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Effects of sewage sludge addition to Norway spruce seedlings on nitrogen availability and soil fauna in clear-cut areas

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ABSTRACT

Anaerobically digested and composted sewage sludge (CSS) has been suggested to be a slow-release fertilizer in forestry and an alternative to quick-release inorganic fertilizers. The effects of CSS with or without added carbohydrate on inorganic nitrogen availability and on soil animals were tested in two Norway spruce plantations. Half of the seedlings were individually fertilized with CSS, and the rest were left as controls. Solid sucrose was added to half of the fertilized and untreated seedlings. Soil samples were taken in the autumn in the first and the second year after the treatments. CSS increased soil NH_4-N (2100%), the proportion of soil NO_3-N , and the N concentration of spruce needles. CSS greatly reduced the abundances of enchytraeids, tardigrades and collembolans, but increased the proportion and abundance of bacterial-feeding nematodes irrespective of carbohydrate addition. A better stabilization method needs to be developed before CSS can be used as a forest fertilizer.

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1. Introduction

Waste water treatment produces large amounts of sludge. Land application of unhandled sewage sludge is a common method of disposal in agroecosystems, but legislation has prevented its use as a forest fertilizer in several countries. Some countries have no clear legislation for the soil amendment of sludge in silviculture (Rantanen et al., 2008; European Commission, 2001). Preliminary treatment is necessary before sewage sludge can be used as a fertilizer. Here we focus on the possible use of anaerobically digested and composted sewage sludge (CSS) in forestry.

Forestry is largely based on clear-cutting and artificial regeneration in Northern Europe. For example, in 2010 forest regeneration was carried out by planting on 84,000 ha in Finland, and Norway spruce was the most commonly planted species (>100 million seedlings). Despite its potential profitability, forest fertilization has not been a common practice (Saarsalmi and Mälkönen, 2001). Legislation may be altered so as to permit the use of organic wastes in forest fertilization. Because there is a great potential for land application of organic waste in spruce plantations, the effects of CSS on forest ecosystem components and processes need to be tested.

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In an early experiment conifers grew better than other tree species in soil amended with sewage sludge, though weed competition was considered a problem (Zasoski et al., 1983). According to Prescott and Blevins (2005), biosolids promoted conifer growth as effectively as chemical fertilizers. CSS increased the growth of pine shoots, but reduced the root/shoot ratio (Selivanovskaya and Latypova, 2006). According to Selivanovskaya and Latypova (2006), heavy metal concentrations of soil and pine seedlings did not exceed allowable limits. Selivanovskaya and Latypova (2006) also found that composted sewage sludge increased microbial biomass in a forest nursery. On the other hand, biosolid amendments elevated metal concentrations in soil even after 25 years (Cline et al., 2012). However, the heavy metal concentrations of sewage sludge have greatly decreased in the last two decades in Scandinavian countries. For example, in Finland the heavy metal concentrations of sludge are low, and limits are strict (21% of EC directives, Rantanen et al., 2008).

Collembolans have been used as indicators in assessing the risks of sewage sludge application (Cole et al., 2001). However, little is known on the effects of CSS on soil animals in natural ecosystems. Oribatids were sensitive to sewage sludge in a Mediterranean forest soil (Andrés et al., 2011). Huhta et al. (1979) found that soil organisms rapidly colonized sewage sludge mixed with crushed bark on arable soil. Predictions of CSS effects can also be based on studies using other forms of N fertilization. High doses of nitrogen







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fertilizers can have negative effects on an important mesofaunal species, *Cognettia sphagnetorum* (Huhta, 1984; Lohm et al., 1977). Collembolans were in general more sensitive to NH₄NO₃ than urea in pine forest soil, but the effects were species-specific (Vilkamaa and Huhta, 1986).

Bacterial- and fungal-feeding nematodes are useful indicators of their respective food sources (Sánchez-Moreno et al., 2006). In particular, the proportion of bacterial-feeding nematodes provides information on the dominance of the bacterial-based food chain. Urea increased the abundance of bacterial-feeding nematodes in a Scots pine forest soil at least partly because higher pH favoured bacteria (Hyvönen and Huhta, 1989).

