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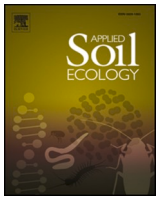

Organic and conventional farming systems shape soil bacterial community composition in tropical arable farming

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ABSTRACT

Soils present a limited resource for agricultural production and bear a vast diversity of organisms crucial for crop health and the provision of ecosystem services. There is growing evidence that agricultural practices affect soil microbial community structure and function but currently, there is a knowledge gap when it comes to tropical arable farming systems. In this study, we investigated the long-term impact of organic and conventional production systems on bacterial communities in two field trial located on a rhodic and humic nitisol in the Central Highlands of Kenya. The field sites operate on a full factorial design, testing farming systems (organic vs conventional) and input levels (high vs low). Including four field replication we assessed soil bacterial community structure via amplicon sequencing of the 16S rRNA gene and soils capacity for nitrification and nitrous oxide reduction via qPCR of functional genes (bacterial and archaeal *amoA*, *nosZ*) after 12 years of distinct management and before the start of the 5th three-year crop rotation period in 2019. The abundances of *amoA* bearing nitrifiers and *nosZ* bearing nitrous oxide reducers were enhanced in the high input organic production system on humic but not in rhodic nitisols. For both soil types, high input organic production system resulted in distinct bacterial community structure with enhanced bacterial richness compared to conventional and low input production systems. In rhodic and humic nitisols 160 and 84 OTUs were found to be indicative for organic production system at high input levels organic. Taxa associated with this system were identified as potential primary decomposers or symbionts related to plant nitrogen fixation, suggesting organic fertilization strategies such as manure composting as major driver for changes in soil bacterial community structure. This study reveals that organic production systems at high input levels on tropical nitisols translates to distinct soil bacterial communities with increased capacity for soil processes that are crucial for crop nutrient supply.

1. Introduction

Soil is a complex structure that provides essential ecosystem services like the production of food, the regulation of nutrient and water cycles and the provision of habitat (Adhikari and Hartemink, 2016). Soil quality describes the inherent capacity of soils to function within natural or managed ecosystem boundaries to provide ecosystem services and is usually assessed by a set of physical, chemical and biological indicators (Bünemann et al., 2018). Yet, soil quality assessments in the last decades were mainly based on chemical and physical quality indicators such as soil nutrient contents and soil pH (Lehmann et al., 2020). Today, the evolving concept of soil health perceives soil as a living entity and calls for a greater inclusion and further development of biological indicators (Lehmann et al., 2020). Among the emerging biological soil quality indicators, the assessment of soil microbial community structure has made the most promising methodological advances and is increasingly used in

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environmental research(Bahram et al., 2018; Delgado-Baquerizo et al., 2018). Although it is inherently difficult to link microbe-based indicators directly with soil health or ecosystem functioning (Fierer et al., 2021), there is growing evidence that soil microbial diversity represents an important biological resource that should be considered in agricultural management decisions (Hartmann and Six, 2023; Wagg et al., 2021).

Agricultural intensification in the last century secured the burgeoning demand for food by a growing population at the expense of soil and environmental health (Ramankutty et al., 2018). Consequently, societal concerns stimulated the development of sustainable agricultural practices in the last decades (Tilman et al., 2011), and today organic farming systems have gained momentum in food production worldwide (Willer and Jan Trávníček, 2021). Since the addition of mineral fertilizers for plant nutrition and chemical plant protection strategies are prohibited, organic farming systems usually show lower yields, but offer benefits for biodiversity, nutrient cycling and environmental quality (Seufert and Ramankutty, 2017). Organic farming systems were shown to enhance key soil quality indicators such as soil organic carbon contents, microbial biomass and soil respiration (Krause et al., 2022). The addition of organic inputs in particular has been shown to drive soil microbial community structure in organic and conventional farming systems in temperate climates (Hartmann et al., 2015).

Due to the omission of mineral nitrogen fertilizers in organic farming, the provision of plant-available nitrogen to secure crop production presents a key challenge. Conventional agricultural practices mitigate this challenge by applying mineral nitrogen sources; globally, >80 million tons of nitrogen fertilizers are applied to soils annually (Lu and Tian, 2017). Negative environmental impacts induced by the loss of nitrogen from agricultural systems include the emission of greenhouse gases, such as nitrous oxide, and the eutrophication of distant ecosystems and surface waters (Fowler et al., 2013). Consequently, managing soil nitrogen cycling is crucial for the development of sustainable agricultural production. The two major microbial processes of soil nitrogen transformation are nitrification and denitrification (Canfield et al., 2010). Nitrification is carried out by ammonia-oxidizing bacteria (AOB) and archaea (AOA) through the enzyme ammonia monooxygenase (*amoA*). Ammonia supplied by mineral nitrogen fertilizers and organic inputs is a major source of energy for nitrifying prokaryotes in agricultural soils (Rasche and Cadisch, 2013). Denitrification is the stepwise reduction of nitrate under anoxic conditions to dinitrogen through the obligatory intermediates nitrite, nitric oxide and nitrous oxide. The last step of denitrification, the conversion of nitrous oxide to dinitrogen, is encoded by the functional gene *nosZ* (Philippot et al., 2007) and is of special environmental relevance due to its mitigation potential for agricultural N_2O emissions (Domeignoz-Horta et al., 2016).

A global meta-analysis revealed enhanced soil microbial abundance and activity under organic compared to conventional farming systems (Lori et al., 2017) and there is growing evidence for organic farming practices to shape soil microbial community structure especially in temperate arable systems (Hartmann et al., 2015). However, there is limited data about the impact of farming systems on soil microbial community structure and function in tropical climates (Lori et al., 2017). To address this knowledge gap, we characterized soil bacterial community structure and functional capacity for nitrification and nitrous oxide reduction in two 12-year-old long-term field trials in the Central Highlands of Kenya that compare organic and conventional production systems at high and low input levels (Adamtey et al., 2016).

We hypothesized that: i) high input systems enhance soil microbial capacity for nitrification and nitrous oxide reduction compared to low input systems due to increased nitrogen fertilizer supply, ii) organic and conventional farming systems harbour distinct soil bacterial community structure depending on input levels, and iii) organic production systems enhance bacterial richness as consequence of the organic fertilization strategy.

2. Material and methods

2.1. Field sites

The long-term experimental fields were set up in 2007 at Chuka and Thika in the sub-humid Central Highlands of Kenya. The characteristics of the fields´ soil and design of the long-term experiments is described in detail in Adamtey et al. (2016). Briefly, the field experiments are set up in a randomized complete block design with four and five replicates in Chuka and Thika, respectively. Plot sizes are 8 m × 8 m with a net sampling area of 6 m × 6 m.

The Chuka experimental site is located at the Kiereni Primary School (0° 19′ 39.49" S 37° 38′ 52.40″ E) and has a warm, humid climate. It lies at 1458 m above sea level, with a mean annual temperature of 20 °C and a mean annual rainfall of 1373 mm. The soil type is a humic nitisol with medium to high innate fertility. It is fine-textured and well-drained with a bulk density of ~1 g cm^{-3}. The soil texture is composed of 74 % clay, 17 % silt, and 9 % sand (Adamtey et al., 2016). After 9 years of distinct management organic carbon contents differed between farming systems with highest contents in Org-High (28.37 g kg^{-1}) followed by Con-High (24.17 g kg^{-1}), Con-Low (22.82 g kg^{-1}) and Org-Low (21.86 g kg^{-1}) (von Arb et al., 2020).

The Thika experimental site located at the Kenya Agricultural and Livestock Research Organization KALRO Horticultural Research Centre (1° 2′ 57.76" S 37° 5′ 34.62″ E) lies at 1500 m above sea level. It has a mean annual temperature of 20 °C, and a mean annual rainfall of 840 mm. The soil type is a rhodic Nitisol with low soil fertility and a mean bulk density of 1.1 g cm^{-3}. The soil texture constitutes 81 % clay, 14 % silt, and 5 % sand with distinct soil organic carbon contents between farming systems after three crop rotation periods (Org-High - 22.91 g kg^{-1}, Con-High - 21.11 g kg^{-1}, Con-Low - 19.02 g kg^{-1}, Org-Low - 18.85 g kg^{-1}) (von Arb et al., 2020).

