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RESEARCH ARTICLE

Microbial biomass activity of a sodic Lixisol reclaimed with gypsum and clean water irrigation in urban vegetable systems of Burkina Faso

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Abstract

Background: Little is known about the effects of gypsum application to remediate saline–sodic soils in the tropics and the role of microbial indicators in soil reclamation.

Aims: Our study aimed at (1) remediating a highly weathered, irrigated sodic Lixisol under prolonged urban crop production by clean water and gypsum application and (2) to determine the remediation effects on soil microbial indices.

Methods: A three-factorial on-farm experiment with maize (*Zea mays* L.) was used to study effects on soil microbial biomass of (1) soil degradation at two levels of salinity, (2) irrigation with clean water and wastewater, and (3) the impact of added gypsum during a typical growing season.

Results: At the high-degradation site, the 0.5 M K_2SO_4 extractable carbon (C) content was 40% higher than at the low-degradation site. In addition, microbial biomass carbon (MBC) and microbial biomass nitrogen (MBN) were 20% lower than at the low-degradation site, while fungal ergosterol was even 40% lower, leading to a 33% lower ergosterol/MBC ratio. Wastewater irrigation increased MBN but decreased ergosterol content at the low-degradation site while having no effect at the high-degradation site. Gypsum amendment led to higher MBN at the low-degradation site but to lower MBN at the high-degradation site. Gypsum amendment always increased the ergosterol content whereby this increase was stronger at the low-degradation site, especially in combination with wastewater irrigation.

Conclusions: From a microbial perspective, high soil degradation levels should be avoided by treatment of a saline–sodic wastewater prior to its use for irrigation rather than relying on future remediation strategies of affected field sites.

KEYWORDS
ergosterol, salt leaching, soil remediation, urban agriculture, West Africa

1 | INTRODUCTION

Urban and peri-urban agriculture (UPA) in Sub-Saharan Africa is under increasing pressure by rapid urbanization, which forces it to frequently relocate production sites. Across the region, UPA is challenged by water scarcity and therefore untreated wastewater is often used to irrigate crops (Levy et al., 2014). Irrigation with wastewater improves crop yields as a result of the waters' high nutrient concentration (Fritz et al., 2022). However, untreated, and partially treated wastewaters may contain unwanted heavy metals or other ions, organic compounds, and pathogens, putting animal and human health at risk, as well as jeopardizing the quality of groundwater resources and soil fertility. Accumulation of heavy metals such as Pb, Co, and Cr and salinization-alkalinization as a consequence of the deposition of exchangeable Na deposits followed by damage of soil structure in urban vegetable gardens were reported at the vegetable production site of Kossodo in Ouagadougou (Kiba et al., 2012).

It is well known that the use of wastewater and the accumulation of Na^+ ions may trigger displacement of Ca^{2+} ions from soil aggregates leading to dispersion of clays and organic matter (Mavi, Marschner, et al., 2012). This results in poor soil physical properties, losses of soil permeability and stability, low soil water content, and hyper-osmosis (Mavi, Marschner, et al., 2012; Qadir & Schubert, 2002). Consequently, plant growth is compromised, and soil microbiological activity is reduced. Given the importance of UPA, there is therefore an urgent need to find effective solutions for soil remediation at saline–sodic vegetable production sites such as Kossodo.

Techniques proposed for the remediation of saline–sodic soils include the use of plants for phytoremediation, chemical and organic amendments, and leaching or drainage processes with clean water alone or mixed with saline water. Leaching or drainage of access salts and gypsum application ($CaSO_4 \times 2\ H_2O$) is often used for the recovery of saline–sodic soils (Stenchly et al., 2017). Numerous studies focused on gypsum application to such soils and reported an improvement in their physico-chemical and biological properties (Ahmad et al., 2016). Gypsum application may also lead to changes in soil microbial properties (Carter, 1986), governing the plant availability of nutrients such as phosphorus (P), sulfur (S), nitrogen (N), and micronutrients. For example, the application of gypsum increases the N use efficiency of organic fertilizers through the reduction of denitrifying bacteria (Batra & Manna, 1997). In this context, virtually nothing is known about the effects of gypsum on total soil microbial and fungal biomass, the key drivers of decomposition processes in soils (Joergensen & Wichern, 2018), during the reclamation stage in urban and peri-urban gardens.

