Effects of 3,4-dimethylphyrazole phosphate-added nitrogen fertilizers on crop growth and N_2O emissions in Southern Italy

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ABSTRACT

The effect of the nitrification inhibitor 3,4-dimethylphyrazole phosphate (DMPP) on N-fertilized crop growth and soil N_2O emissions were studied at two experimental sites in Southern Italy, characterised by a Mediterranean climate and different soil texture. The experiments were a randomized block design of two treatments: crop fertilized with NH_4NO_3 (considered the control treatment) or amended with DMPP plus NH_4NO_3 (considered the DMPP treatment). ANOVA was performed to assess differences between treatments and fertilization periods whereas simple and multiple linear regressions were performed in order to assess the effect of the soil-related independent variables on soil gases emissions. Growth of potato plants fertilized with DMPP-added nitrogen was enhanced compared to control plants, whereas no benefit on maize plants grown during summer was observed. N_2O emissions measured from soil to potato after the first fertilization with DMPP-added nitrogen was reduced during winter, but was higher than control after the second fertilizer application in spring, leading to comparable N_2O emission factors (EF1) between treatments. In maize N_2O emissions and EF1 were lower for DMPP compared to control treatment. The effectiveness of reduction in soil N_2O emission was influenced by soil temperature and water-filled pore space (WFPS) in both experimental sites. However, the overall effect of WFPS was contrasting as N_2O emissions were decreased in potato and enhanced in maize.

Keywords: greenhouse gases; plant growth; nitrous oxide; DMPP; Mediterranean climate

Nitrous oxide (N_2O) is a powerful greenhouse gas with a global warming potential 298 times higher than CO_2. Arable soils are the major source of N_2O and it was shown that agricultural N_2O emissions increased nearly 20% from 1990 to 2005 (IPPC 2007). Soil N_2O production is related to biological processes such as nitrification and denitrification; both processes are affected by soil temperature, nitrogen availability, and moisture. Agricultural management operations such as fertilization, irrigation, tillage, etc. can affect N_2O fluxes (Drury et al. 2006, Rochette et al. 2008). The employment of nitrogenous fertilizers increases N_2O emissions from arable soils, especially when fertilization rates exceed the amount required to optimize crop growth (McSwiney and Robertson 2005, Zebarth et al. 2008) and the soil conditions are favourable to its production.

Among strategies proposed to diminish N_2O emissions from arable soil, the utilization of N-fertilizers added with nitrification inhibitor is proposed as a feasible measure to reduce N_2O losses. DMPP (3,4-dimethylphirazole phosphate) is an inhibitor of nitrification with some advantageous properties, namely high efficiency and low risk for translocation, compared to other nitrification inhibitors (Zerulla et al. 2001). It is known that the beneficial effect of DMPP on plant production is more pronounced in light textured soils (Linzmeier et al. 1999, Pasda et al. 2001) whereas little information is available about the relation of soil texture to DMPP effect on N_2O production. Most published studies focused on the reductive effect of DMPP on soil N_2O emissions in different cropping systems (Weiske et al. 2001, Belastegui Macadam et al. 2003, Menéndez et al. 2006, Pfab et al. 2012). Only few studies were carried out under Mediterranean climate conditions (Ranucci et al. 2011). The Mediterranean climate is characterized by high rainfall during winter that may increase soil moisture up to an ideal condition for denitrification. On the other hand, hot summers may reduce the benefit of DMPP on soil N_2O production because at higher temperature DMPP is degraded faster so its inhibitory capacity is reduced faster than at lower temperatures. Laboratory experiments report that DMPP is less effective in reducing soil N_2O emissions at elevated temperatures (Suter et al. 2010a,b).

The aim of this study was to evaluate the benefits of DMPP on plant growth and soil N_2O emission from maize and potato grown in two locations in Southern Italy characterized by different soil texture and to assess the dependence of emissions from biophysical variables.

