



# Article Soil Macroinvertebrate Response to Paddy Rice Farming Pathways in Mpologoma Catchment, Uganda

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Abstract: Agricultural practices play a major role in influencing soil fauna abundance and diversity. Interest in these practices has increased owing to the growing need for sustainable agricultural systems in this era of increasing agricultural intensification. In this study, two paddy rice farming pathways (smallholder and large-scale commercial) and an adjacent natural wetland in Mpologoma catchment were studied to determine the response of soil macroinvertebrates to paddy rice farming pathways. Eighteen macroinvertebrate taxa were observed, some of which were not the usual soil taxa (Hirudinea, Decapoda, Ephemeroptera, Trichoptera, and Odonata). SIMPER analysis showed that Oligochaeta, Gastropoda, and Coleoptera were the major taxa responsible for dissimilarity among sites. Macroinvertebrate richness and diversity also varied among sites. Some taxa showed habitat exclusivity: Diptera, Odonata, and Trichoptera were exclusive to both rice paddies; Decapoda, Chilopoda, Diplopoda, and Blattodea to natural wetland; Diplura and Ephemeroptera were exclusive to large-scale commercial paddies. NMDS ordination showed that macroinvertebrate distribution among sites was strongly correlated with soil pH and calcium and moderately correlated with phosphorus. These results indicate that wetland conversion to rice paddies could affect macroinvertebrate richness and diversity and underscore the importance of soil environment in influencing the macroinvertebrate community in rice paddies.

Keywords: soil fauna; soil quality; agricultural systems; macroinvertebrates; rice paddies; wetlands

# 1. Introduction

Establishing sustainable food production systems is a major global concern, and the need to balance the social, economic, and environmental aspects of crop production has attracted vast research in the area of agricultural sustainability [1,2]. In the rice production sector, however, much of the attention has been focused on yield maximization to meet the high rice demand for nutrition and poverty reduction [3–9]. Research on the environmental impacts of rice production has been limited [10]. In particular, the response of soil invertebrates to paddy rice farming pathways has received negligible focus, yet these invertebrates play a key role in leveraging sustainable paddy rice production [11,12].

Soil invertebrates range in size from the small microfauna (average size < 0.2 mm), such as nematodes, through the medium-sized mesofauna (0.2–2 mm), such as microarthropods and enchytraeids, to the largest macrofauna (>2 mm), such as arthropods, molluscs, annelids, and crustaceans [13,14]. By decomposing organic matter, modifying soil structure, and mediating nutrient cycling, among other functions, soil invertebrates enhance soil



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**Copyright:** © 2022 by the authors. Licensee MDPI, Basel, Switzerland. This article is an open access article distributed under the terms and conditions of the Creative Commons Attribution (CC BY) license (https:// creativecommons.org/licenses/by/ 4.0/). quality for sustainable crop production [13–19]. However, conservation of soil invertebrates continues to be a challenge in agricultural landscapes. This is largely attributed to their dwelling in the surface litter or in nests and burrows, such that they create in the top 20 cm of the soil profile, where they are prone to physical, chemical, or biological disturbance [13,20,21]. The sensitivity to disturbance is of utmost ecological significance in the function of soil invertebrates as bioindicators [22–25], and changes in their community characteristics can provide valuable feedback on prevailing soil management practices.

Many authors have highlighted the effects of conventional farm management practices on soil invertebrates [26–29]. They have shown how agricultural practices, such as tillage and field traffic (machine compaction), can affect soil invertebrate populations and reduce crop yields [30–32]. Irrigation has also been reported to reduce soil arthropod abundance: Menta et al. [20] observed that Acari, Collembola, and Hymenopteran numbers were higher in corn and wheat fields which had conservation (non-irrigated) soil management practices than in fields with conventional (irrigated) soil management practices. John et al. [33] also observed that microarthropod (mite and collembola) preferred a non-flooded crop rotation environment, unlike the enchytraeids. Similarly, agrochemical use is also reported to interfere with the soil invertebrate environment. For instance, Förster et al. [34] observed that the use of fungicide carbendazim and insecticide lambda-cyhalothrin reduces millipede *Trigoniulus corallinus* and earthworm *Pontoscolex corethrurus* abundance in terrestrial systems. However, it should be noted that, although the majority of the studies have reported detrimental effects, not all conventional farm management practices cause negative effects on soil biodiversity. For example, nitrogen (N) fertilizer addition has been demonstrated to increase mesostigmata mite richness and collembolan abundance, whereas reduced soil disturbance increased the species richness, abundance, and diversity of oribatid mites and collembola [35].