This study therefore used saline–sodic fields of Kossodo as a case study for reclamation by gypsum amendment and clean-water irrigation (Dao et al., 2018, 2019; Stenchly et al., 2017). The specific objective of our study was to determine the effects of clean-water irrigation and gypsum amendment on soil microbial biomass carbon (MBC), nitrogen (MBN), fungal ergosterol (Joergensen & Wichern, 2018), as well as on basal respiration (Anderson & Domsch, 1989). The following hypotheses were tested: (1) both reclamation approaches improve the microbial and especially the fungal habitat properties and (2) the positive gypsum amendment and clean-water irrigation effects are stronger at the high-degradation site than with low soil degradation.

TABLE 1 Characteristics of clean water and wastewater used for irrigation during the wet season (October 2015 to February 2016) at the Kossodo site of urban agriculture in Ouagadougou, Burkina Faso. For mineral elements, data show total concentrations in μg mL^{-1}

Variables (unit)	Clean water (mean ± SE)	Wastewater (mean ± SE)
pH	8.7 ± 0.3	9.0 ± 0.1
EC (dS m^{-1})	0.29 ± 0.07	2.28 ± 0.03
N	1.8 ± 0.7	6.5 ± 1.4
P	0.4 ± 0.2	18.1 ± 0.7
K	10.4 ± 3.1	51.9 ± 1.2
Mg	4.5 ± 1.3	4.3 ± 2.0
Ca	27 ± 9	267 ± 5
Na	16 ± 4	514 ± 19
Cl	15 ± 5	13 ± 7
COD (μg mL^{-1})	22 ± 9	868 ± 16

Abbreviations: COD, chemcial oxygen demand; EC, electrical conductivity; SE, standard error; SAR, sodium absorption ratio.
Source: Dao et al. (2019), modified.

2 | MATERIALS AND METHODS

2.1 | Study area

The study was carried out on farmers' fields at the Kossodo garden site located in the North-Eastern district of Ouagadougou, Burkina Faso (N 12° 25′ 44.277″ W 1° 28′ 22.394″; 300 m asl, World Geodetic System 1984 - WGS 84). The site covers 35 ha of the downstream area of an urban wastewater treatment plant. It consists of a conventional system to clean effluents of municipal, industrial, agricultural, and runoff flows typical for many urban centers (Dao et al., 2018, 2019; Stenchly et al., 2017). Three anaerobic basins are connected in two sedimentation basins followed by three maturation basins that are working in parallel. It was set up to collect and treat 5400 m^3 day^{-1} of domestic and industrial wastewater from a brewery, slaughterhouse, a cement factory, and steel sheet factories. Within a few years of vegetable cultivation, the irrigated Lixisol (IUSS Working Group WRB, 2015) with the partially treated wastewater from the purification plant was rapidly alkalinized given high concentrations of Na in the wastewater (Table 1) and absence of any reclamation or mitigation efforts. As a result, soil $pH_{water1:2.5}$ currently exceeds 8.5 (Table 2).