2.2. Experimental setup

Arable cropping at Chuka and Thika field site follows a six-season, three-year crop rotation with maize and baby corn (*Zea mays*) in the long seasons (March to September) and vegetables, legumes, and potato in the short seasons (October to February). A detailed overview on the crop rotation is listed in Adamtey et al. (2016). At each site, organic (Org) and conventional (Con) production systems are compared at two input levels (High vs. Low) resulting in a total of four production systems: Con-High, Org-High, Con-Low and Org-Low. Systems at high input levels receive supplemental water through drip irrigation during drought periods, while the low input farming systems are strictly rainfed (Table 1). The application rates of fertilizer and management practices for High and Low input levels were based on the recommendations and practices for commercial and subsistence production systems, respectively (Adamtey et al., 2016). In the organic systems, nutrients are applied in form of composted farmyard manure, *Tithonia* tea and mulch, and rock phosphate while in the conventional systems fertilization with farmyard manure is further amended by synthetic fertilizers such as diammonium phosphate, calcium ammonium nitrate, and triple-superphosphate (Table 1). Organic carbon and total nitrogen inputs from organic fertilizers are determined annually via wet digestion (dichromate) and Kjehldahl digestion, respectively. As fertilization strategy is adjusted to fertilizer nitrogen inputs, farming systems differ in total carbon inputs (Table 1). In the conventional systems crop residues are exported, while in the organic systems crop residues remained on the field. Conventional pest control employs synthetic pesticides while microbial and plant based biopesticides are used in the organic systems (Table 1).

2.3. Soil sampling procedure and DNA extraction

Soil sampling was carried out in the dry season in March 2019,

Table 1
Farming system specific management practices in the long-term systems comparisons trials at Chuka and Thika in the Central Highlands of Kenya. Rates of nutrients in the high input and low input farming systems were as recommended by research institutions and as practiced by small scale farmers. Details on mean annual nutrient inputs, field production systems and plant protection strategies can be reviewed in Adamtey et al., 2016, Adamtey et al., in preparation.

	Soil input type	Irrigation	Selected plant-protective substances	Mean annual inputs (kg ha^{-1})		
				Nitrogen Thika / Chuka	Carbon Thika / Chuka	Phosphorus Thika / Chuka
Con-High	Farmyard manure, calcium ammonium nitrate, di-ammonium phosphate and triple super phosphate	Supplementary irrigation	Bulldock™ (Beta-cyfluthrin), Dragnet™ (Permethrin), Confidor™ (Limidacloprid)	222 / 222	1273 / 1528	127 / 127
Con-Low	Farmyard manure, calcium ammonium nitrate, di-ammonium phosphate and triple super phosphate	Rain-fed	Wood ash, Dragnet™ (Permethrin), Confidor™ (Limidacloprid)	47 / 47	524 / 520	31 / 31
Org-High	Composted farmyard manure, rock phosphate, Tithonia mulch and Tithonia tea	Supplementary irrigation	Neem oil extract (*Azadirachta indica*), Delfin (*Bacillus thuringiensis*)	228 /228	5397 / 5920	127 / 127
Org-Low	Composted farmyard manure, rock phosphate, Tithonia mulch and Tithonia tea	Rain-fed	Wood ash	47 / 47	672 / 560	31 / 31

before the vegetation period of maize growth. 16 soil cores to a soil depth of 20 cm from each experimental plot were retrieved, pooled, thoroughly homogenized, and cooled for transport to the International Centre of Insect Physiology and Ecology (ICIPE), Nairobi, Kenya. At Thika four out of five blocks were sampled to ensure equal sample numbers at both field sites. Soil samples were stored at −20 °C and a subsample of approximately 50 g were freeze-dried at ICIPE and sent to the Research Institute of Organic Agriculture FiBL, Switzerland. DNA was extracted from a subsample of 450 mg per plot using the FastDNA SPIN Kit and FastPrep Homogenizer (MP Biomedicals, USA) according to the manufacturer's instruction. The DNA was eluted in 100 μL of Tris-EDTA buffer. DNA quantity was assessed using Qubit double-stranded DNA high sensitivity assay kit (ThermoFisher Scientific, USA), and relative fluorescent units were measured with the CFX96 Optical Real-Time PCR Detection System (Bio-Rad Laboratories, Switzerland).

2.4. Quantification of functional genes

Extracted DNA was transported on dry ice to the Institute of Applied Microbiology at Justus Liebig University in Giessen and stored at −20 °C upon arrival. Quantitative polymerase chain reaction (qPCR) was used to quantify gene copies of the bacterial 16S rRNA gene as well as functional genes indicative for nitrification (bacterial and archaeal *amoA*) and nitrous oxide reduction (*nosZ*). Dilutions of DNA extracts (diluted in PCR-grade water) were tested for presence of inhibitors using the 16S rRNA gene primer system and 1:30 diluted samples were used for all qPCR measurements. qPCR standards were obtained by reamplified DNA fragments from plasmids containing the gene fragment of interest using the M13 primer system (Table S3). The reamplified PCR products were purified (QIAquick PCR Purification Kit, QIAGEN) and the DNA concentration was determined fluorometrically (Quant-iT PicoGreen dsDNA Assay Kit, Invitrogen). DNA concentrations and fragment lengths were used to determine the concentration of gene copy numbers per μl as described in Kolb et al. (2003). Quantitative PCR assays were performed with 10 μl reaction volume using the SsoFast™ EvaGreen Supermix (BioRad, USA) on a BioRad CFX 96™ Real-Time System (BioRad, Germany). Per reaction 5 μl of 2 x SsoFast EvaGreen Supermix, 0.2 μl at 10 μM each of forward and reverse primer, 1 μl of 1:30 diluted DNA extracts, and 3.6 μl PCR grade water was included. For each qPCR assay, biological replicates were analyzed with three technical replicates together with a no template control and a serial dilution of the respective qPCR standard. Primer sequences, references, and cycling conditions used are listed in Tables S1 and S2. R^2s of standard curves ranged from 0.972 to 0.999 and mean efficiencies were 70.7 % (archaeal *amoA*), 76.1 % (16S rRNA gene), 80.1 % (bacterial *amoA*), and 83.9 % (*nosZ*). Amplification of *nosZ*-II was envisaged as described in Jones et al. (2013), but concentrations were mostly below detection limit. The quality and size of the generated amplicons were checked by gel electrophoresis on agarose gels (1.4 %) stained with ethidium bromide and melting curve analyses. The initial target gene concentration of biological replicates was retrieved from the BioRad CFX Manager software and target gene copies per weight of soil sample were obtained by multiplying the mean initial target gene concentration by the volume of eluted DNA from soil samples and the dilution factor of 30 for DNA samples, and then divided by the dry weight of individual soil samples used for DNA extraction.

2.5. 16S rRNA gene amplicon sequencing

For each sample, 20 μL of the original DNA extract dissolved in Tris-buffer (5 mM, pH 8.5) with a concentration ranging between 1 and 10 ng μL^{-1} was sent to LGC Genomics (Germany) for 16S rRNA gene amplicon-based high-throughput next-generation sequencing using the Illumina technology. A negative control containing distilled H_2O and positive control containing ZymoBIOMICS Microbial Community DNA Standard (ZymoResearch, Germany) were also included (Fig. S1). PCR

amplification, tagging, library preparation, and sequencing on an Illumina MiSeq sequencing platform (Illumina, USA) were performed according to the amplicon guidelines provided by Illumina. A two-step PCR approach employed fluidigm tagged 16S rRNA gene primers 341F (5'-CCTAYGGGRBGCASCAG-3') and 806R (5'-GGACTACNNGGGTATCTAAT-3') targeting the V3-V4 regions (Sundberg et al., 2013). 16S rRNA gene amplicon library was run with a paired-end 300 bp Reagent Kit and a total of 1,719,184 raw sequences were deposited on NCBI under Accession number PRJNA853512.

2.6. Bioinformatic analysis

The bioinformatics pipeline was performed at the Genetic Diversity Center at ETH Zürich, Switzerland. USEARCH v11.0.667 was used to remove phiX and low complexity sequences, trim read ends, and merge read pairs (Edgar, 2010). Next, primer sequences were removed, and paired reads were size selected, quality filtered. Operational taxonomic units (OTUs) were defined by clustering at 97 % similarity in UPARSE (Edgar, 2013). Taxonomy was assigned with SINTAX (Edgar, 2016) employing SILVA v128 (Quast et al., 2013) as a reference database.

2.7. Chemical, physical and biological soil quality indicators

For correlation analysis of microbial community structure with soil parameters, data for chemical, physical and biological soil quality indicators was retrieved from an earlier study investigating the evolution of soil quality in Chuka and Thika field sites during the first three crop rotations (von Arb et al., 2020). Briefly, chemical soil quality indicators included soil pH, measured in an aqueous suspension(1/2.5 *w/v*), soil organic carbon and total nitrogen concentrations, determined via combustion in an elemental analyser (FlashEA®1112, Thermo Fischer), as well as total and available phosphorous contents, quantified via wet digestion and quantification of resin extractable P. Physical soil quality indicators were limited to soil aggregate stability, determined by wet sieving. Soil biological indicators included soil microbial biomass carbon and nitrogen, as quantified via chloroform fumigation extraction method. Enzymatic activity of dehydrogenases, acid and alkaline phosphatases were determined colorimetrically and soil basal respiration and nitrogen mineralization capacity was determined in small incubation trial using the CO_2 capture technique and subsequent titration and extraction of mineral nitrogen, respectively (von Arb et al., 2020).