MATERIAL AND METHODS

Experiments were carried out at two sites in Southern Italy, Acerra (40°57'N, 14°25'E) and Ponticelli (40°86'N, 14°33'E) (Naples), characterized by Mediterranean climate and different soil textures (Table 1). Potato (*Solanum tuberosum* L.) was grown in Acerra, from March to June 2011, and maize (*Zea mays* L.) in Ponticelli, from May to August 2011. In both sites the experiments were set up according to a randomized block design. In Acerra two treatments were applied with three replicates (3 × 4 m plots): NH_4NO_3, SO_3, P_2O_5 (C, control plots) and NH_4NO_3 added with nitrification inhibitor (DMPP, Entec®, K + S nitrogen, DMPP plots) plus SO_3 and P_2O_5. The same treatments, replicated four times with 6 × 4 m plots, were imposed in Ponticelli. Total N supplied to potato and maize crops was 180 kg/ha N and 250 kg/ha N, respectively. The fertilizer was provided in two applications, at sown and during the vegetative growth stage (30 and 66 days after sowing, respectively for maize and potato), respectively the 40% and 60% of total N supplied. The potato crop was irrigated by furrow while maize crop by sprinklers to provide optimal water regime for crop growth.

Biometrical determination. Plant growth was followed during the whole cropping cycle and the biometrical determinations performed during both the vegetative stage and at reproductive stage (harvest). Two plants per plot were collected from each treatment and transferred into laboratory for biomass determination. The latter was determined by oven drying plants at 60°C up to constant weight. The total biomass and its partitioning in root, leaf, stem, and tubers for potato and leaf, stem, and ears for maize was determined.

Soil gas flux measurement and soil sampling. Soil N_2O emission was measured by static chamber technique. Air samples, collected by means of a Polypropylene syringe, stored in vials and analysed by means of gas chromatography (SRI 8610C, gas chromatograph, Torrance, USA), were collected before closing the lid of the chamber and after

Table 1. Soil properties at 0–0.20 m depth at the two experimental sites

	Acerra	Ponticelli
Soil type	Sandy-loam	Sandy
Texture (%):		
Clay	15.5	8
Silt	26	12
Sand	58.5	80
Bulk density (g/cm^3)	1.0	1.37
pH_{H_20}	7.37	7.90
EC (µS/cm)	289	409
$CaCO_3$ (%)	2.0	–
Organic carbon (%)	2.54	1.47
$N-NO_3^-$ (mg/kg)	10.3	15.2
$N-NH_4^+$ (mg/kg)	17.3	34.6
Total N (g/kg)	1.82	1.81

closing chamber at three subsequent times. The N_2O flux was estimated as:

$$F = \Delta C/\Delta t \times V/A$$

Where: V and A – volume and surface area of the chamber; ΔC – difference in N_2O concentration between the start and the end of gas sampling, and Δt – time between consecutive air samples.

N_2O emission factor (EF1) was calculated as:

$$EF1 = fc/N$$

Where: fc (calculated by a linear interpolation) – N_2O cumulative flux, and N – total nitrogen input.

Soil CO_2 fluxes were measured by means of a soil chamber (Li-6400-09) connected to Li-6400 Portable Photosynthesis System (LiCor Inc., Lincoln, USA). Both N_2O and CO_2 fluxes were measured in the morning at weekly intervals.

Soil NH_4 and NO_3 content were determined on samples collected near the chambers at a depth of 0–0.20 m. Soil was air-dried and sieved up to a particle size of 2 mm. NH_4^+-N and NO_3^--N content was determined in a 2 mol/L KCl soil extract and measured colorimetrically (HACH DR/2000 spectrophotometer, Loveland, USA).

Soil temperature and volumetric soil water content (VSWC) were determined at a depth of 0–0.20 m by means of a thermocouple and a TDR (Tektronix 1502B Metallic Cable Tester, Refurbished, Melrose, Scottish), respectively. Water-filled pore space (WFPS) was calculated by VSWC and bulk density taking into account of an average density of the solid matrix of 2.65 g/cm^3.