TABLE 2 Soil chemical characteristics at the Kossodo site of urban agriculture in Ouagadougou, Burkina Faso (0–20 cm depth)

	Low-degradation site		High-degradation site	
Variables (unit)	Clean water	Wastewater	Clean water	Wastewater
Sand (%)[a]	70.5 ± 0.5	70.0 ± 0.1	65.6 ± 0.2	66.9 ± 1.5
Silt (%)[a]	26.0 ± 0.4	26.3 ± 0.1	30.5 ± 0.3	29.2 ± 1.4
Clay (%)[a]	3.3 ± 0.2	3.7 ± 0.4	3.8 ± 0.1	3.6 ± 0.2
Soil pH (H_2O 1:2.5)	7.2 ± 0.8	7.3 ± 0.1	8.5 ± 0.3	8.5 ± 0.1
$EC_{1:2}$ (dS m^{-1})	0.26 ± 0.15	0.21 ± 0.004	0.64 ± 0.11	0.60 ± 0.10
ESP (%)	12.3 ± 5.0	10.9 ± 2.9	28.6 ± 3.2	22.4 ± 3.6
SAR	4.9 ± 2.5	4.0 ± 0.8	14.5 ± 1.3	12.2 ± 2.9
CEC ($mmol_C$ g^{-1} soil)	40.4 ± 6.0	40.4 ± 1.6	48.5 ± 4.2	55.0 ± 3.6
Ca_{EX} ($mmol_C$ g^{-1} soil)	16.8 ± 0.9	17.9 ± 1.2	16.3 ± 4.9	18.7 ± 5.6
Mg_{EX} ($mmol_C$ g^{-1} soil)	5.0 ± 2.5	5.9 ± 0.5	2.4 ± 0.5	2.9 ± 0.8
K_{EX} ($mmol_C$ g^{-1} soil)	3.5 ± 1.6	3.0 ± 0.2	7.3 ± 1.1	8.8 ± 1.7
Na_{EX} ($mmol_C$ g^{-1} soil)	5.2 ± 2.8	4.4 ± 1.0	14.0 ± 2.7	12.2 ± 1.8
SOC (mg g^{-1} soil)	8.3 ± 0.2	7.3 ± 0.5	6.0 ± 0.5	7.2 ± 1.1
Total N (mg g^{-1} soil)	0.63 ± 0.05	0.56 ± 0.05	0.55 ± 0.09	0.66 ± 0.10

[a]Our data and data from Dao et al. (2019).
Abbreviations: COD, chemcial oxygen demand; EC, electrical conductivity; ESP, exchangable sodium percentage; SE, standard error; SAR, sodium absorption ratio; SOC, soil organic carbon.

2.2 | Experimental design

In 2015, prior to the onset of the experiment, fields of the site were surveyed and grouped into three levels of soil degradation according to soil pH (Dao et al., 2019). For each level of soil degradation, four fields were chosen as replicates and each of them were split into three sub-plots. Two of the three sub-plots were considered in this study. The wastewater-treated plots and the clean water-treated plots were divided into two sub-plots with and without gypsum treatment. There were three factors used for this study. The first factor was the soil degradation level as defined by pH at low severity (pH < 7.5) versus highly degraded plots (pH > 8.5), and the second experimental factor was gypsum application at 0 t ha^{-1} (control) and at the calculated gypsum requirement (4 t ha^{-1} for low-degraded plots and 12 t ha^{-1} for highly degraded plots) based on Equation (1) proposed by Ayers and Westcot (1985) and Rasouli et al. (2013):

$$\text{Gypsum amendment}\left(\text{tha}^{-1}\right) = (\text{ESP}_{\text{initial}} - \text{ESP}_{\text{final}}) \times \text{CEC} \times \text{BD} \times D \times F \times 0.00086, \qquad (1)$$

where *ESP* is the exchangeable sodium percentage (%), *CEC* is the cation exchange capacity ($mmol_C$ kg^{-1}), *BD* is the soil bulk density (g cm^{-3}), *D* is the soil depth (cm), and *F* is Ca and Na exchange efficiency, which was considered equal to 1.

The third experimental factor was irrigation water quality comprising wastewater and clean water. While the wastewater was taken from the effluents of the Kossodo treatment plant (Table 3), the clean water came from the tap.