2.8. Statistical analyses

Statistical analysis of univariate data was performed in R (version 4.0.3) and R Studio (version 1.3.1093). The effect of organic and conventional systems (Con vs. Org), inputs levels (High vs. Low), and their interaction was assessed using linear mixed-effects models with the experimental block as random term using the lme function of the nlme package (Pinheiro et al., 2022). The anova_lme function retrieved the significance of tested main effects and posthoc tests (Tukey-Kramer HSD test) were conducted when significant main effects were observed at $p < 0.05$.

For analysis of bacterial alpha- and beta-diversity, the phyloseq package was used to remove non-bacterial (chloroplasts, mitochondria and archaea) sequences using the subset_taxa function (McMurdie and Holmes, 2013) (Table S4). In total we identified 42 and 58 archaeal OTUs in Chuka and Thika field sites respectively, for which we limited our study on bacterial diversity which accounted for 3500 and 3856 OTUs (Table S4). The filtered dataset contained 15,454 to 61,812 bacterial sequences per sample in Chuka and 7313 to 47,913 bacterial sequences per sample in Thika. Rarefaction curves are displayed in the Fig. S2. Shannon index (H) and the observed richness (S) were calculated using the estimate_richness function and (H/log(S)) was used to calculate community evenness as measures for alpha diversity. Before the assessment of beta diversity, OTUs with a prevalence of <10 reads and occurring in <5 % of the samples were removed using the phyloseq_filter_prevalence function (Fig. S3). The adonis2 function of the vegan package (Oksanen et al., 2012) was used to calculate the effect of production system (Org vs. Con), input levels (High vs. Low) and their interaction on bacterial community composition on the base of euclidean distances of center log transformed data (Gloor et al., 2017). Permutations were set at 9999 and restricted on the field blocks. Differences between production systems were compared by pairwise PERMANOVA with false discovery rate *p*-value adjustment. Unconstrained ordination was visualized via non-metric multidimensional scaling (NMDS) and constrained ordination via distance-based redundancy analysis (db-RDA) using metaMDs and dbrda functions of the vegan package, respectively (Oksanen et al., 2012). Db-RDA analysis constrained ordination for production system and input level since both factors significantly affected bacterial community structure (Table 6). Environmental variables including gene copy numbers of the 16S rRNA gene and functional genes *nosZ* and bacterial and archaeal *amoA* as well as soil physicochemical and biological parameters, retrieved from von Arb et al. (2020), correlating with the projections of the ordination ($R^2 > 0.7$ and $p = 0.05$) were bi-plotted using the envfit function (Table S5).

Indicative OTUs associated with one or more treatment group combinations and a minimum abundance of 300 were determined using the multipatt function of the indicspecies package with 9999 permutations and the "r.g" function (De Cáceres et al., 2012). Lists of indicator species associated with one farming system are provided in the Supplementary Information. Effect of production system and input level on the eight most abundant indicator species was assessed via linear mixed-effects model and 95 % confidence interval of estimated marginal means (Figs. 2 and 3).

3. Results

3.1. Abundance of 16sRNA gene copies and nitrifying and nitrous oxide reducing marker genes

On the humic nitisol in Chuka, soils functional capacity for nitrification, as indicated by the abundance of archaeal and bacterial *amoA* gene copy numbers, was significantly elevated in Org-High compared to all other production systems (Table 2). Abundance of archaeal and bacterial *amoA* gene copies did not differ between Con-High, Con-Low and Org-Low. Yet, total bacterial abundances as indicated by 16S rRNA gene copy numbers, remained unaffected by production systems and input levels (Table 2). Tukey post hoc test showed significantly enhanced *nosZ* gene copy numbers in the Org-High system compared to Org-Low, Con-High and Con-Low and nitrous oxide reducers were significantly enhanced in organic production systems ($p = 0.046$) (Table 2).

On the rhodic nitisol in Thika, significantly higher bacterial 16S rRNA gene numbers were observed in organic compared to conventional production systems ($p = 0.03$) (Table 3). Despite slightly elevated abundances of *nosZ* and archaeal and bacterial *amoA* gene copies in Org-High, no significant effect of production systems or input levels was observed. Notably, in the humic and the rhodic nitisol, the abundance of archaeal *amoA* exceeded bacterial *amoA* by one to two orders of magnitudes across all treatments.

3.2. Microbial alpha-diversity

Significantly elevated Shannon diversity was found on the humic nitisol at Chuka in Org-High, followed by Org-Low, Con-High, and Con-Low, with production system ($p = 0.013$) and input levels ($p = 0.021$) as discerning factors (Table 4). Significantly higher bacterial richness was found in organic compared to conventional systems ($p > 0.001$) and in high compared to low input levels ($p = 0.013$) (Table 4). Highest species richness was found in Org-High with 1796 Operational Taxonomic Units (OTUs), followed by Org-Low with 1240 OTUs, Con-High with 1084

Table 2
Impact of farming systems on bacterial 16S rRNA gene and functional gene abundance. Data shows mean and standard error ($n = 4$) of functional gene abundances (gene copies g^{-1} dry weight soil) as affected by production system (Con vs. Org) and input levels (Low vs. High) in a humic (Chuka) nitisol. F and p-value of the main effects were obtained by ANOVA based on a linear mixed effects model. Tukey post hoc test results are given if significant main effects were found with different letters indicating significant differences at $p < 0.05$.

	16S rRNA gene copies [g^{-1} DW soil]			Archaeal *amoA* gene copies [g^{-1} DW soil]			Bacterial *amoA* gene copies [g^{-1} DW soil]			*nosZ* gene copies [g^{-1} DW soil]		
Humic nitisol - Chuka	mean	SE		mean	SE		mean	SE		mean	SE	
Con-High	3.88×10^9	6.34×10^8	–	1.39×10^8	3.83×10^7	a	6.74×10^5	1.32×10^5	a	4.22×10^8	9.55×10^7	a
Con-Low	4.03×10^9	5.03×10^8	–	1.25×10^8	1.52×10^7	a	7.84×10^5	4.20×10^5	a	4.38×10^8	9.68×10^7	a
Org-High	8.72×10^9	2.40×10^9	–	2.82×10^8	1.92×10^7	b	6.92×10^6	1.78×10^6	b	1.26×10^9	3.03×10^8	b
Org-Low	4.54×10^9	4.02×10^8	–	1.49×10^8	1.29×10^7	a	2.60×10^5	9.98×10^4	a	5.00×10^8	5.42×10^7	a

ANOVA	denDF	F-value	p-value	F-value	p-value	F-value	p-value	F-value	p-value
Production system (PS)	9	3.256	0.105	9.41	0.013	7.264	0.025	5.336	0.046
Input levels (IL)	9	1.841	0.208	7.276	0.024	9.517	0.013	3.639	0.089
PS x IL	9	2.122	0.179	4.746	0.057	10.171	0.011	3.978	0.077

Table 3
Impact of farming systems on bacterial 16S rRNA gene and functional gene abundance. Data shows mean and standard error (n = 4) of functional gene abundances (gene copies g^{-1} dry weight soil) as affected by production system (Con vs. Org) and input levels (Low vs. High) in a rhodic (Thika) nitisol. F and p-value of the main effects were obtained by ANOVA based on a linear mixed effects model. Tukey post hoc test results are given if significant main effects were found with different letters indicating significant differences at $p < 0.05$.

	16S rRNA gene copies [g^{-1} DW soil]			Archaeal *amoA* gene copies [g^{-1} DW soil]			Bacterial *amoA* gene copies [g^{-1} DW soil]			*nosZ* gene copies [g^{-1} DW soil]		
Rhodic nitisol - Thika	mean	SE		mean	SE		mean	SE		mean	SE	
Con-High	3.76×10^9	1.17×10^9	ab	6.70×10^7	3.37×10^7	–	5.72×10^6	2.49×10^6	–	3.04×10^8	1.21×10^8	–
Con-Low	1.69×10^9	1.55×10^9	a	2.42×10^7	5.71×10^7	–	3.48×10^6	2.57×10^6	–	1.39×10^8	1.85×10^7	–
Org-High	1.31×10^{10}	3.66×10^9	b	1.10×10^8	2.79×10^7	–	1.97×10^7	8.82×10^6	–	8.80×10^8	2.66×10^8	–
Org-Low	4.62×10^9	1.46×10^9	ab	8.87×10^7	3.39×10^7	–	3.48×10^6	2.30×10^6	–	3.98×10^8	1.55×10^8	–

ANOVA	denDF	F-value	p-value	F-value	p-value	F-value	p-value	F-value	p-value
Production system (PS)	9	6.632	0.030	2.805	0.128	1.53	0.247	4.752	0.057
Input levels (IL)	9	4.902	0.054	0.998	0.344	2.666	0.137	2.842	0.126
PS x IL	9	1.803	0.212	0.11	0.747	1.526	0.248	0.687	0.429

Table 4
Impact of farming systems on bacterial alpha-diversity. Data shows mean and standard error (n = 4) of OTU based Shannon diversity, richness and community evenness as affected by production system (Con vs. Org) and input levels (Low vs. High) in a humic (Chuka) nitisol. F and p-value of the main effects were obtained by ANOVA based on a linear mixed effects model. Tukey post hoc test results are given if significant main effects were found with different letters indicating significant differences at $p < 0.05$.

