, and extended phospholipid fatty acid (PLFA) analysis confirmed the presence of abundant microbial communities in the topsoil and also in the deepest Cg2 horizon of the AS soil. The patterns of microbial counts, biomass and activity in the profile of the AS soil and partly also in the non-AS soil therefore differed from the general tendency of gradual decreases in soil profiles. High respiration in the deepest Cg2 horizon of the AS soil (5.66 μg C $g^{-1}$ $h^{-1}$, as compared to 2.71 μg C $g^{-1}$ $h^{-1}$ in a top Ap horizon) is unusual but reasonable given the large amount of organic carbon in this horizon. Nitrous oxide production peaked in the BCgc horizon of the AS and in the BC horizon of the non-AS soil, but the peak value was ten-fold higher in the AS soil than in the non-AS soil (82.3 vs. 8.6 ng N $g^{-1}d^{-1}$). The data suggest that boreal AS soils on the Baltic coast contain high microbial abundance and activity. This, together with the abundant carbon and total and mineral nitrogen in the deep layers of AS soils, may result in substantial gas production. Consequently, high GHG emissions could occur, for example, when the generally high water table is lowered because of arable farming.

## 1. Introduction

The Baltic coasts of Finland contain the largest area of acid sulphate (AS) soils in Europe, with approximately 130,000 ha of agricultural land meeting the AS soil criteria of the U.S. Soil Taxonomy system (Yli-Halla et al., 1999). When used for arable farming, AS soils are often drained to lower the generally high water table and to maintain aeration in the topsoil. AS soils in Finland are usually drained with subsurface pipes installed to a depth of about 1.0–1.2 m. This practice exposes large volumes of previously waterlogged soils to oxygen and promotes oxidation of various reduced substances, including sulphides, leading to sulphuric acid formation, which in turn causes acidification of drainage waters. Oxidation processes result in a number of on- and off-site effects (see Šimek et al., 2011).

One potentially serious off-site effect of AS soils is the formation of increased amounts of microbial gaseous metabolites. Subsoils of boreal AS soils can contain large amounts of organic matter and mineral nutrients including nitrogen (Yli-Halla, 1997; Joukainen and Yli-Halla, 2003; Paasonen-Kivekäs and Yli-Halla, 2005; Šimek et al., 2011). Under natural conditions, these stocks of organic carbon and nitrogen in the waterlogged subsoils are well protected against microbial decomposition and

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mineralisation. Under the conditions of temporary and/or permanent aeration, however, decomposition of organic matter will be accelerated. It is then likely that enhanced microbial activity in the AS soils will lead to accumulation of gases in the soil matrix and consequently leads to increased emissions of these gases from the soil and into the atmosphere. These emissions include two significant greenhouse gases (GHGs), namely nitrous oxide and carbon dioxide.

In addition to the presence of organic substrates and oxygen, the prerequisite for increased microbial activity in soils is a potent microbial community. Our previous research (Šimek et al., 2011) detected unexpectedly high abundances of culturable microbial cells in the C horizon of the boreal AS soil, occurring ca. 1.2 m below the soil surface, and very high basal and glucose-induced respiration in soil samples incubated under oxic conditions. It is therefore likely that the abundant carbon and mineral nitrogen stocks in the deep horizons of AS soils serve as substrates and nutrients for enhanced aerobic microbial growth when oxygen diffuses into these horizons. We concluded that mineralisation of organic matter in the subsoil of AS soils is likely to result in large emissions of $CO_2$ and $N_2O$ (Šimek et al., 2011). Because microbially mediated methane production is a strictly anaerobic process (Wang and Patrick, 2000), temporary and/or permanent aeration of subsoil horizons of AS soils would be expected to decrease methane emissions. Methane emissions are decreased by methane oxidation in topsoil as well. Denmead et al. (2010) found that methane emissions from drained AS sugarcane fields were of the same order of magnitude as the emissions from comparable non-acid fields in Australia.

To our knowledge, the above-mentioned findings represent the first evidence of a rich microbial community in the subsoil of boreal AS soil and the first evidence of the community's theoretical potential to produce large amounts of gases. Several recent studies found larger $N_2O$ emissions from AS soils than from non-AS soils in Australia (e.g., Macdonald et al., 2008, 2011; Denmead et al., 2010), and a fluctuating water table was generally found to increase $N_2O$ emission from soils (e.g., Jiao et al., 2006; Peng et al., 2011). These studies, however, were all conducted in warmer climates.

Restoring the previously drained AS soils to wetlands by inundation (Johnston et al., 2009) or by raising the groundwater table where cultivation is locally vital (Joukainen and Yli-Halla, 2003; Anda et al., 2009; Indraratna et al., 2005) is generally considered effective for mitigating the environmental off-site effects of AS soil on water bodies. Although these mitigation methods inhibit the diffusion of oxygen into the sulphide-bearing horizons, the development of anaerobic conditions may be conducive to emissions of GHGs, especially methane and nitrous oxide, from waterlogged soil.