The experiment was conducted during the rainy season of 2016 (lasting from June to October), using a local maize (*Zea mays* L.) variety as a test crop. All plots were hoed at about 15 cm depth followed by application of cattle manure (C:N ratio = 8.97) at 10 t ha^{-1}. Subsequently, NPK-SB (14–23–14–6–1) was broadcast at a rate of 150 kg ha^{-1} and urea (46% N) at 100 kg ha^{-1}. The NPK-SB fertilizer was applied once at day 16 after sowing, and urea was split into two equal rates applied at 23 and 40 days after sowing. Soil samples were collected at 10 cm depth after harvest in October 2016 for assessing soil microbiological indices. To this end, five soil samples per plot were taken and pooled. Subsequently, samples were sieved at 2 mm and stored in paper bags.

2.3 | Soil microbial properties

Basal respiration and soil microbial biomass were determined after moistening and adjusting soil samples to 50% water holding capacity followed by pre-incubation at 22°C in the dark for 14 days before analysis. For assessment of basal respiration, 50 g of moist soil sample were incubated with 5 mL of 0.5 M NaOH for 14 days at 22°C in the dark. CO_2 was measured by back-titration of the unconsumed NaOH with 0.5 M HCl after adding 5 mL of saturated $BaCl_2$ to precipitate carbonate as insoluble $BaCO_3$.

MBC and MBN were measured by chloroform fumigation extraction using 0.5 M K_2SO_4 as extractant (Brookes et al., 1985; Vance et al., 1987). Organic C and total N were measured in the extracts using a Multi N/C 2100S analyzer (Analytik Jena GmbH, Jena, Germany). MBC was E_C/k_{EC}, where E_C = (organic C extracted from fumigated soils) –

TABLE 3 Main effects of degradation level, water quality, and gypsum amendment on 0.5 M K_2SO_4 extractable carbon C and soil biological characteristics (0–10 cm depth) at the Kossodo site of urban agriculture in Ouagadougou, Burkina Faso, in October 2016

	Extractable C (μg g^{-1} soil)	MBC	MBN	MB-CN	Basal respiration (μg CO_2C g^{-1} soil day^{-1})	qCO_2 (mg CO_2C g^{-1} MBC day^{-1})	Ergosterol (μg g^{-1} soil)	Ergosterol/ MBC (%)
Low degradation	92	227	40	6.1	6.6	30	0.76	0.35
High degradation	130	180	32	5.7	6.1	34	0.41	0.23
Clean water	124	214	33	6.6	6.6	32	0.61	0.29
Wastewater	103	187	38	5.1	6.1	33	0.51	0.27
Without gypsum	115	201	35	6.0	6.8	34	0.45	0.22
With gypsum	112	199	36	5.7	5.9	30	0.67	0.34
Probability levels								
Degradation	<0.01	0.02	0.02	NS	NS	NS	<0.01	0.01
Water quality	0.04	NS	0.05	0.01	NS	NS	0.04	NS
Gypsum	NS	NS	NS	NS	NS	NS	<0.01	<0.01
D × W	0.03	NS	0.04	0.01	NS	NS	NS	NS
D × G	0.03	NS	0.02	0.03	NS	NS	0.01	NS
CV (± %)	17	19	21	19	21	19	24	29

Note: Probability values of a three-way ANOVA. All gypsum × water (G × W) interactions were not significant. All degradation × gypsum × water (D × G × W) interactions were not significant.

Abbreviations: CV, mean coefficient of variation (n = 3, low degradation, n = 4, high degradation); MBC, microbial biomass carbon; MBN, microbial biomass nitrogen; NS, not significant.

(organic C extracted from non-fumigated soils) and k_{EC} = 0.45 (Wu et al., 1990). MBN was E_N/k_{EN}, where E_N = (total N extracted from fumigated soils) – (total N extracted from non-fumigated soils) and k_{EN} = 0.54 (Brookes et al., 1985; Joergensen & Mueller, 1996).