	Exp Shannon			Observed richness			Community evenness		
Humic nitisol - Chuka	mean	SE		mean	SE		mean	SE	
Con-High	223.6	12.1	a	1084.0	63.4	a	0.774	0.006	–
Con-Low	200.2	10.5	a	1067.5	74.9	a	0.760	0.003	–
Org-High	299.4	20.9	b	1796.3	67.9	b	0.759	0.006	–
Org-Low	227.9	13.0	ab	1240.7	108.8	a	0.763	0.007	–

ANOVA	denDF	F-value	p-value	F-value	p-value	F-value	p-value
Production system (PS)	9	9.29	0.013	22.5	0.001	0.78	0.400
Input levels (IL)	9	7.82	0.021	9.40	0.013	0.60	0.458
PS x IL	9	2.01	0.190	8.34	0.017	1.74	0.219

OTUs, and Con-Low with 1067 OTUs.

On the rhodic nitisol at Thika field site, Shannon diversity did not differ between production system or input level (Table 5). Yet, richness showed a similar trend as for the humic nitisol in Chuka with highest richness in Org-High with 1596 OTUs, followed by Con-High with 1264 OTUs, Org-Low 1167 OTUs, and Con-Low with 812 OTUs, with elevated richness in organic production systems ($p = 0.034$) and at High input levels ($p = 0.01$). An inverse trend was observed for community evenness, which was elevated in Con-High and Con-Low compared to Org-High, with production system ($p = 0.06$) and input level ($p = 0.029$) as discerning factors (Table 5).

3.3. Distinct bacterial communities in organic and conventional farming systems

In humic and rhodic nitisols at Chuka and Thika field sites, the

Table 5
Impact of farming systems on bacterial alpha-diversity. Data shows mean and standard error (n = 4) of OTU based Shannon diversity, richness and community evenness as affected by production system (Con vs. Org) and input levels (Low vs. High) in a rhodic (Thika) nitisol. F and p-value of the main effects were obtained by ANOVA based on a linear mixed effects model. Tukey post hoc test results are given if significant main effects were found with different letters indicating significant differences at $p < 0.05$.

	Exp Shannon			Observed richness			Community evenness		
Rhodic nitisol - Thika		mean	SE		mean	SE		mean	SE
Con-High	257.1	17.7	–	1264.7	131.1	ab	0.778	0.007	–
Con-Low	205.6	37.5	–	812.7	129.6	a	0.789	0.011	–
Org-High	216.8	1.6	–	1596.3	121.7	b	0.731	0.007	–
Org-Low	221.6	14.6	–	1167.3	150.6	ab	0.769	0.008	–

ANOVA	denDF	F-value	p-value	F-value	p-value	F-value	p-value
Production system (PS)	9	0.40	0.540	6.34	0.032	12.81	0.006
Input levels (IL)	9	1.55	0.245	10.46	0.010	6.64	0.029
PS x IL	9	2.43	0.153	0.01	0.934	2.08	0.182

interactive effect of the organic production system with high input levels significantly affected bacterial community structure (Table 6). Pairwise PERMANOVA revealed distinct bacterial community structure in Org-High compared to Con-High, Con-Low and Org-Low, while at both field sites no significant difference in bacterial community structure was observed for Con-High, Con-Low and Org-Low (Table 6). Unconstrained and constrained ordination of bacterial community structure via NMDS and db-RDA showed a clear distinction of bacterial communities of Org-High from Con-High, Con-Low and Org-Low at both field sites (Fig. 1).

Correlation of soil physicochemical and biological parameters with bacterial community structure revealed elevated soil pH, dehydrogenase activity, acid phosphatase activity, total and microbial nitrogen, soil organic carbon and microbial carbon to be positively associated with Org-High in the humic nitisol in Chuka (Fig. 1, Table S5). On the rhodic nitisols in Thika, bacterial community structure in Org-High was correlated with enhanced soil basal respiration, total phosphorous contents, and soil aggregate stability (Fig. 1).

3.4. Identifying indicative taxa for distinct farming systems

Identification of farming system specific indicator species in humic nitisol in Chuka resulted in 84 indicative OTUs for Org-High, 10 for Con-High, and 1 for Org-Low, while no indicative OTU was found for Con-Low. In the rhodic nitisol in Thika 160 indicative OTUs were found for Org-High, 1 for Con-High, 2 for Org-Low, and 2 for Con-Low (Fig. 1).

Table 6
Impact of farming systems on microbial beta-diversity. Effects of production systems (Con vs. Org) and input levels (Low vs. High) on bacterial community composition were assessed by PERMANOVA (9999 permutations) based on euclidean distances of a center-log transformed OTU counts. Lower panel shows *p*-values of pairwise PERMANOVA assessing contrasts between farming systems with false discovery rate *p*-value corrections.

		Humic nitisol - Chuka			Rhodic nitisol - Thika		
PERMANOVA	Df	R^2	F-value	p-value	R^2	F-value	p-value
Production system (PS)	1	0.16	3.49	0.001	0.13	2.68	0.022
Input levels (IL)	1	0.14	2.94	0.008	0.15	3.03	0.015
PS x IL	1	0.11	2.50	0.021	0.11	2.20	0.047

Pairwise PERMANOVA	Con-High	Org-Low	Con-Low	Con-High	Org-Low	Con-Low
Org-High	0.027	0.024	0.030	0.026	0.021	0.035
Con-Low	0.224	0.338		0.502	0.690	
Org-Low	0.089			0.551		

A complete list of indicative OTUs is provided in the supplementary information and relative abundances of the most abundant indicator species for each farming system are shown in Figs. 2 and 3 for humic and rhodic nitisols, respectively.

The three most abundant OTUs associated with Org-High on the Humic nitisol in Chuka were assigned to the families of *Micrococcaceae* (OTU14) and Intrasporangiacea (OTU225) and to the genus *Micromonospora* (OTU29) (Fig. 2). For all three OTUs, the input level was found to be a statistically significant factor for its relative abundance. OTU1792, assigned to the genus of *Massilia,* was associated to Con-High with generally elevated abundances in conventional production systems. The two OTUs indicative for Org-Low were annotated to the order *Chthoniobacterales* (OTU4780) and the genus *Gemmantirosa* (OTU651) and their relative abundances show strong interactive effect of production system and input level. Indicative OTUs for Con-Low assigned to the genus *Gemmatimonas* (OTU532) and the genus Lysinibacillus (OTU775). Whereas elevated relative abundance of *Gemmatimonas* (OTU 532) was found in low compared to high input levels and *Lysinibacillus* (OTU775) occurred with a higher relative abundance under conventional compared to organic production systems.

The four most abundant OTUs associated with Org-High in the rhodic nitisol in Thika were assigned to the family *Micrococcaceae* (OTU8) and the genera *Nocardioides* (OTU25), *Micorvirga* (OTU53), and *Massilia* (OTU816) (Fig. 3). For all OTUs, a statistically significant interaction between production system and input level was found with strongly elevated relative abundance in Org-High compared to Con-High, Con-Low and Org-Low. The three most abundant indicative OTUs associated with Con-High assigned to the genera *Sphingomonas* (OTU41), *Gemmatimonas* (OTU87), and *Nitrosospira* (OTU259). A statistically significant interaction effect between production system and input level was found for OTU41 and OTU87 with higher relative abundance under Con-High compared to the other farming systems. In contrast, *Nitrosospira* (OTU259) was predominantly found in conventionally managed systems. OTU806 assigned to the family of *Gemmatimonadaceae* and was found indicative for Org-Low with generally elevated abundances at low input levels.