In this study, we further tested the hypothesis that the subsoil horizons of a boreal AS soil support a rich microbial community. The data obtained from a drained AS soil were compared with those from a drained non-AS soil as well as from a pristine wetland soil from the same region. The pristine soil is at the sea level where sulphidic materials currently accumulate just below the decaying reed residues (Yli-Halla et al., 2008).

## 2. Materials and methods

### 2.1. Study area and soils

The soils under study (one AS soil, one non-AS soil, and one pristine reedy soil) were all sampled on the research farm of the University of Helsinki. The area, described in detail by Mokma et al. (2000), is emerging from the Baltic Sea as a consequence of the post-glacial isostatic rebound at a rate of about 3 mm per year. The mean annual air temperature in the region is 5 °C (30-year mean, 1971–2000), the average annual precipitation is 650 mm, and the average duration of permanent snow cover is about 100 days.

The AS soil is in agricultural use (location *Patoniitty*: 60°13′ N, 25°0′ E, elevation at sea level). It was classified as a Sulfic Cryaquept (Soil Survey Staff, 2010) and is a representative of the AS soils on the coasts of Finland. The field was poldered in the 1950s and artificially drained in 1958 with subsurface pipes installed to a depth of about 1 m. Because of drainage, some of the sulphidic materials have been oxidised resulting in a current minimum pH of 3.8 at the depth of 72–102 cm. The field is used for growing grasses in rotation with small grains.

The other sulphidic soil (Histic Sulfaquent; referred to as R soil or reedy soil here) is in a pristine area under a dense, reedy vegetation (*Phragmites communis*), where sulphidic materials are currently accumulating below the abundant decaying plant residues. The site is at the average sea level. At the time of sampling, the site was covered with 20 cm of brackish water.

The non-AS soil (location *Alaniitty*: 60°13′ N, 25°1′ E, elevation about 1.5 m above average sea level) is located about 1000 m from the AS soil and was classified as a Typic Cryaquept. The non-AS soil represents a poorly drained soil type formed in fine-grained sediments and is common in the area. The field, which is used as pasture, is drained with subsurface pipes, and the level of groundwater is usually at 85–100 cm depth. Selected properties of the three soils are presented in Table 1.

### 2.2. Soil sampling

The chemical and microbial properties were determined in soil samples taken from several soil horizons to about 2 m depth on 22 September 2010. Three locations were randomly selected at each of the three sites. Soil was sampled using a core sampler (split tube sampler, Eijkelkamp, The Netherlands), equipped with transparent PVC liner (diameter 5 cm, length 30 cm). After the top soil core (depth 0–30 cm) was collected, the liner was replaced, the sampler was carefully inserted into the hole made during sampling of the top core, and the next soil core was sampled, and so on. Samples from the horizons below the groundwater table were collected in the same way, and from the previously made holes, with a sediment sampler (piston sampler, Eijkelkamp, The Netherlands). Three individual and randomly selected soil samples, formed of several sub-sequential 30-cm cores, were sampled from each soil. All samples were transported immediately to the laboratory, and the soil cores were cut according to the major soil horizons; four horizons (AS soil and non-AS soil) or two horizons (R, reedy soil) were selected for analyses. The subsamples for mineral nitrogen analyses were stored at −20 °C, and those for the determination of biological properties were stored in plastic bags at 4 °C; the analyses were performed during a 3-month period. The redox-potential measurements of selected soil horizons were carried out independently in soil pits concurrently with soil classification (AS in 2011, non-AS in 2009, and R in 2011). Variations of redox-potential related to the different dates of sampling and subsequent analyses can be ignored in subsoil horizons that were permanently waterlogged.

### 2.3. Determination of soil physical and chemical properties

Soil organic carbon ($C_{org}$) and total nitrogen ($N_{tot}$) were determined by dry combustion at 900 °C (Vario Max CN macro elementar analyzer, Elementar Analysensysteme GmbH, Germany) with 0.5 g of air-dried and crushed subsamples (method ISO 11464). Because these soils are non-calcareous, all carbon was assumed to be organic. Mineral nitrogen ($NH_4^+$–N and $NO_3^-$–N) was extracted from frozen, thawed subsamples with a KCl solution (2 M, 1:2.5 w/w; Esala, 1995) and was determined spectrophotometrically with a flow injection autoanalyser (AIM 1250 Autosampler and LaChat QuikChem 8000, Lachat Instruments, USA).

The redox potential of soil horizons was measured with a Pt electrode and a calomel or silver/silver chloride reference electrode (90-01, Orion, USA and P5E, Jensen Instruments, USA) in situ, and fresh soil pH and temperature were measured at the same time (IQ 160 ISFET pH meter with a built-in thermistor, PHW37-SS, Hach