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# Effects of transient anaerobic conditions in the presence of acetylene on subsequent aerobic respiration and $N_2O$ emission by soil aggregates

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**Abstract**

Our objective was to assess the effect of anaerobic conditioning in the presence of acetylene on subsequent aerobic respiration and $N_2O$ emission at the scale of soil aggregates. Nitrous oxide production was measured in intact soil aggregates Δ (compacted aggregates without visible porosity) and Γ (aggregates with visible porosity) incubated under oxic conditions, with or without anaerobic conditioning for 6 d. $N_2O$ emissions were much higher in aggregates that had been submitted to anaerobic conditioning than in aggregates that did not experience this conditioning, although very little $NO_3^-$ remained in soil after the anaerobic period. $^{15}N$ isotope tracing technique was used to check whether $N_2O$ came from nitrification or denitrification. The results showed that denitrification was the major process responsible for $N_2O$ emissions. The aerobic $CO_2$ production rate was also measured in intact soil aggregates. It was greater in aggregates submitted to anaerobic conditioning than in those that were not, suggesting that the anaerobic conditioning lead to an accumulation of small compounds including fatty acids that are readily available for microbial decomposition in aerobic conditions. This process increases the aerobic $CO_2$ production and favours the $N_2O$ emissions through denitrification.


*Keywords:* Denitrification; Anaerobiosis; Nitrous oxide; Aerobic respiration; Soil aggregates

## 1. Introduction

Nitrous oxide ($N_2O$) is a trace gas involved in atmospheric pollution; it contributes to the greenhouse effect (Smith, 1990; IPCC, 1996), and affects the chemistry of $O_3$ in the upper troposphere and lower stratosphere (Graedel and Crutzen, 1992). $N_2O$ is mainly produced in soils during biological denitrification and nitrification (Tortoso and Hutchinson, 1990; Groffman, 1991; Conrad, 1996).

Various models, more or less complex, have been proposed to estimate $N_2O$ emissions through nitrification and denitrification. Most of them account for the variations with time in environmental variables such as soil water, $NO_3^-$ content and temperature. Simplified models (Parton et al., 1988; Hénault and Germon, 1995; Parton et al., 1996) do not account for the microbial dynamics, while more complex ones (Grant, 1995) explicitly consider these dynamics. However, the latter models do not consider variations in potential microbial activities, particularly the variations in respiration, $N_2O$ production and $N_2O$ reduction activities, whereas they are likely to vary with time in arable soils.

Potential denitrification has been shown to be correlated with soluble organic matter and easily mineralisable C (Burford and Bremner, 1975). Anaerobic conditions may lead to the accumulation of small organic compounds, including acetate (Tsusuki and Ponnamperuma, 1987; Dassonville et al., 2004) that may be consumed later in aerobic conditions and decrease temporarily the pH of the soil solution (Dassonville et al., 2004). Such changes in

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easily mineralisable C compounds and pH might affect potential respiration and denitrification activities.

Our aim was to assess the consequence of a prolonged anaerobic period (6 d) on subsequent aerobic respiration and net $N_2O$ emission through denitrification. Experiments were performed at the soil aggregate level. The micro-scale approach to study denitrification was motivated by the fact that, in many cases, the conditions experienced by soil organisms at the microscale are not reflected by measurements on bulk soil samples (Parry et al., 2000). For example, $O_2$ concentrations may decrease from values nearly equal to the atmospheric concentration to zero within a few millimetres in soil aggregates (Greenwood, 1961; Greenwood and Berry, 1962; Sextone et al., 1985; Sierra et al., 1995).

## 2. Materials and methods

### 2.1. Soil aggregate sampling and conservation

Experiments were performed on an Orthic Luvisol (FAO classification) sampled at Mons-en-Chaussée in Northern France (49°80' N, 3°60' E). The soil was cropped with maize in 2000. The properties of the soil were as follows: clay, 194 g $kg^{-1}$; silt, 706 g $kg^{-1}$; sand, 68 g $kg^{-1}$; total $CaCO_3$, 32 g $kg^{-1}$; pH (water), 8.2; organic C, 8.52 g $kg^{-1}$; total N, 1.00 g $kg^{-1}$. At sampling time, the soil contained 4.7 mg $NO_3^-$-N $kg^{-1}$. Large aggregates were sampled in the ploughed layer (10–30 cm depth) on 12 September 2000. Two sets of aggregates were distinguished according to Richard et al. (1999): aggregates Δ, with a massive structure and no visible porosity resulting from compaction due to traffic (Fig. 1a), and aggregates Γ, with a fragmentary structure and visible porosity (Fig. 1b). The larger aggregates were gently broken down immediately after sampling and then calibrated: we kept aggregates between 25 and 30 mm diameter. In order to reduce microbial activity during storage, the aggregates were air-dried over 3 d to obtain a residual moisture close to 0.11 g $g^{-1}$ soil, and then stored at 2 °C until the beginning of the experiments, i.e. until November 2000, January 2001 and May 2001, for experiments 2, 3 and 1, respectively. Because of water evaporation during storage, the soil moisture at the beginning of experiments was 0.11, 0.07 and 0.06 g $g^{-1}$, for experiments 2, 3 and 1, respectively.