4. Discussion

4.1. Effect of production system and input levels on nitrifying and nitrous oxide reducing marker genes

Soil nitrogen cycling is mainly mediated by soil microbial activity and the abundance and activity of nitrogen cycling bacteria can be affected by multiple management practices such as mineral nitrogen fertilization and compost application (Ouyang et al., 2018; Segal et al., 2017). Due to four to five times increased nitrogen inputs in High

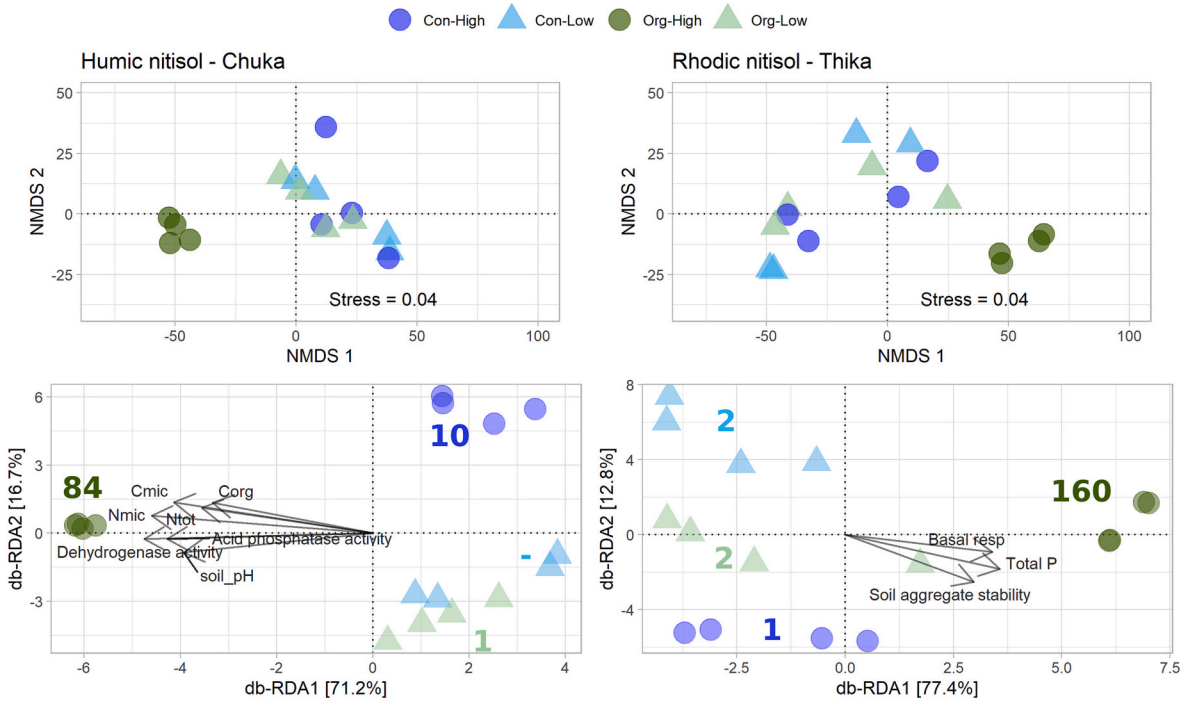

Fig. 1. Impact of farming systems on soil bacterial community composition in humic (Chuka) and rhodic (Thika) nitisols, visualized via unconstrained (NMDS) and constrained (db-RDA) ordination on the base of euclidean distances of center-log transformed OTU counts. Numbers show the amounts of indicative taxa for each farming system and arrows represent correlations of biological and geochemical parameters with the ordination scores. Only correlations with $r^2 > 0.7$ are shown and arrow length is scaled according to correlation strength.

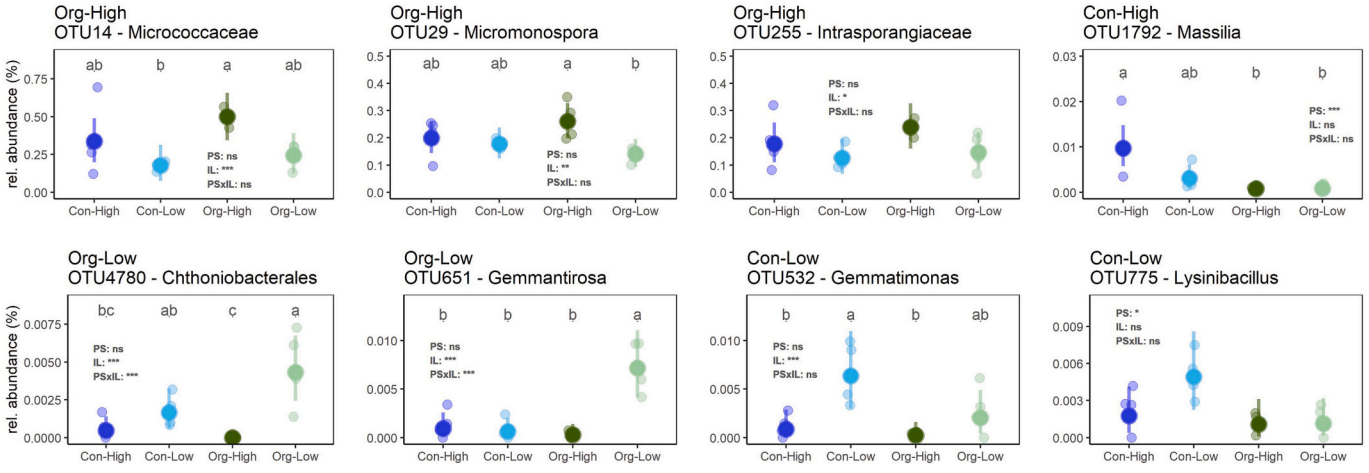

Fig. 2. Effect of farming systems on the relative abundance of the most abundant indicative OTUs in a humic nitisol in Chuka. Data displays individual observations, estimated marginal means and confidence intervals (95 %) and of a linear mixed effect model assessing the impact of production system (PS) and input levels (IL)(n = 4). Different numbers at top of the graphs means significant statistical differences between each farming system (ANOVA, $p < 0.05$). Significant levels for experimental factors are displayed as follows: *** for $p < 0.001$, ** for $p < 0.01$, * for $p < 0.05$ and ns for not significant.

compared to Low input systems (Table 1), enhanced soil microbial capacity for nitrification and nitrous oxide reduction was expected in Org-High and Con-High compared to Org-Low and Con-Low. Yet, we observed enhanced capacity for nitrification and nitrous oxide reduction only for high-input organic production system for the humic nitisol in Chuka but not for the rhodic nitisol in Thika. Greater organic matter inputs via compost in Org-High seem to foster abundance of heterotrophic nitrous oxide reducers in humic nitisols, which is in line with a recent review describing the stimulation of complete denitrification as measure for N_2O mitigation in manure-based systems (Lazcano et al., 2021). Through the continuous release of ammonia nitrogen during decomposition of compost amendments also the abundance and activity of nitrifiers can be stimulated (Ouyang and Norton, 2020). As for nitrous oxide reducers, also this effect seems too dependent on the prevailing soil type, as significantly higher gene copy number of nitrifying marker genes could only be observed for the humic nitisol in Chuka. It needs be noted that bacterial and archaeal *amoA* target genes as proxy for nitrifying capacity has been complemented by the discovery of comammox bacteria (Daims et al., 2015), which might have further added to soils nitrifying capacity but were not accounted for in this study. But also, distinct natural soil fertility levels of humic and rhodic nitisols might have affected microbial response to management practices. Similar to our observations a previous study found more pronounced effects of production systems on key soil quality indicators on humic nitisols in

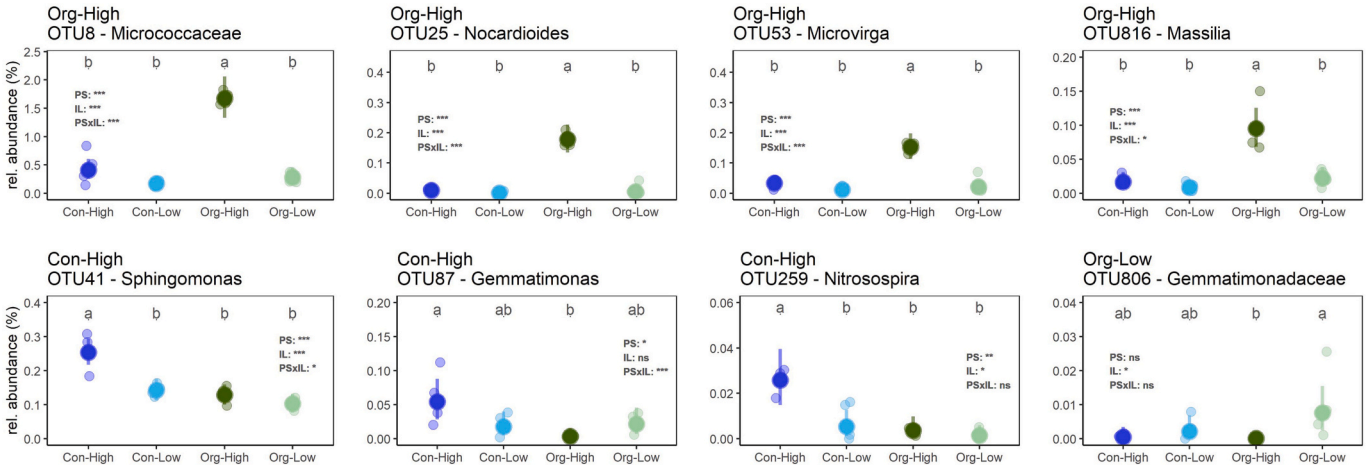

Fig. 3. Effect of farming systems on the relative abundance of the most abundant indicative OTUs oi a rhodic nitisol in Thika. Data displays individual observations, estimated marginal means and confidence intervals (95 %) and of a linear mixed effect model assessing the impact of production system (PS) and input levels (IL)(n = 4). Different numbers at top of the graphs means significant statistical differences between each farming system (ANOVA, $p < 0.05$). Significant levels for experimental factors are displayed as follows: *** for p < 0.001, ** for p < 0.01, * for p < 0.05 and ns for not significant.

