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Impact of salinity on soil microbial communities and the decomposition of maize in acidic soils

Jannike Wichern, Florian Wichern *, Rainer Georg Joergensen

Department of Soil Biology and Plant Nutrition, University of Kassel, Nordbahnhofstr. 1a, 37213 Witzenhausen, Germany

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Abstract

Soil salinity, as an increasingly important process of land degradation, is a major threat to microbial communities and thus strongly alters organic matter turnover processes. This study was conducted to determine the influence of salinity on the decomposition of maize and on the response of soil microbial communities. Soil samples were collected from two pasture sites in Heringen (Germany). One of the sites has previously been influenced by salinity caused by saline effluent from a potassium mine. These sandy soils were washed, resulting in equal levels of electrical conductivity. Moist soils were then incubated with 2% incorporated maize straw and at three levels of salinity (0, 15, 50 mg NaCl g^{-1} soil) for almost 7 weeks at 25 °C. The amount of recovered maize derived particulate organic matter (POM) increased with increasing salinity, exhibiting reduced decomposition of substrate. Furthermore, inorganic N, which consisted almost exclusively of NH_4^+, increased with increasing levels of salinity. Corresponding to this, biological indices like soil respiration and microbial biomass decreased with increasing levels of salinity, underlining the detrimental effect of salinity on soil microorganisms. This effect was reduced after addition of maize straw, documenting the importance of organic matter amendment in counteracting the negative effects of salinity on microbial communities and related mineralisation processes. Addition of organic matter also led to a spatial differentiation of the microbial community in the soil, with bacteria dominating the surface of the substrate, indicated by a low glucosamine-to-muramic acid ratio. This ratio, however, was not altered by salinity. On the other hand, the ergosterol-to-microbial biomass C ratio was an evidence of fungal dominance in the soil. The ratio increased with elevated salt content, either showing a shift towards fungi, a change in fungal cell morphology, or accumulation of ergosterol in the soil. The metabolic quotient qCO_2 was higher in the soil previously subjected to osmotic stress, showing a physiologically more active population that is using substrate less efficiently. We assume that it might further reflect adaptation mechanisms to the increased osmotic pressure.

Keywords: Amino sugars; Fungi-to-bacteria ratio; Microbial activity; Osmotic stress; Physiological adaptation; Salinisation

1. Introduction

Soil salinity is part of natural ecosystems under arid and semi-arid conditions (Pathak and Rao, 1998), and an increasing problem in agricultural soils throughout the world (Keren, 2000; Qadir et al., 2000). In temperate humid climates soil salinity occurs on a smaller scale, mainly in salt marshes, along roads, and on saline waste dumps.

The influence of salt as a major stress to soil microorganisms, which is more potent than that of heavy metals (Sardinha et al., 2003), has been the subject of several studies (e.g. Sarig and Steinberger, 1994; Pankhurst et al., 2001; Mamilov et al., 2004). A decrease in CO_2-evolution, enzymatic activity, or microbial biomass has often been observed (e.g. Laura, 1974; Pathak and Rao, 1998; Rietz and Haynes, 2003). Increasing salinity thus has detrimental effects on biologically mediated processes in the soil, such as C and N-mineralisation (Pathak and Rao, 1998). In saline soils and under drought, microbes suffer from osmotic stress, which results in drying and lysis of cells.

Despite this, soil microorganisms have the ability to adapt to or tolerate osmotic stress caused by drought or salinity, especially when regularly confronted with such conditions (Sparling et al., 1989). There are also examples of microbes thriving in strongly saline ponds (e.g. Casamayor et al., 2002), documenting the evolutionary potential of microorganisms. However, fungi tend to be sensitive to salt stress, indicated by

* Corresponding author. Tel.: +49 5542 981664; fax: +49 5542 981596.
E-mail address: fwichern@uni-kassel.de (F. Wichern).

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decreasing ergosterol contents in the soil (Sardinha et al., 2003). Van Bruggen and Semenov (2000) reported that long-term stress results in decreasing fungal diversity. On the other hand, Killham (1994) mentioned that filamentous fungi are highly tolerant to water stress. They nevertheless have to cope with increasing osmotic pressure and thus might change their physiology (Killham, 1994) and morphology in response to this (Zahran, 1997). Killham (1994) describes two main adaptation strategies of microorganisms to osmotic stress, such as salinity, drought or freezing, all of which result in accumulation of solutes in the cell to counteract the increased osmotic pressure. One is to selectively exclude the incorporated solute (e.g. Na^+, Cl^-) and thus accumulate other ions necessary for metabolism instead (e.g. NH_4^+). The other adaptation mechanism of the cell is to produce organic compounds which will antagonise the concentration gradient between soil solution and cell cytoplasm. This adaptation ultimately results in a physiologically more active microbial community and consequently in a lower substrate use efficiency. However, these mechanisms are known from single microorganisms but have hardly ever been studied on a community level. The metabolic quotient as the specific respiration of a resting microbial community (Anderson and Domsch, 1985) provides a suitable tool for investigating the physiological reaction of a microbial community under osmotic stress.