Statistical analysis. Differences in plant characteristics and in soil N_2O emissions between treatments within each sampling date and influence of sampling periods on N_2O emissions were tested by one-way ANOVA using SigmaPlot 12.0 (Systat Software Inc. Releases, San Joses, USA). Simple linear regressions and multiple linear regressions (best subset method) were performed to assess the effect of the soil-related independent variables (WFPS, NO_3^--N, NH_4^+-N, and T_{soil}) on soil gases emissions for the periods in correspondence of the fertilization events.

RESULTS AND DISCUSSION

Plant growth. The height of potato plants was higher ($P < 0.05$) in plots where the fertilizer was added with the nitrification inhibitor (DMPP) compared to control plots (Figure 1a), while no difference

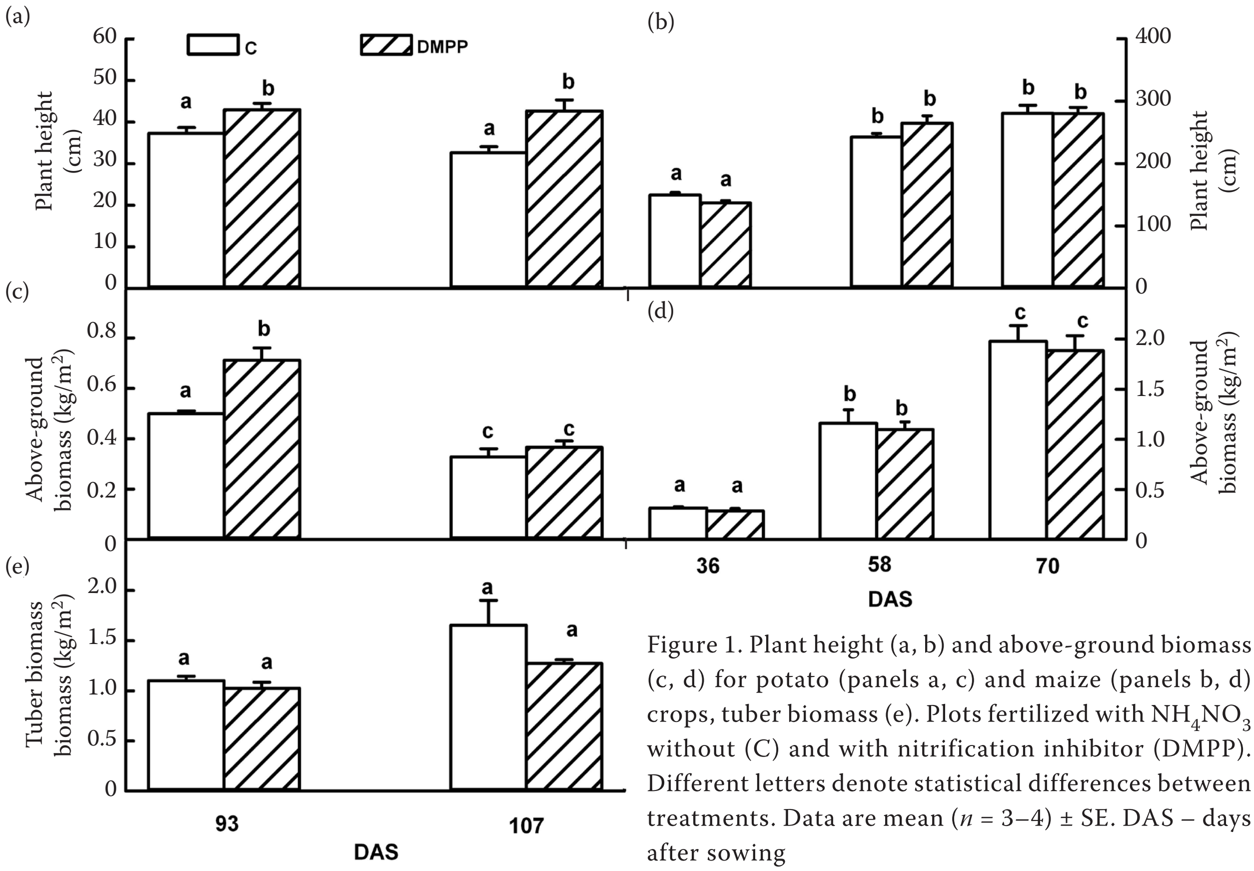

Figure 1. Plant height (a, b) and above-ground biomass (c, d) for potato (panels a, c) and maize (panels b, d) crops, tuber biomass (e). Plots fertilized with NH_4NO_3 without (C) and with nitrification inhibitor (DMPP). Different letters denote statistical differences between treatments. Data are mean (n = 3–4) ± SE. DAS – days after sowing

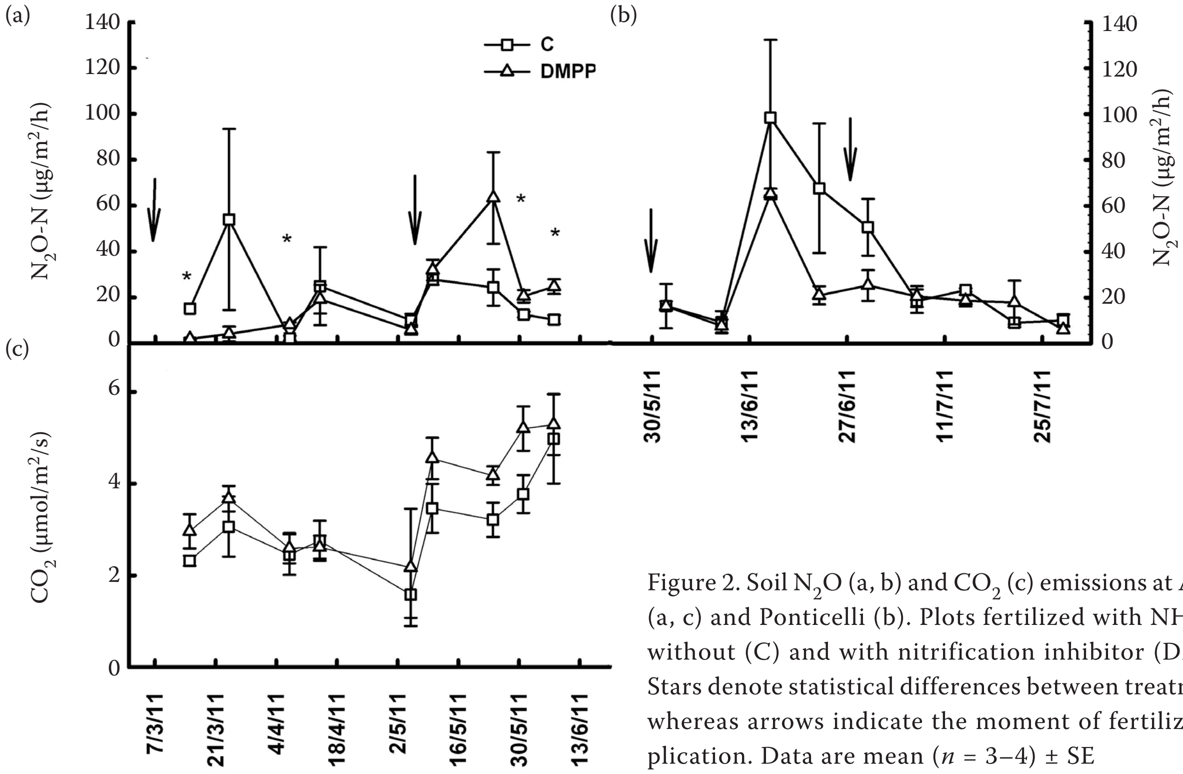

Figure 2. Soil N_2O (a, b) and CO_2 (c) emissions at Acerra (a, c) and Ponticelli (b). Plots fertilized with NH_4NO_3 without (C) and with nitrification inhibitor (DMPP). Stars denote statistical differences between treatments, whereas arrows indicate the moment of fertilizer application. Data are mean (n = 3–4) ± SE