Chuka compared to rhodic nitisols in Thika and relate their findings to longer drought periods and generally lower soil biological activity in rhodic compared to humic nitisols (von Arb et al., 2020). Still, high input organic production systems in a rhodic nitisol in Thika led to enhanced 16S rRNA gene copy numbers as a measure for total bacterial abundance (Table 3) which fortifies the finding of higher microbial biomass carbon under high compared to low input levels on both soil types after the third crop rotation period (von Arb et al., 2020). Also, Kiboi et al. (2018) showed that the sole use of organic inputs, like farmyard manure and crop residues, significantly increased microbial biomass carbon and nitrogen in a Kenyan field trial. Summarizing enhanced bacterial abundance and increased capacity for nitrification and nitrous oxide reducing capacity due to high input levels in organic production systems were observed on humic but not rhodic nitisols.

4.2. *Distinct bacterial community composition and enhanced diversity in organic production systems at high input levels*

The observation that soil bacterial community structure differentiated between Org-High and the other production systems on humic and rhodic nitisols in tropical arable farming complements existing studies on soil microbial diversity in organic and conventional farming systems of the temperate climates (Hartmann et al., 2015; Lori et al., 2017). Distinct soil bacterial communities in Org-High coincide with elevated soil pH and Corg contents and might well be linked to organically based fertilization strategy including composting of farmyard manure, mulching, cover cropping and application of *Tithonia* tea (Fig. 1, Table 6). The strong linkage between soil microbial activity and carbon cycling is rooted in the respiration and breakdown of organic matter that serves as energy substrate and allows soil carbon to be transformed in differed carbon pools (Lehmann and Kleber, 2015). In line with this the most abundant OTUs indicative for Org-High on humic (OTU14) and rhodic (OTU8) nitisols were assigned to the genus of *Micrococcaceae*, which is known as primary decomposer with strong cellulolytic activity (Taylor et al., 2012) and was reported to be among the first degraders of crop residues (De la Cruz-Barrón et al., 2017; Ortiz-Cornejo et al., 2017). Additionally, OTU25, assigned to the genus *Nocardioides*, was found to be active in straw decomposition (Guo et al., 2022) and indicative for Org-High on the humic nitisol in Chuka. Hence, indicative taxa show that the high amount of organic matter inputs and remaining crop residues in organic production systems foster bacterial decomposers and increased capacity for decomposition in soils under organic production system at high input levels (Table 1).

OTUs assigning to the genus of *Massilia* (OTU1792 and OTU816) were found to be especially abundant under high input levels on humic and rhodic nitisols in Chuka and Thika, respectively. This genus is known for its root colonizing activity, thrives under copiotrophic conditions and is sensitive to soil organic matter amendments (Ofek et al., 2012). On humic nitisol in Chuka, the abundance of OTU29, was significantly elevated in Org-High and representatives of this genus (*Micromonospora*) are known to act as plant growth promoting bacteria (Trujillo et al., 2015). *Micromonospora* can form symbiotic relationship with leguminous and actinorhizal plants and previous studies identified *Micromonospora* as major component of nitrogen-fixing root nodules across several climates (Martínez-Hidalgo et al., 2014; Trujillo et al., 2010). On humic nitisol in Chuka, soil bacterial community structure in Org-High was positively associated with soil parameters reflecting enhanced soil microbial activity, such as phosphatase and dehydrogenase (Fig. 1, Table S5). In line with that, a global meta-analysis found that organic management particularly enhanced soil microbial activity parameters, such as protease activity by up to 83 % (Lori et al., 2017).

In rhodic nitisols in Thika soil aggregate stability was linked to bacterial community structure in Org-High, (Fig. 2, Table S5), which indicates that the soil microbiota interacts with physical soil structure in less fertile soil conditions. Possibly, raising Corg contents and irrigation practice in high input farming reduced water stress under rather arid conditions in Thika, as indicated by Bogati and Walczak (2022) who found that drought stress declines microbial activity and ultimately affects soil nutrient cycling.

Conventional production systems, especially at high input levels, in rhodic nitisol in Thika yielded increased abundances of OTU259, which was assigned to the genus of *Nitrosospira*. High abundance of this genus suggests high availability of mineralized nitrogen such as ammonium (Lin et al., 2018). As, soil nitrogen supply in Org-High need to be covered through organic matter mineralization or symbiotic nitrogen fixation, it is not surprising that, OTU53 which assigned to the *Microvirga,* was described for its capacity for symbiotic nitrogen fixation under semiarid conditions (Radl et al., 2014). Hence indicative taxa in high input organic and conventional farming hint towards distinct strategies for nitrogen provision, with enhanced mineralization capacity in Org-High. In line with this Musyoka et al. (2019) found highest nitrogen release from amended soils in Org-High during a season experiment on potato, maize and vegetables.

Summarizing, organic production systems shape bacterial community structure at high input levels and promote abundance of organic matter decomposing and symbiotic nitrogen fixing bacteria.

4.3. Enhanced bacterial richness in organically managed arable farming systems

Although, humic nitisols present in Chuka are considered as soil types with higher inherent soil fertility, in line with a previous study of Karanja et al. (2020), we found generally higher bacterial richness in rhodic nitisols in Thika (Table 3). However, in contrast to Karanja et al. (2020), we found the highest bacterial richness under Org-High for humic and rhodic nitisols (Table 3), which might be explained by different ecological target groups (prokaryotes vs. bacteria) or sampling four years later at a different stage of the crop rotation (Chamberlain et al., 2020). As indicated by the ongoing temporal development of soil organic carbon contents (von Arb et al., 2020), soil biogeochemical processes might not have reached a new steady-state conditions after the setup of the field trial 12 years ago. Yet, considering the important role of soil organic carbon turnover as the energetic base for the soil microbial community, raising Corg contents in Org-High are well in line with enhanced bacterial richness in Org-High (Table 3). Long-term field observations in the temperate climate demonstrated increased soil bacterial richness in organic compared to conventional arable cropping systems (Hartmann et al., 2015; Lupatini et al., 2017), and Durrer et al. (2021) found distinct bacterial communities but similar richness in an organic arable cropping system in the subtropical climate.

Interestingly, community evenness declined under Org-High in rhodic nitisol in Thika, suggesting the presence of highly abundant bacterial OTUs to dominating soil bacterial community. Indeed, effect of Org-High on indicative bacterial OTUs was especially pronounced in rhodic nitisols in Thika compared to humic nitisols in Chuka (Figs. 2-3). This indicates that soil bacterial communities show a stronger response to organic production systems on low inherent soil quality of rhodic compared to humic nitisols.

5. Conclusion

In conclusion, we could show that organic production systems at high input levels increased soil bacterial richness and shaped community structure on humic and rhodic nitisols in tropical arable farming systems. Also, microbial capacity for nitrification and nitrous oxide reduction was enhanced in the organic high input systems. Archaeal and bacterial nitrifying capacity was increased by a factor of 2 and 10, respectively, in humic nitisols. Based on taxonomic identification, the ecological niches of most abundant indicator species in the organic high input system were linked to organic matter mineralization, which suggests the use of organic inputs (including cover cropping) as the major drivers for shifts in bacterial community composition. Consequently, this study shows that enhanced bacterial diversity and capacity for nitrogen cycling can be achieved through the integration of organic management principles, but relies on high organic input levels in tropical arable farming systems.

Declaration of competing interest

The authors declare that they have no competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Data availability

Data will be made available on request.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.apsoil.2023.105054.

References

Adamtey, N., Musyoka, M.W., Zundel, C., Cobo, J.G., Karanja, E., Fiaboe, K.K.M., Muriuki, A., Mucheru-Muna, M., Vanlauwe, B., Berset, E., Messmer, M.M., Gattinger, A., Bhullar, G.S., Cadisch, G., Fliessbach, A., Mäder, P., Niggli, U., Foster, D., 2016. Productivity, profitability and partial nutrient balance in maize-based conventional and organic farming systems in Kenya. Agric. Ecosyst. Environ. 235, 61–79. https://doi.org/10.1016/j.agee.2016.10.001.