In energy-limited conditions microbes use N-containing organic compounds as energy sources, and excess N is released as ammonium (Nieminen, 2008a; Smolander et al., 2001). Thus, N fertilization in a C-limited system probably increases the inorganic N pool. On the other hand, labile C represses the production of protease and other enzymes at the cellular level (Geisseler et al., 2010). Sufficient labile C availability is therefore essential to prevent N leaching in forest ecosystems fertilized with nitrogen. Carbon resource amendments such as wood chips or sucrose have been used to reduce soil inorganic N pools to prevent N leaching in real and model ecosystems (Eschen et al., 2007; Homyak et al., 2008; Miller and Seastedt, 2009; Nieminen, 2009; O'Connell et al., 2004). Labile C additions to C-limited systems can also increase the biomass of organisms higher in the soil food web, such as nematodes (Nieminen and Setälä, 2001) and enchytraeids (Nieminen, 2008b). However, woody mulch decomposes slowly and it can reduce the growth of spruce seedlings (Hallsby, 1995). A soluble carbohydrate addition can be suggested to have fewer undesirable effects than woody mulch.

In the present study we investigated if it is safe to use composted sewage sludge as a fertilizer in Norway spruce forests. In particular, we asked if carbohydrate addition can retard the release of inorganic N from CSS, and lessen possible side-effects of high N concentrations on soil biota. The short-term effects of composted sewage sludge with or without carbohydrate (sucrose) addition on the inorganic soil nitrogen pools, the N uptake of spruce seedlings and the soil animal abundances were tested in clear-cut Norway spruce forests. We hypothesized that CSS would increase the inorganic N availability in forest soil and the N uptake by Norway spruce seedlings, but that sucrose addition would reduce inorganic N pools. Because enchytraeids, mites and collembolans seem to be sensitive to high levels of nitrogen, we hypothesized that CSS would reduce the abundances of these groups in contrast to, for example, bacterial-feeding nematodes. Finally, we expected that sucrose addition would alleviate the effects of compost on soil animals.

2. Materials and methods

2.1. Study sites and experimental design

The experiment was established at the end of June 2009 in two Norway spruce forests situated ca. 100 km apart in Viitasaari ($63^{\circ}14^{\circ}N$, $26^{\circ}5'E$) and in Åhtäri ($62^{\circ}43^{\circ}N$, $24^{\circ}18^{\circ}E$), Central Finland. Both forest sites were clear-cut in the previous winter. The soil type is Haplic podzol. The biological, physical and chemical properties of the sites have been described in Nieminen et al. (2012). Meteorological data are given in Table 1.

The experimental area at each site was divided into 3 plots (15 m*15 m each) in which the treatment replicates were interspersed. The experimental unit in our study is a Norway spruce seedling and the rhizosphere soil associated with it. Norway spruce seedlings were planted into each plot in a regular grid using the planting devices M-Planter and Bräcke. Half of the seedlings were fertilized individually by adding CSS (15 kg (seedling)⁻¹, equalling 20 Mg ha⁻¹) as surface mulch to circular tree-centred patches (diam. 1 m). A circular (20 cm diam.) untreated area was left around each seedling to avoid salinity effects. Solid sucrose (262 g (seedling)⁻¹, equalling 1 Mg C ha⁻¹) was then added to half of the fertilized and non-fertilized seedlings. Each of the four possible treatment combinations (control

Table 1

Meteorological data for the study sites (Finnish Meteorological Institute).

	Viitasaari	Ähtäri
Long-term mean annual temperature (°C)	3.3	2.7
Long-term mean annual precipitation (mm)	604	632
Mean annual temperature (°C) 2009	3.8	3.2
Mean annual temperature (°C) 2010	2.4	1.3
Mean temperature (°C) during growing season 2009	13	12
Mean temperature (°C) during growing season 2010	13.9	13.5
Precipitation (mm) 2009	421	492
Precipitation (mm) 2010	588	633

without amendments, sucrose addition, compost addition, combined sucrose and compost addition) was replicated three times in each plot. The total number of seedlings in the experiment was 72 (2 sites*3 plots*2 factors*2 levels*3 replicates).