Ergosterol was extracted from 2 g moist soil with 100 mL ethanol for 30 min by oscillating shaking at 250 rev min^{-1} (Djajakirana et al., 1996). Subsequently, ergosterol was determined by reversed phase HPLC (Gynkotek M 480 pump, UVD 340 S detector, and Gina 50 autosampler, Germering, Germany), using 100% methanol as the mobile phase and detected at a wavelength of 282 nm.

2.4 | Statistics

Data are presented as arithmetic means on an oven-dry weight basis (24 h at 105°C). Statistical analyses were carried out using SigmaPlot 13.0 (Systat, San José, CA, USA). Data were tested for normality of residuals using the Shapiro–Wilk test. The significance of treatment effects was analyzed by a three-way analysis of variance. Interrelationships between the soil biological properties were tested by linear regression analysis.

3 | RESULTS

At the high-degradation site, the 0.5 M K_2SO_4 extractable C content was generally 40% higher than at the low-degradation site (Table 3). Wastewater irrigation had no effect at the low-degradation site but decreased the extractable C content at the high-degradation site (Figure 1), leading to a significant degradation level × water quality interaction (Table 3). Gypsum amendment led to higher extractable C contents at the low-degradation site but to lower extractable C contents at the high-degradation site (Figure 1), causing a significant degradation level × gypsum amendment interaction (Table 3).

At the high-degradation site, MBC and MBN were 20% lower than at the low-degradation site (Table 3), whereas fungal ergosterol was even 40% lower, leading to a 33% lower ergosterol/MBC ratio. The MB-C/N ratio, basal respiration, and the metabolic quotient qCO_2 did not significantly differ between degradation levels.

Wastewater irrigation increased MBN at the low-degradation site but had no effect at the high-degradation site (Figure 2a), leading to a significant degradation level × water quality interaction (Table 3). The MB-C/N ratio showed the reverse response to wastewater irrigation than MBN as MBC was not affected by this treatment. Wastewater irrigation decreased ergosterol at the low-degradation site but had no effect at the high-degradation site (Figure 2b), leading to a significant degradation level × water quality interaction (Table 3). In contrast, wastewater irrigation did not significantly affect the ergosterol/MBC ratio. The same was true for the basal respiration and the metabolic quotient qCO_2.

Gypsum amendment led to higher MBN at the low-degradation site but to lower MBN at the high-degradation site (Figure 2a), causing a significant degradation level × gypsum amendment interaction (Table 3). The MB-C/N ratio showed the reverse response to gypsum amendment than MBN. Gypsum amendment always increased the ergosterol content, but this increase was stronger at the low-degradation site, especially in combination with wastewater irrigation