in the height of maize plants was found (Figure 1b). The above-ground biomass of potato plants resulted higher ($P < 0.005$) in DMPP plots compared to control plots only 93 days after sowing (DAS) (Figure 1c), whereas no difference in above-ground biomass for maize plants was found (Figure 1d). No significant difference in tuber biomass between treatments and sampling dates could be detected (Figure 1e). The lack of enhancement in potato yield and maize growth and yield in DMPP plots could be related to the soil conditions that might have reduced DMPP inhibitory effect. It is known that high temperature accelerates DMPP degradation affecting the effectiveness of DMPP on soil N loss (Zerulla et al. 2001). At the end of potato cropping cycle (May–June), during fast tuber swelling stage and during the whole maize growing season (June–July), WFPS was low (Figures 3c,d), contributing to maintain the soil temperature higher than 20°C (Figure 3a,b). As a consequence, the DMPP action on NH_4^+ oxidation could have been depressed. This hypothesis seems to be consistent with the absence of significant differences in soil NH_4^+ concentration between treatments in both cropping systems (Figure 3g,h). An enhanced NH_4^+ content was found to be advantageous for maize crops grown on coarse-texture soils with slightly alkaline pH (Teiker and Hobbs 1992).

Soil greenhouse gases emissions. In the days following the winter-time fertilizer application in Acerra, N_2O emissions from DMPP potato planted plots were lower ($P < 0.01$) than those from control potato planted plots (Figure 2a). Following the second fertilizer application (during spring), N_2O fluxes measured in DMPP plots remained higher ($P < 0.05$) than those measured in control plots until the end of crop cycle (Figure 2a), leading to comparable N_2O emission factors (EF1) between treatments (0.29% and 0.26% for control and DMPP, respectively). A sharp peak in N_2O emission from control plots about 30 days after the first fertilizer application was observed; this peak appeared notably delayed in DMPP plots, a likely consequence of a prolonged persistence of DMPP in the soil at moderately low temperature (Figure 3a). As reported by Zerulla et al. (2001) DMPP degradation is temperature-dependent, resulting faster as temperature increases. After the second application, a similar peak in N_2O emission from DMPP plots was detected about 18 days from fertilization. It is supposed that in late spring, a faster DMPP degradation due to soil temperatures higher than 20°C (Figure 3a) could have occurred, enhancing NH_4^+ oxidation and in turn N_2O production by nitrifiers. This is consistent with the positive relationship found between soil

