## Is co-application of municipal sewage sludge and inorganic fertilizer on soils a double-edged sword? Trade-offs among nitrogen kinetics, carbon sequestration and greenhouse gas emissions.

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Under the framework of Circular Economy, EU Green Deal and UN Sustainable Development Goals the use of organic amendments is highly promoted as a cost-efficient solution to improve soil quality and agrosystem sustainability. Municipal sewage sludge is a by-product of wastewater treatment plants and it is projected to exceed 7 million ton in the Eurozone (Eurostat, 2023). Only a small part of it, less than 30% is re-utilized for agriculture, while the remaining majority is disposed in landfills. This in an unsustainable practice. Nonetheless, their agronomic use comes with an uncertainty of their potential to release ample plant-available nitrogen (N), and to emit soil greenhouse gasses (GHG). All policies consider the crucial role of soil and soil microbes as nutrient and GHG regulators. For example, when organic amendments are co-applied with inorganic N, soil microbes through their biochemical functioning may release nutrients for plant growth and GHG that are intermediates and products of their assimilatory, dissimilatory and respiratory processes. Therefore, any changes in soil organic carbon (C) may feed-back into the N Cycle, enhancing soil nitrous oxide (N<sub>2</sub>O) emissions or offsetting any soil C sequestration action. Which leads us to the question in our study: Is combined application of organic and inorganic N a double-edged sword?

We investigated the short-term (90 d) soil N dynamics of sandy soil mesocosms receiving municipal sewage sludge (MSS) amendments (50 t/ha), without and with N fertilization (200 kg/ha; urea-N). An unamended soil mesocosm was included as control. Municipal sewage sludge was thermally dried after dewatering to reduce moisture and microbial load, and was collected from Thessaloniki Wastewater Treatment Plant, Thessaloniki, Greece. Urea-N was commercial grade with 46% N, was supplied by Corteva Agriscience Hellas SA, Thessaloniki, Greece. The agricultural soil was collected from an agricultural field (upland cotton; Gossypium hirsutum) at Aiginion, Greece. Soil mesocosms consisted of 2 kg (dw) soil, 15% soil moisture (w/w), and the respective amendment as indicated in table 1. During the incubation soil ammonium ( $NH_4^+$ ), nitrate ( $NO_3^-$ ), nitrite ( $NO_2^-$ ), N<sub>2</sub>O and carbon dioxide ( $CO_2$ ) were regularly monitored. Soil  $NH_4^+$ , NO<sub>3</sub>-, NO<sub>2</sub>-, were extracted with 30 ml 2 M potassium chloride (KCl) and their concentration was determined following standard assays (SSSA, 1994; CSSS, 2007). CO<sub>2</sub> and N<sub>2</sub>O emissions were determined using an infrared gas analyser (LI-800; LI-COR, USA) and a gas chromatographer (Agilent, USA), as previously described (Giannopoulos et al., 2011; Thers et al., 2019; Giannopoulos et al., 2020). At the end of the incubation integrated and cumulative values were calculated to better compare the time series in our experiment. Two sustainability indexes were calculated, to better understand the C and N footprint of our mesocosms. The first index was the emission factor, which is the proportion of N<sub>2</sub>O emissions per N applied, and the second index was the CO<sub>2</sub> equivalent per organic C stored, which is the proportion of total greenhouse gas emissions equivalent (cumulative CO<sub>2</sub> and N<sub>2</sub>O soil emissions) per soil organic C gained (corrected by soil organic C content of control treatment).

Table 1. Experimental design.

Index	Treatment	Fertilizer (Urea- N; kg/ha)	Municipal Sewage Sludge (t/ha)
C	Control	0	0
C+U	Control+Urea-N	200	0
MSS	Municipal Sewage Sludge	0	50
MSS+U	Municipal Sewage Sludge+Urea-N	200	50

Urea hydrolysed quickly and increasing soil  $NH_4^+$  concentrations were observed in all treatments just after 5 d. Peak concentrations of  $NH_4^+$  were observed in all urea-N treatments (C+U, 88 mg kg<sup>-1</sup> and MSS+U, 371 mg kg<sup>-1</sup>) and MSS treatment (127 mg kg<sup>-1</sup>) at 14d. (Fig. 2). Soil  $NH_4^+$  reached background concentrations at 35 d for C+U, contrary soil  $NH_4^+$  was detectable until 42 d in the MSS and MSS+U treatment. It is evident that the addition of urea-N (C+U), municipal sewage sludge (MSS) and their combined application (MSS+U) increased the availability of soil  $NH_4^+$  by 3x,

5x and 12x times relative to unamended soil, respectively. Interestingly, we observed a tremendous release of soil NO<sub>2</sub>only in the urea treatment (C+U; 128 mg kg<sup>-1</sup>), and not in the other remaining treatments (C, MSS and MSS+U). In terms of soil NO<sub>3</sub>, which is a product of nitrification and a substrate for denitrification, we observed a similar pattern of NO<sub>3</sub>accumulation in all treatments (C+U, 399; MSS, 563 and MSS+U, 771 mg kg<sup>-1</sup>), except the unamended soil treatment (C) where soil NO<sub>3</sub><sup>-</sup> remained below 5 mg kg<sup>-1</sup>. Though-out the incubation approx. 12.7x, 13.4x and 19.7x more soil NO<sub>3</sub><sup>-</sup> was observed for the C+U, MSS, and MSS+U treatment, relative to the control treatment (C), respectively. If soil NO<sub>3</sub>is not assimilated by crops, there is a high risk of NO3<sup>-</sup> leaching in aquatic bodies. Considering the gaseous emissions of CO<sub>2</sub> and N<sub>2</sub>O, that are generally products of soil respiration, nitrification and denitrification, the addition of municipal sewage sludge (MSS) and co-application municipal sewage sludge with urea-N (MSS+U), enhanced soil CO<sub>2</sub> by 2.4x and 2.4x, and by 13.6x and 16.9x for soil N<sub>2</sub>O emissions, respectively. Fertilization by urea-N alone (C+U) did not affect much soil CO<sub>2</sub> (526 mg CO<sub>2</sub>-C kg<sup>-1</sup>) and N<sub>2</sub>O (1258 µg N<sub>2</sub>O-N kg<sup>-1</sup>) emissions when compared to the unamended soil treatment (C). The MSS+U reduced the emission factor, which is the percentage of N<sub>2</sub>O per applied N, by 5x when compared to MSS treatment, however it was well above the IPPC emission factor of 1%. Municipal sewage sludge is a source of C, though ee observed that municipal sewage sludge (MSS, 74%) and the combined treatment (MSS+U, 96%) enhanced the CO<sub>2</sub>-equivenat per soil C gain index, indicating a complete loss of the added organic C though greenhouse gas emissions.

In this set of soil, we observed beneficial aspects of municipal sewage sludge co-application with chemical fertilization (urea-N) in soil fertility (soil  $NH_4^+$  and  $NO_3^-$ ). Due to the evident accumulation of soil  $NO_3^-$ , we argue that mineralization and nitrification are driving  $N_2O$  emissions. The addition of easily degradable C through MSS mineralization, enhanced gaseous losses of the added C and N. Considering our key question, whether co-application of inorganic fertilizer and municipal sewage sludge is a Double-edged sword, we conclude that municipal sewage sludge and fertilizer co-application should be carefully evaluated case per case, as it affects several key soil parameters differently, and therefore we should seek new ways to minimize gaseous losses thus to improve sustainability in agrosystems.

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