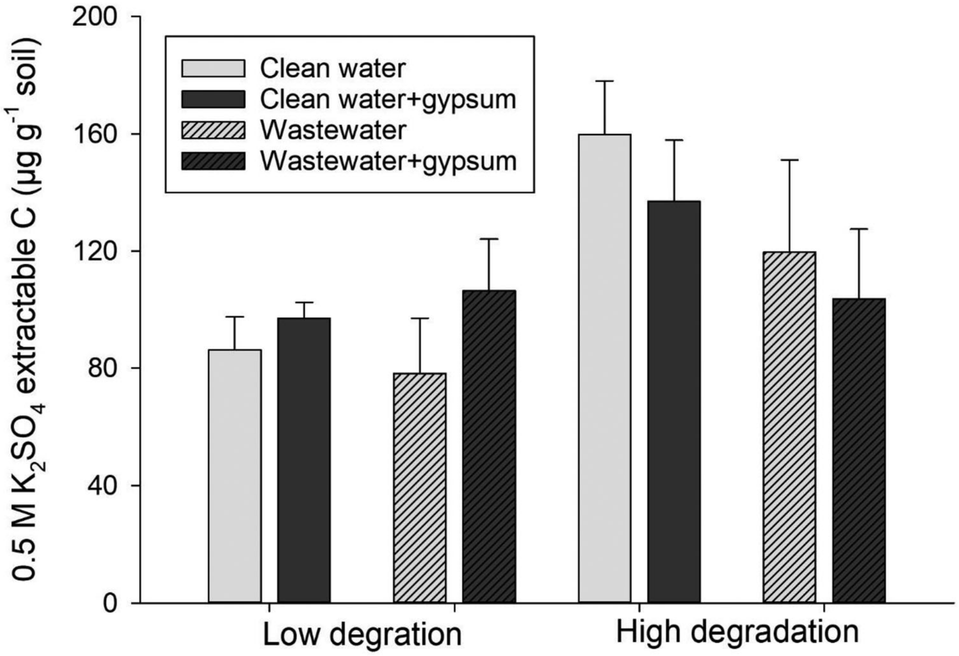

FIGURE 1 Effects of degradation level, water quality, and gypsum amendment on 0.5 M K_2SO_4 extractable carbon (C) (0–10 cm depth) at the Kossodo site of urban agriculture in Ouagadougou, Burkina Faso, in October 2016. Bars on top of the columns show one standard deviation.

(Figure 2b). The same was true for the ergosterol/MBC ratio (Table 3). Gypsum amendment did not significantly affect MBC, basal respiration, nor the metabolic quotient qCO_2.

4 | DISCUSSIONS

4.1 | Treatment effects on extractable organic C

The high Na load of the current wastewater increased the sodium absorption ratio values over several years, weakening the Ca bridges between clay and soil organic matter (SOM) (Fei et al., 2017; Igwe, 2005; Mavi, Sandermann, et al., 2012). This process transferred an increasing percentage of SOM into an extractable form (Curtin et al., 2021; Setia et al., 2013), especially at the sodic high-degradation site (Table 3; Figure 1). Consequently, elution may additionally contribute to the decline in SOM in the long-term.

In the current study, wastewater irrigation had no additional increasing effect on soil Na content at the low-degradation site and even a decreasing effect at the high-degradation site (Table 2), which is difficult to explain. One reason might be the high Ca concentration of the wastewater (Table 1). This argument is supported by the results of a study by Setia et al. (2013) where gypsum amendment led to a reduction in the content of 0.5 M K_2SO_4 extractable C in a highly sodic soil from Australia. In contrast, the increased concentration of sulfate in the soil solution at the low-degradation site is most likely the reason for the increased content of 0.5 M K_2SO_4-extractable C at the neutral low-degradation site (Figure 1). Sulfate solutions have been shown to increase the contents of extractable SOM from non-fumigated soil in comparison with water as extractant (Haney et al., 1999, 2001). However, this increase was small in comparison to the overall effects of high-degradation on SOM loss and microbial biomass reduction at the high-degradation site.

4.2 | Treatment effects on microbial biomass and bacterial activity

At the high-degradation site, MBC and MBN were markedly reduced by salinity and sodicity, without any effect on the MB-C/N ratio (Table 3). Clean-water irrigation had no positive effect on MBC and MBN, whereas gypsum amendment had a small but significant negative effect on MBN. In addition, Carter (1986) observed a decrease in MBN with gypsum amendment compared to no gypsum use in highly Solonetzic soils from Canada. This underlines that soil degradation as a consequence of irrigation with saline water should be avoided as the gypsum-based remediation of such sites is despite quick leaching of excess sulfur—difficult and slow if not impossible. In contrast to many previous studies (Muhammad et al., 2008; Sardinha et al., 2003; Wichern et al., 2006), our results show that basal respiration and qCO_2 values were not affected by any treatment. This may, however, be partly due to the high variation among the replicates and the low level of bacterial activity. However, the significant linear regression ($r = 0.55$, $p < 0.01$) between MBC and basal respiration suggests that both microbial indices were in principle similarly affected by the degradation level, wastewater irrigation, and gypsum addition.