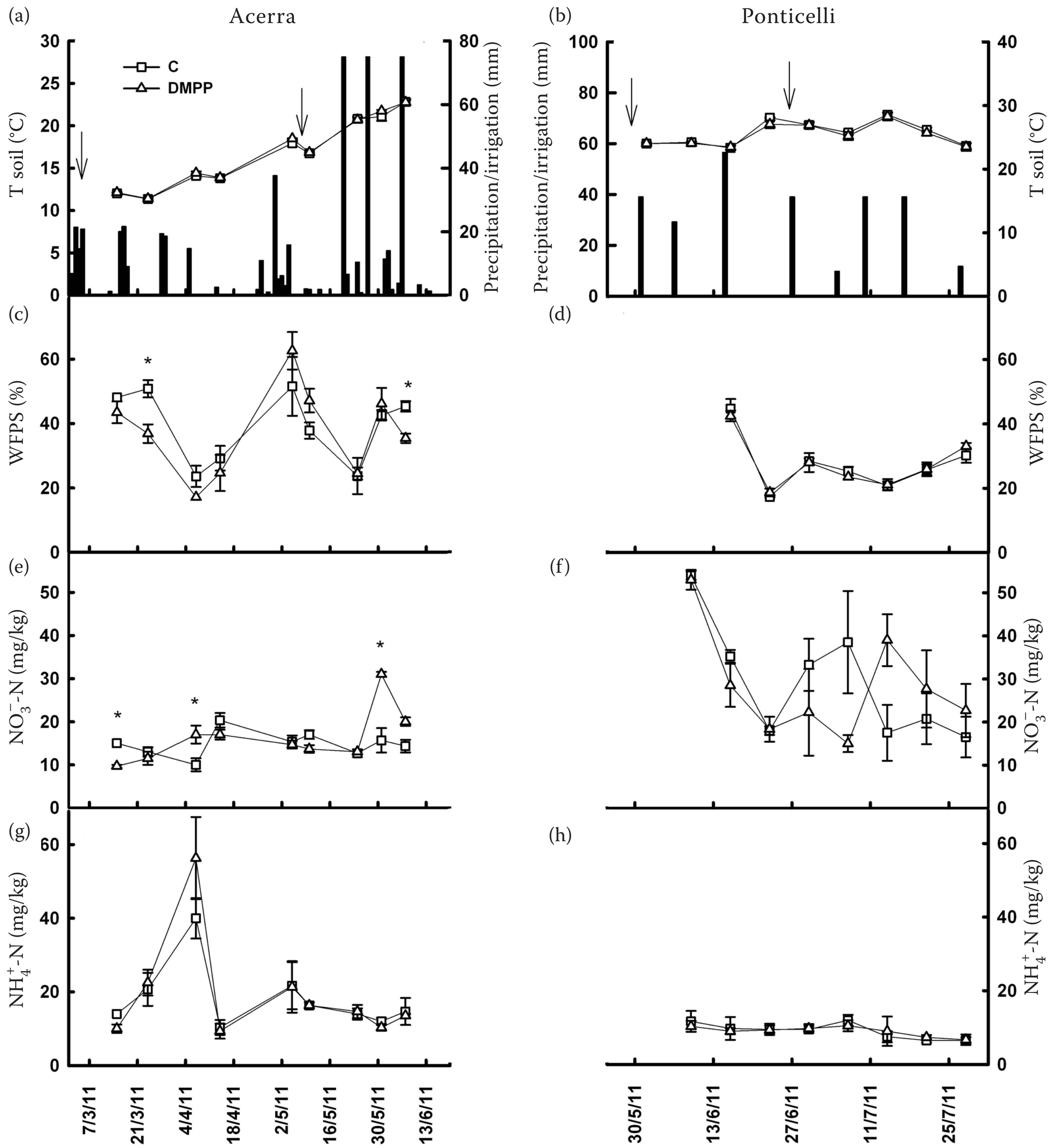

Figure 3. Soil temperature (open symbols) and water supply to crop (bars) by precipitation or irrigation (a, b); water-filled pore space (WFPS; c, d); nitrate (e, f) and ammonium (g, h) concentration. Plots fertilized with NH_4NO_3 without (C) and with nitrification inhibitor (DMPP). Stars denote statistical differences between treatments, whereas arrows indicate the moment of fertilizer application. Data are mean (*n* = 3–4) ± SE