Adhikari, K., Hartemink, A.E., 2016. Linking soils to ecosystem services - a global review. Geoderma 262, 101–111. https://doi.org/10.1016/j.geoderma.2015.08.009.

von Arb, C., Bünemann, E.K., Schmalz, H., Portmann, M., Adamtey, N., Musyoka, M.W., Frossard, E., Fliessbach, A., 2020. Soil quality and phosphorus status after nine years of organic and conventional farming at two input levels in the Central Highlands of Kenya. Geoderma 362, 114112. https://doi.org/10.1016/j.geoderma.2019.114112.

Bahram, M., Hildebrand, F., Forslund, S.K., Anderson, J.L., Soudzilovskaia, N.A., Bodegom, P.M., Bengtsson-Palme, J., Anslan, S., Coelho, L.P., Harend, H., Huerta-Cepas, J., Medema, M.H., Maltz, M.R., Mundra, S., Olsson, P.A., Pent, M., Põlme, S., Sunagawa, S., Ryberg, M., Tedersoo, L., Bork, P., 2018. Structure and function of the global topsoil microbiome. Nature 560, 233–237. https://doi.org/10.1038/s41586-018-0386-6.

Bogati, K., Walczak, M., 2022. The impact of drought stress on soil microbial community, enzyme activities and plants. Agronomy 12, 1–26. https://doi.org/10.3390/agronomy12010189.

Bünemann, E.K., Bongiorno, G., Bai, Z., Creamer, R.E., De Deyn, G., de Goede, R., Fleskens, L., Geissen, V., Kuyper, T.W., Mäder, P., Pulleman, M., Sukkel, W., van Groenigen, J.W., Brussaard, L., 2018. Soil quality – a critical review. Soil Biol. Biochem. 120, 105–125. https://doi.org/10.1016/j.soilbio.2018.01.030.

Canfield, D.E., Glazer, A.N., Falkowski, P.G., 2010. The evolution and future of earth's nitrogen cycle. Science 1979 (330), 192–196. https://doi.org/10.1126/science.1186120.

Chamberlain, L.A., Bolton, M.L., Cox, M.S., Suen, G., Conley, S.P., Ané, J.M., 2020. Crop rotation, but not cover crops, influenced soil bacterial community composition in a corn-soybean system in southern Wisconsin. Appl. Soil Ecol. 154, 103603 https://doi.org/10.1016/j.apsoil.2020.103603.

Daims, H., Lebedeva, E.V., Pjevac, P., Han, P., Herbold, C., Albertsen, M., Jehmlich, N., Palatinszky, M., Vierheilig, J., Bulaev, A., Kirkegaard, R.H., Von Bergen, M., Rattei, T., Bendinger, B., Nielsen, P.H., Wagner, M., 2015. Complete nitrification by Nitrospira bacteria. Nature 528, 504–509. https://doi.org/10.1038/nature16461.

De Cáceres, M., Legendre, P., Wiser, S.K., Brotons, L., 2012. Using species combinations in indicator value analyses. Methods Ecol. Evol. 3, 973–982. https://doi.org/10.1111/j.2041-210X.2012.00246.x.

De la Cruz-Barrón, M., Cruz-Mendoza, A., Navarro–Noya, Y.E., Ruiz-Valdiviezo, V.M., Ortíz-Gutiérrez, D., Ramírez-Villanueva, D.A., Luna-Guido, M., Thierfelder, C., Wall, P.C., Verhulst, N., Govaerts, B., Dendooven, L., 2017. The bacterial community structure and dynamics of carbon and nitrogen when maize (Zea mays L.) and its neutral detergent fibre were added to soil from Zimbabwe with contrasting management practices. Microb. Ecol. 73, 135–152. doi:https://doi.org/10.1007/s00248-016-0807-8.

Delgado-Baquerizo, M., Oliverio, A.M., Brewer, T.E., Benavent-González, A., Eldridge, D. J., Bardgett, R.D., Maestre, F.T., Singh, B.K., Fierer, N., 2018. A global atlas of the dominant bacteria found in soil. Science 1979 (359), 320–325. https://doi.org/10.1126/science.aap9516.

Domeignoz-Horta, L.A., Putz, M., Spor, A., Bru, D., Breuil, M.C., Hallin, S., Philippot, L., 2016. Non-denitrifying nitrous oxide-reducing bacteria - an effective N2O sink in soil. Soil Biol. Biochem. 103, 376–379. https://doi.org/10.1016/j.soilbio.2016.09.010.

Durrer, A., Gumiere, T., Zagatto, M.R.G., Feiler, H.P., Silva, A.M.M., Longaresi, R.H., Homma, S.K., Cardoso, E.J.B.N., 2021. Organic farming practices change the soil bacteria community, improving soil quality and maize crop yields. PeerJ 9, 1–24. https://doi.org/10.7717/peerj.11985.

Edgar, R.C., 2010. Search and clustering orders of magnitude faster than BLAST. Bioinformatics 26, 2460–2461. https://doi.org/10.1093/bioinformatics/btq461.

Edgar, R.C., 2013. UPARSE: highly accurate OTU sequences from microbial amplicon reads. Nat. Methods 10, 996–998. https://doi.org/10.1038/nmeth.2604.

Edgar, R., 2016. SINTAX: a simple non-Bayesian taxonomy classifier for 16S and ITS sequences. bioRxiv 074161.

Fierer, N., Wood, S.A., Bueno de Mesquita, C.P., 2021. How microbes can, and cannot, be used to assess soil health. Soil Biol. Biochem. 153, 108111 https://doi.org/10.1016/j.soilbio.2020.108111.

Fowler, D., Coyle, M., Skiba, U., Sutton, M.A., Cape, J.N., Reis, S., Sheppard, L.J., Jenkins, A., Grizzetti, B., Galloway, J.N., Vitousek, P., Leach, A., Bouwman, A.F.,