In contrast, wastewater irrigation and gypsum amendment both had a strong increasing effect on MBN, but not MBC, significantly reducing the MB-C/N ratio (Table 3). A low MB-C/N ratio in soil usually indicates C limitation in combination with sufficient N availability to

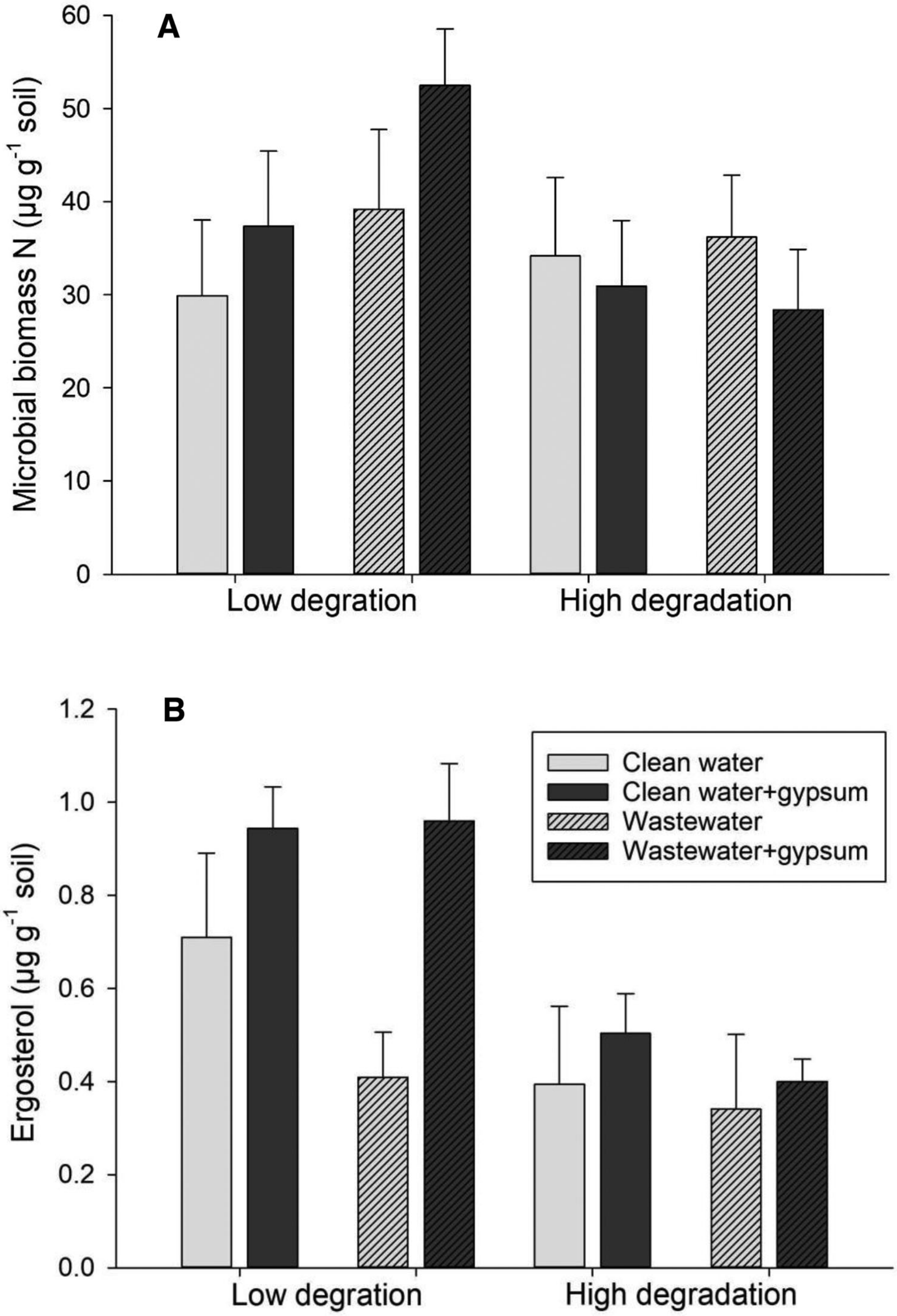

FIGURE 2 Effects of degradation level, water quality, and gypsum amendment on (a) microbial biomass N (MBN) and (b) ergosterol (0–10 cm depth) at the Kossodo site of urban agriculture in Ouagadougou, Burkina Faso, in October 2016. Bars on top of the columns show one standard deviation.