### 2.2. Batch incubations and measurements

Three experiments were carried out. Experiment 1 was performed to check the effect of anaerobic conditioning on the subsequent aerobic respiration, by measuring $CO_2$ production. Aggregates were rewetted with water; half of them were then submitted to 6.6 d of anaerobic conditioning. Experiment 2 was performed to check the effect of anaerobic conditioning on the subsequent $N_2O$ emissions.

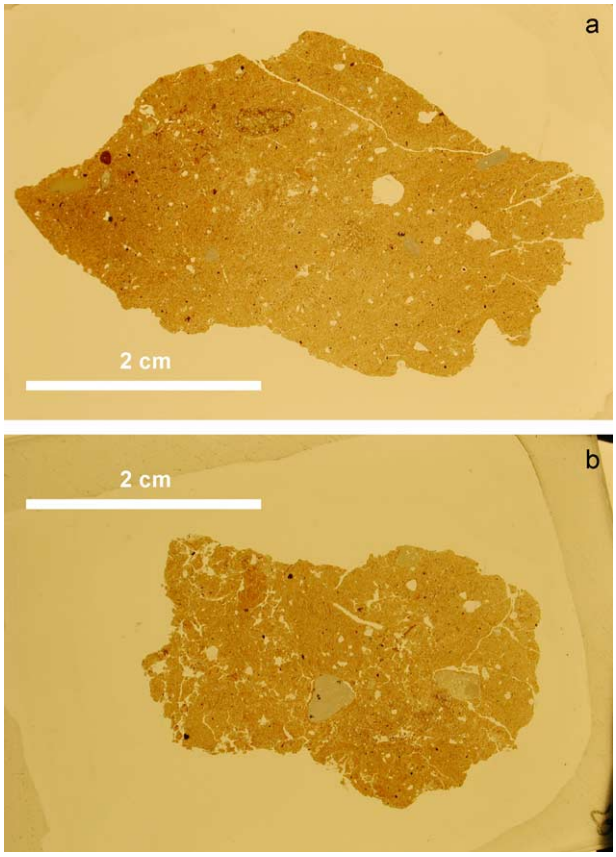


Fig. 1. Photographs of thin sections of Δ (a) and Γ (b) aggregates, The thin sections were obtained after the inclusion of dry clods in resin. Care was then taken to center the section on the center of the clods.

Net $N_2O$ emissions were measured in air for all aggregates, and $N_2O$ gross emissions for aggregates that did not experience an anaerobic conditioning. The objective of Experiment 3 was to verify that most $N_2O$ emissions came from denitrification, by using a $^{15}N$ isotope tracing technique.

In all experiments, Δ and Γ aggregates were first rewetted with either water or $KNO_3$ solution (4 g $L^{-1}$) at 20 °C on tension tables successively at −10 kPa suction for 1 d, −5 kPa for 1 d, −1 kPa for 1 d, and −0.5 kPa for 4 d. This procedure ensured a slow rewetting process, which prevented crack formation. The soil moistures obtained after 7 d was $0.21 \pm 0.01$ and $0.24 \pm 0.01$ g $g^{-1}$ for Δ and Γ aggregates, respectively. Half of the aggregates rewetted with water then experienced a 6.6 d period of anaerobic conditioning: they were incubated in anaerobic conditions in flasks that received 3 successive cycles of 3 min vacuum and 3 min of pure $N_2$ gas addition. Approximately 5% of $N_2$ (2% in experiment 1) was removed and replaced by the same volume of $C_2H_2$ in order to record $NO_3^-$ consumption through denitrification in experiment 2 and create the same conditions in experiments 1 and 3. In experiment 2, approximately 1% additional $N_2$ was removed and replaced by the same volume of Kr, in order to check for gas leakage. During anaerobic conditioning in experiment 2, gas samples