NO_3^- concentration and soil temperature (NO_3^--N = 0.86 + 0.93T_{soil}, r^2 = 0.350, P < 0.005) in DMPP plots, which might be related to an increase in NH_4^+ oxidation attributable to a progressive loss of the inhibitory effect of DMPP due to temperature. This conclusion is in agreement with data obtained in Ponticelli, where no difference in N_2O emissions from DMPP and control maize planted plots during summer was found (Figure 2b), likely due to higher soil temperatures than 20°C (Figure 3b) that negatively affected the DMPP effectiveness. However, a clear trend of higher emissions from control plots could be detected (Figure 2b), that led to a higher EF1 in control plots (0.21%) compared to DMPP plots (0.14%). Differently from Acerra, in Ponticelli a single peak in N_2O emissions was detected from both plots 15 days following the first fertilization, likely in response to an increase of soil moisture (WFPS higher than 50%) (Figure 3d) due to a precipitation event. Overall, our data confirm the influence of temperature on the efficiency of DMPP into contrasting NH_4^+ oxidation and the effectiveness of DMPP in soils with coarse texture.

In Acerra, DMPP application did not reduce soil CO_2 emissions (Figure 2c), differently from findings of Weiske et al. (2001) and Pfab et al. (2012). For the potato crop, as NO_3^- concentration in DMPP plots paralleled N_2O emission trend (Figures 2a, 3e), it is hypothesized that, diversely from Kleineidam et al. (2011), the lower N_2O fluxes in DMPP plots during winter could be ascribed to the NH_4^+ oxidation inhibition by DMPP rather than to a negative effect of DMPP on nitrifying bacteria.

ANOVA evidenced a significant effect ($P < 0.005$) of sampling periods on soil N_2O emissions at both experimental sites. These periods were identified as the days following the first and the second fertilizer application. Therefore, we separately analysed the influence of soil-climatic conditions on N_2O emissions by means of single or multiple linear regressions during these two periods. In both sites, no relationship between N_2O fluxes and soil-climatic conditions for the first period were found; conversely WFPS and soil temperature affected N_2O emissions in the days following the second fertilizer application. In Ponticelli WFPS positively influenced soil N_2O emissions in DMPP plots (N_2O-N = −11.4 + 1.79 WFPS, $r^2 = 0.920$, $P < 0.001$). In control plots a synergic effect of WFPS and T_{soil} on N_2O emission (N_2O-N = −223.1 + 6.8 WFPS + 2.59 T_{soil}, $r^2 = 0.316$, $P < 0.05$) was found. This positive influence of soil moisture on N_2O emissions is likely to be ascribed to the nitrification process that represents the main source of N_2O in soils at 35–60% WFPS (Bateman and Baggs 2005, Kiese et al. 2008). Surprisingly, a significant negative relationship between N_2O fluxes and WFPS for control (N_2O-N = 41.1 – 0.6 WFPS, $r^2 = 0.345$, $P < 0.05$) and DMPP (N_2O-N = 86.7 – 01.31 WFPS, $r^2 = 0.373$, $P < 0.05$) plots as well as between N_2O fluxes and T_{soil} for control plots (N_2O-N = 74.5 – 2.7 T_{soil}, $r^2 = 0.412$, $P < 0.05$) was found for data collected in Acerra. The different response of N_2O fluxes observed at the two experimental sites could be ascribed to differences in soil properties, or to a direct effect of soil temperature on soil moisture which, drying the soil, negatively affects nitrifiers activity. Regarding this aspect we observed a reduction of N_2O emission with increase in soil temperature in control plots. However, no relationship between soil temperature and WFPS was found for both nitrogen treatments (data not shown), thus we attribute the reduction of N_2O emissions to soil properties such as pore size distribution and bulk density that, influencing water holding capacity, affect the optimal % WFPS for N_2O production. Mathieu et al. (2006) hypothesized that nitrification belonged to a water-limited phase (i.e., nitrification increases with increasing soil water content) under unsaturated conditions and to an aeration-limited phase (i.e., nitrification decreases with increasing soil water content) under saturated conditions. In Acerra the soil is sandy-loam, characterized by higher water holding capacity compared to the sandy soil of Ponticelli. Therefore, it is supposed that a WFPS higher than 40% could become limiting for nitrification in Acerra, but this was not the case in Ponticelli where a coarse texture allows a better soil aeration.

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