soil microorganisms (Khan et al., 2016; Khan & Joergensen, 2019). However, this seems to be different in the current study. Wastewater typically contains high concentrations of total N and organic C but reportedly only N can be taken up by soil microorganisms in considerable amounts, especially by a transfer into fungi and their vacuoles (Khan et al., 2016; Klionsky et al., 1990). This suggests that the organic matter in the wastewater applied to a soil is hardly available to soil microorganisms (Fritz et al., 2022). Reasons for this might be the high salinity of most wastewaters (Musazura et al., 2019), but heavy metal concentrations (Diogo et al., 2010) may also contribute to this phenomenon. Wastewater may also contain high concentrations of recalcitrant purine- and pyrimidine-derived biogenic heterocyclic organic N compounds depending on the feedstock for the gut microbiome (Bosshard et al., 2011; Leinweber et al., 2013).

4.3 | Treatment effects on soil fungi

At the high-degradation site, ergosterol and the ergosterol/MBC ratio were markedly reduced by both salinity and sodicity. Clean water irrigation had no positive effect on ergosterol, whereas gypsum amendment had a small but significant positive effect under these severe environmental conditions (Figure 2). The current study confirms the negative effects of wastewater irrigation on soil fungi as previously reported under acidic (Sardinha et al., 2003; Wichern et al., 2006) and alkaline conditions (Muhammad et al., 2008; Pankhurst et al., 2001). It should be noted that salinity has not only negative effects on fungal biomass, but also on fungal necromass (Khan et al., 2016; Wichern et al., 2006), reducing the possibility to increase soil organic carbon stocks as a result of C application in the wastewater.

The claim of specifically higher sensitivity of fungi against salinity has been challenged by Rath et al. (2016), who observed a higher sensitivity of bacterial growth than fungal growth against salinity. However, they used the H_3-leucine in protein (Bååth, 1994) and ^{14}C-acetate in ergosterol incorporation (Bååth, 2001) methods, which presumably measure only the active fraction of bacteria and fungi, respectively (Joergensen & Wichern, 2018). However, it might be possible that highly adapted soil fungi exhibit a higher activity in the absence of their fungal competitors. This points to the advantage in combining the biomarker approach with the H_3-leucine in protein and acetate in ^{14}C-acetate in ergosterol methods in one study, to elucidate the relationship of active and dormant fungi in their response to salinity and sodicity.

At the low-degradation site, wastewater irrigation led to a strong decline in ergosterol, which was fully compensated by gypsum application under clean water irrigation (Figure 2). Gypsum application is known to have a positive effect on soil fungi. One reason could be the high sulfur demand of many soil fungi (Heinze et al., 2010, 2021). The much stronger response of ergosterol at the low-degradation site in comparison to the high-degradation site suggests again that soil degradation should be avoided rather than its consequences be remediated.

5 | CONCLUSIONS

Salinity and sodicity had severe negative effects on microbial biomass, especially on soil fungi. Short-term clean-water irrigation had only minor remediating effects on soil microorganisms at both sites. In contrast, gypsum addition had generally strong positive effects on soil fungi only at the low-degradation site. From a microbial perspective, high soil degradation levels as a result of irrigation with saline–sodic wastewater should be avoided given the difficulties in remediation.

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DATA AVAILABILITY STATEMENT

The data that support the findings of this study are available from the corresponding author upon reasonable request.

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