Butterbach-Bahl, K., Dentener, F., Stevenson, D., Amann, M., Voss, M., 2013. The global nitrogen cycle in the Twentyfirst century. Philos. Trans. R. Soc. B Biol. Sci. 368 https://doi.org/10.1098/rstb.2013.0164.
Gloor, G.B., Macklaim, J.M., Pawlowsky-Glahn, V., Egozcue, J.J., 2017. Microbiome datasets are compositional: and this is not optional. Front. Microbiol. 8, 1–6. https://doi.org/10.3389/fmicb.2017.02224.
Guo, T., Zhang, Q., Song, D., Ai, C., Zhang, S., Yue, K., Huang, S., Zhou, W., 2022. Varying microbial utilization of straw-derived carbon with different long-term fertilization regimes explored by DNA stable-isotope probing. Eur. J. Soil Biol. 108, 103379 https://doi.org/10.1016/j.ejsobi.2021.103379.
Hartmann, M., Six, J., 2023. Soil structure and microbiome functions in agroecosystems. Nat. Rev. Earth Environ. 4, 4–18. https://doi.org/10.1038/s43017-022-00366-w.
Hartmann, M., Frey, B., Mayer, J., Mäder, P., Widmer, F., 2015. Distinct soil microbial diversity under long-term organic and conventional farming. ISME J. 9, 1177–1194. https://doi.org/10.1038/ismej.2014.210.
Jones, C.M., Graf, D.R.H., Bru, D., Philippot, L., Hallin, S., 2013. The unaccounted yet abundant nitrous oxide-reducing microbial community: a potential nitrous oxide sink. ISME J. 7, 417–426. https://doi.org/10.1038/ismej.2012.125.
Karanja, E.N., Fliessbach, A., Adamtey, N., Kambura, A.K., Musyoka, M., Fiaboe, K., Mwirichia, R., 2020. Diversity and structure of prokaryotic communities within organic and conventional farming systems in central highlands of Kenya. PLoS One 15. https://doi.org/10.1371/journal.pone.0236574.
Kiboi, M.N., Ngetich, K.F., Mugendi, D.N., Muriuki, A., Adamtey, N., Fliessbach, A., 2018. Microbial biomass and acid phosphomonoesterase activity in soils of the Central Highlands of Kenya. Geoderma Regional 15, e00193. https://doi.org/10.1016/j.geodrs.2018.e00193.
Kolb, S., Knief, C., Stubner, S., Conrad, R., 2003. Quantitative detection of methanotrophs in soil by novel. Society 69, 2423–2429. https://doi.org/10.1128/AEM.69.5.2423.
Krause, H.M., Stehle, B., Mayer, J., Mayer, M., Steffens, M., Mäder, P., Fliessbach, A., 2022. Biological soil quality and soil organic carbon change in biodynamic, organic, and conventional farming systems after 42 years. Agron. Sustain. Dev. 42 https://doi.org/10.1007/s13593-022-00843-y.
Lazcano, C., Zhu-Barker, X., Decock, C., 2021. Effects of organic fertilizers on the soil microorganisms responsible for n2o emissions: a review. Microorganisms 9, 1–18. https://doi.org/10.3390/microorganisms9050983.
Lehmann, J., Kleber, M., 2015. The contentious nature of soil organic matter. Nature 528, 60–68. https://doi.org/10.1038/nature16069.
Lehmann, J., Bossio, D.A., Kögel-Knabner, I., Rillig, M.C., 2020. The concept and future prospects of soil health. Nat. Rev. Earth Environ. 1, 544–553. https://doi.org/10.1038/s43017-020-0080-8.
Lin, Y., Ye, G., Luo, J., Di, H.J., Liu, D., Fan, J., Ding, W., 2018. Nitrosospira cluster 8a plays a predominant role in the nitrification process of a subtropical Ultisol under long-term inorganic and organic fertilization. Appl. Environ. Microbiol. 84, 1–15. https://doi.org/10.1128/AEM.01031-18.
Lori, M., Symnaczik, S., Mäder, P., De Deyn, G., Gattinger, A., 2017. Organic farming enhances soil microbial abundance and activity—a meta-analysis and meta-regression. PLoS One 12. https://doi.org/10.1371/journal.pone.0180442.
Lu, C., Tian, H., 2017. Global nitrogen and phosphorus fertilizer use for agriculture production in the past half century: shifted hot spots and nutrient imbalance. Earth Syst. Sci. Data 9, 181–192. https://doi.org/10.5194/essd-9-181-2017.
Lupatini, M., Korthals, G.W., de Hollander, M., Janssens, T.K.S., Kuramae, E.E., 2017. Soil microbiome is more heterogeneous in organic than in conventional farming system. Front. Microbiol. 7, 1–13. https://doi.org/10.3389/fmicb.2016.02064.
Martínez-Hidalgo, P., Galindo-Villardón, P., Igual, J.M., Martínez-Molina, E., 2014. Micromonospora from nitrogen fixing nodules of alfalfa (Medicago sativa L.). a new promising plant probiotic bacteria. Sci. Rep. 4 https://doi.org/10.1038/srep06389.
McMurdie, P.J., Holmes, S., 2013. Phyloseq: an R package for reproducible interactive analysis and graphics of microbiome census data. PLoS One 8. https://doi.org/10.1371/journal.pone.0061217.
Musyoka, M.W., Adamtey, N., Bünemann, E.K., Muriuki, A.W., Karanja, E.N., Mucheru-Muna, M., Fiaboe, K.K.M., Cadisch, G., 2019. Nitrogen release and synchrony in organic and conventional farming systems of the Central Highlands of Kenya. Nutr. Cycl. Agroecosyst. 113, 283–305. https://doi.org/10.1007/s10705-019-09978-z.
Ofek, M., Hadar, Y., Minz, D., 2012. Ecology of root colonizing Massilia (Oxalobacteraceae). PLoS One 7. https://doi.org/10.1371/journal.pone.0040117.
Oksanen, A.J., Blanchet, F.G., Kindt, R., Legen-, P., Minchin, P.R., Hara, R.B.O., Simpson, G.L., Solymos, P., Stevens, M.H.H., 2012. Community Ecology Package. … Ecology Package …, 263.
Ortiz-Cornejo, N.L., Romero-Salas, E.A., Navarro-Noya, Y.E., González-Zúñiga, J.C., Ramirez-Villanueva, D.A., Vásquez-Murrieta, M.S., Verhulst, N., Govaerts, B., Dendooven, L., Luna-Guido, M., 2017. Incorporation of bean plant residue in soil with different agricultural practices and its effect on the soil bacteria. Appl. Soil Ecol. 119, 417–427. https://doi.org/10.1016/j.apsoil.2017.07.014.
Ouyang, Y., Norton, J.M., 2020. Short-term nitrogen fertilization affects microbial community composition and nitrogen mineralization functions in an agricultural soil. Appl. Environ. Microbiol. 86 https://doi.org/10.1128/AEM.02278-19.
Ouyang, Y., Reeve, J.R., Norton, J.M., 2018. Soil enzyme activities and abundance of microbial functional genes involved in nitrogen transformations in an organic farming system. Biol. Fertil. Soils 54, 437–450. https://doi.org/10.1007/s00374-018-1272-y.
Philippot, L., Hallin, S., Schloter, M., 2007. Ecology of denitrifying prokaryotes in agricultural soil. Adv. Agron. 96, 249–305. https://doi.org/10.1016/S0065-2113(07)96003-4.
Pinheiro, J., Bates, D., R Core Team, 2022. nlme: Nonlinear Mixed Effects Models.
Quast, C., Pruesse, E., Yilmaz, P., Gerken, J., Schweer, T., Yarza, P., Peplies, J., Glöckner, F.O., 2013. The SILVA ribosomal RNA gene database project: improved data processing and web-based tools. Nucleic Acids Res. 41, 590–596. https://doi.org/10.1093/nar/gks1219.
Radl, V., Simões-Araújo, J.L., Leite, J., Passos, S.R., Martins, L.M.V., Xavier, G.R., Rumjanek, N.G., Baldani, J.I., Zilli, J.E., 2014. Microvirga vignae sp. nov., a root nodule symbiotic bacterium isolated from cowpea grown in semi-arid Brazil. Int. J. Syst. Evol. Microbiol. 64, 725–730. https://doi.org/10.1099/ijs.0.053082-0.
Ramankutty, N., Mehrabi, Z., Waha, K., Jarvis, L., Kremen, C., Herrero, M., Rieseberg, L. H., 2018. Trends in global agricultural land use: implications for environmental health and food security. Annu. Rev. Plant Biol. 69, 789–815. https://doi.org/10.1146/annurev-arplant-042817-040256.
Rasche, F., Cadisch, G., 2013. The molecular microbial perspective of organic matter turnover and nutrient cycling in tropical agroecosystems - what do we know? Biol. Fertil. Soils 49, 251–262. https://doi.org/10.1007/s00374-013-0775-9.
Segal, L.M., Miller, D.N., McGhee, R.P., Loecke, T.D., Cook, K.L., Shapiro, C.A., Drijber, R.A., 2017. Bacterial and archaeal ammonia oxidizers respond differently to long-term tillage and fertilizer management at a continuous maize site. Soil Tillage Res. 168, 110–117. https://doi.org/10.1016/j.still.2016.12.014.
Seufert, V., Ramankutty, N., 2017. Many shades of gray—the context-dependent performance of organic agriculture. Sci. Adv. 3 https://doi.org/10.1126/sciadv.1602638.
Sundberg, C., Al-Soud, W.A., Larsson, M., Alm, E., Yekta, S.S., Svensson, B.H., Sørensen, S.J., Karlsson, A., 2013. 454 pyrosequencing analyses of bacterial and archaeal richness in 21 full-scale biogas digesters. FEMS Microbiol. Ecol. 85, 612–626. https://doi.org/10.1111/1574-6941.12148.
Taylor, C.R., Hardiman, E.M., Ahmad, M., Sainsbury, P.D., Norris, P.R., Bugg, T.D.H., 2012. Isolation of bacterial strains able to metabolize lignin from screening of environmental samples. J. Appl. Microbiol. 113, 521–530. https://doi.org/10.1111/j.1365-2672.2012.05352.x.
Tilman, D., Balzer, C., Hill, J., Befort, B.L., 2011. Global food demand and the sustainable intensification of agriculture. Proc. Natl. Acad. Sci. U. S. A. 108, 20260–20264. https://doi.org/10.1073/pnas.1116437108.
Trujillo, M.E., Alonso-Vega, P., Rodríguez, R., Carro, L., Cerda, E., Alonso, P., Martínez-Molina, E., 2010. The genus Micromonospora is widespread in legume root nodules: the example of Lupinus angustifolius. ISME J. 4, 1265–1281. https://doi.org/10.1038/ismej.2010.55.
Trujillo, M.E., Riesco, R., Benito, P., Carro, L., 2015. Endophytic actinobacteria and the interaction of Micromonospora and nitrogen fixing plants. Front. Microbiol. 6, 1–15. https://doi.org/10.3389/fmicb.2015.01341.
Wagg, C., Hautier, Y., Pellkofer, S., Banerjee, S., Schmid, B., van der Heijden, M.G.A., 2021. Diversity and asynchrony in soil microbial communities stabilizes ecosystem functioning. Elife 10, 1–19. https://doi.org/10.7554/eLife.62813.
Willer, Helga, Jan Trávníček, C.M., B.S., 2021. The world of organic agriculture. Statistics and emerging trends 2021. In: Research Institute of Organic Agriculture FiBL, Frick, and IFOAM – Organics International, Bonn (v20210301).