The sewage sludge was anaerobically digested and hygienized by drying and composting shortly two turns within two months. The concentrations of nitrogen, phosphorus and potassium were 3%, 5% and 0.07% of dm (dry mass) while soluble nitrogen and phosphorus concentrations were 0.3% and <0.1%, respectively. The organic matter content of CSS was 43%, and the moisture content was ca. 50%. Heavy metal concentrations were As 18 mg/kg, Cd 0.56 mg/kg, Hg, 0.6 mg/kg, Pb 18 mg/kg, Ni 45 mg/kg, Cr 58 mg/kg, Cu 290 mg/kg and Zn 650 mg/kg, which are below the acceptable limits for fertilizer contaminants in Finland. The elemental concentrations were analysed with ICP-OES at the laboratory of Suomen Ympäristöpalvelu Oy.

2.2. Sampling

Two similar and complete samplings were carried out in September 2009 and 2010. A soil core for enchytraeid and another for microarthropod extraction (57 mm diam, 4–5 cm deep), and one 35 mm diam. soil core for nematode extraction and physico-chemical determinations were taken with a steel corer from each experimental unit. Compost was removed from a small area before taking the soil sample. The soil cores were put individually in plastic bags and transported in cooled polystyrene boxes to the laboratory where the fresh mass (fm) of each soil core was weighed. The samples were stored below +5 °C until extractions.

Spruce needles were sampled in November 2010. Because of high needle mortality, no needle samples were taken in Ähtäri. The needle samples were oven dried, pulverized in a mortar and analyzed with a CNS analyzer at the laboratory of Suomen Ympäristöpalvelu Oy.

Sub-samples of the soil cores were weighed, oven dried ($105 \,^{\circ}$ C) for 24 h, cooled in a desiccator, and weighed again. Soil organic matter content (1 – ash mass/dry mass) was measured on a loss-on-ignition basis (4 h at 550 $^{\circ}$ C) and the moisture percentage on an ash-free basis as 100*(fresh mass – dry mass)/(om content*dry mass).

2.3. Soil animals

Enchytraeids were extracted from about 20 g (fm) for 4 h with heating and nematodes from about 3 g fm for 24 h without heating using the Baermann wet funnel technique (O'Connor, 1957). Microarthropods were extracted for ca. one week from the soil cores with a computer-controlled high-gradient extractor (dry funnels).

Collembolans and mites were enumerated on preserved samples. Living enchytraeids, nematodes and tardigrades were counted after the extraction and animal abundances were calculated per organic matter. Because the size of individual enchytraeids varies greatly, we estimated the biomass of the enchytraeid populations as described in Nieminen et al. (2012). Briefly, we estimated the biomass of the enchytraeid population as the product of the mean individual dry mass and the abundance. The mean individual dry mass was calculated using measured lengths and existing length–mass regressions (Makulec, 1983) and moisture contents (Lundkvist, 1982).

A subset of nematodes in each sample was classified as bacterial feeders, predators or others (fungal feeders, herbivores and omnivores) using Yeates et al. (1993) and the proportions of these groups were expressed as % of total nematode abundance.

2.4. Chemical analyses

Water-extractable NH₄–N and NO₃–N (full sampling in 2010 only) were extracted by soaking 5 g (fm) soil overnight in 50 ml of distilled deionized water. The extracts were filtered through 1.6 μ m glass microfiber filters (Whatman No 1822055) and 0.45 μ m membrane filters (Supor 450 Membrane 61854, Pall Corporation 600 South Wagner Road, Ann Arbor, Michigan), stored at –20 °C until analysis, and analyzed for pH, conductivity, NH₄–N and NO₃–N (autumn 2009 and 2010) according to standards SFS-EN ISO 11732:2005 and SFS-EN ISO 13395:1996. Chemical analyses were carried out at the laboratories of the Institute for Environmental Research, University of Jyväskylä (NO₃–N), PP Laboratoriopalvelut (NH₄–N, pH, conductivity), and Suomen Ympäristöpalvelu Oy (elemental analyses).

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