Therefore, we conducted an incubation experiment to study the response of soil microbial communities *in situ* to elevated salinity and the effects on soil microbial properties and related processes. We hypothesised that (1) increasing salinity decreases soil microbial biomass and activity and thus the turnover of organic matter, and that (2) elevated salinity would lead to a stronger decline of fungi than bacteria. We assumed (3) that soil microorganisms in the soil previously prone to salinity are adapted to salt stress, which results in (4) a higher mineralisation of organic matter in comparison to the other soil.

2. Materials and methods

2.1. Soil sampling and conditioning

Soil samples were taken from two grassland sites with low pH used as meadow. They were situated in the flood plain of the river Werra close to Heringen in North Hessia, Germany, and were approximately 100 m apart. At one site (A) plants showed no symptoms of salinity, whereas the other site (B) has been prone to salinity to a high extent, which was illustrated by no growth of plants and establishment of highly salt tolerant plant species on a neighbouring site (Sardinha et al., 2003). Soil type at site A was classified as a Dystric Fluvisol, whereas soil type at site B was a Salic Fluvisol (Sardinha et al., 2003). Monthly rainfall varies between less than 10 and more than 150 mm, with an average annual rainfall of about 690 mm. For more than 100 years, liquid saline waste from a nearby potassium plant has been injected into underground geological formations that emerged at different sites in this area. The salt was predominantly NaCl (Sardinha et al., 2003). Site A was dominated by *Alopecurus pratensis* L., *Poa pratensis* L., and *Arrhenatherum elatius* L., whereas at site B no vegetation could be detected, but particulate plant residues were present. At a distance of approximately 2 m to site B, small patches of the halophyte *Spegularia salina* L. were observed. At both sites, five core samples (100 cm^{-3}) were taken at 0 to 5 cm depth in May 2004 to determine bulk density. In addition, a larger bulk of soil was taken at 0 to 10 cm depth, put into polyethylene bags, transferred to the laboratory in a cooled box immediately, sieved wet (<2 mm) and stored at 4 °C until further processing. Both soils were washed repeatedly to reduce the salt content until the electrical conductivity in the extract was at a low level in the salt affected soil (site B) similar to that in the soil from site A. We assumed that the washing procedure did not influence the microbial biomass, as most soil particles with adhering microorganisms were recovered after washing.

2.2. Experimental set-up

An incubation experiment was conducted as a three-factorial design with four replicates of each treatment. The washed soils from sites A and B were weighed (120 g) into 1 litre stoppered Pyrex jars and incubated either with or without 2% (on an oven-dry basis) maize straw (42.9% C, 1% N) addition at three different levels of added salt (0, 15, 50 mg NaCl g^{-1} soil, corresponding to a calculated electrical conductivity of approximately 1, 8 and 24 dS m^{-1}). Maize straw was thoroughly mixed into the soil samples. Samples were then incubated for 47 days at 25 °C and approximately 60% water holding capacity in the dark. Soil respiration was measured after 2, 4 and 7 days after incubation commenced and thereafter every 7 days. Then, the evolved CO_2 was captured in 5 to 15 ml 1 M NaOH and titrated with 1 M HCl after addition of $BaCl_2$ and phenolphthalein indicator solution. The maize derived CO_2–C (μg d^{-1} g^{-1} soil) was estimated by subtracting the soil respiration of the control treatments from the respiration in the respective maize treatments, with the assumption that no priming effects occurred. The specific CO_2 evolution of the microbial biomass, the metabolic quotient (qCO_2), was calculated from the basal respiration as follows: (μg CO_2–C d^{-1} g^{-1} soil evolved during the last 12 days of incubation in the control samples)/(μg microbial biomass C g^{-1} soil at the end of the incubation experiment) × 1000 = mg CO_2–C g^{-1} microbial biomass C d^{-1}. At the end of the experiment, sub-samples of each treatment were analysed for soil organic C and total N, microbial biomass C and N, ergosterol content and inorganic N.

2.3. Measurement of soil chemical and physical properties

Clay content was determined by a pipette method after pretreatment with H_2O_2 to remove organic matter (Gee and Bauder, 1986). The pH of the soil was determined before the incubation experiment in water and in 0.01 M $CaCl_2$ using a soil-to-solution ratio of 1:2.5. The cation exchange capacity of the original soils was measured according to Ryan et al. (1996) in sodium acetate solution and in unbuffered 0.1 M $BaCl_2$ solution (Schlichting et al., 1995) as were the washed soils. Exchangeable cations were measured by atomic absorption spectrometry (M series,