Most studies on the effect of management practice on soil biodiversity in croplands have been carried out in terrestrial areas. Studies targeting wetland environments and rice paddies in particular are few and have mainly focused on the effect of crop rotation [33,36]. No study has dealt with the response of soil macroinvertebrates to paddy rice farming pathways. A farming pathway is defined, in this study, as the course of agricultural management practices followed on a particular farm during a cropping cycle. Accordingly, two categories of paddy rice farming pathways are recognized in the paddy rice farming system: subsistence smallholder pathway and large-scale commercial farming pathway. These pathways differ in many respects including extent of mechanization, sown crop varieties, irrigation, technical agronomic services (fertilizer, pesticide, extension services), and farm size [2,37,38]. Subsistence smallholder farmers cultivate a threshold of 2 ha of land mainly for home consumption and are generally more resource constrained than their large-scale commercial counterparts [38–40]. We assumed that the management practices along each of these paddy rice farming pathways could have unique implications on the community characteristics of the soil macroinvertebrates.

Therefore, in this study, we investigated the response of soil macroinvertebrate taxa in terms of occurrence frequency, richness, abundance, evenness, and diversity in the different paddy rice farming pathways. Macroinvertebrate densities were further correlated with soil characteristics, such as pH, organic matter, nitrogen, phosphorus, potassium, and texture. We hypothesized that the frequency, abundance, richness, evenness, and diversity of macroinvertebrates would reduce along land use intensification gradient from natural wetland, through smallholder paddies, to large-scale commercial paddies. Furthermore, since paddies have unique soil and hydrological conditions, their macroinvertebrate community would be expected to at least encompass some terrestrial or aquatic taxa, such as the arthropod larvae and pupae.

# 2. Materials and Methods

# 2.1. Study Sites

This study was carried out in Mpologoma catchment, a renowned paddy rice growing watershed, in eastern Uganda (Figure 1). The catchment derives its name from River Mpologoma, which forms the major drainage network in the region. In the Mpologoma River's drainage system are several wetlands that the local communities have largely converted for rice growing. Majority of the rice paddies are of the smallholder type but there is also one large-scale commercial farm known as the Kibimba Rice Scheme. The Kibimba Rice Scheme is located at latitude 0°32′2462 N and longitude 33°52′9350 E and covers an area of 4350 hectares. It was established in 1973 by the government of Uganda to increase food production [37,41]. However, it is currently a private venture. The farmed area covering 3900 ha is divided into 18 blocks and each block is further subdivided into 4–6 strips of 1–4 plots. This area receives an annual rainfall of between 900–1400 mm and has 2 rainy seasons, late February–May and August–November, with a peak in April [41].

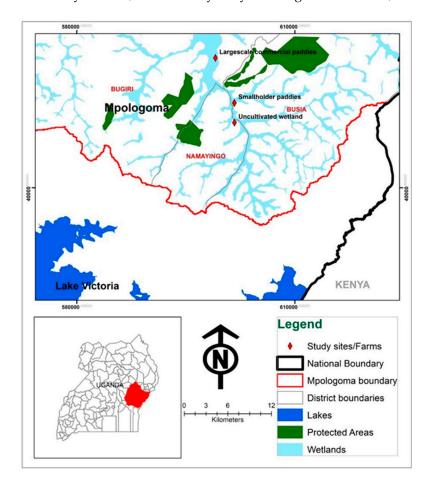


Figure 1. Location of the study sites in Mpologoma catchment.

Rice farming at the large-scale commercial farm (COM) is highly mechanized from land preparation to harvesting. The soil is clayish (Table S3). Ploughing of the land and seeding are mainly carried out twice a year in September–October and March–April. Rice growing is possible throughout the year due to the readily available irrigation water supplied from the reservoir (dam) constructed across River Kibimba. Flooding and drainage of rice fields is regulated through sluice gates constructed across the water canals. Herbicides, fertilizers, and pesticides are regularly used on the farm during the production cycle. The most commonly used herbicides include Glyphosate, which is mainly applied to control volunteer plants that emerge after primary cultivation, and D-Amine for post-emergence weeds in the first weeks of transplanting. The 1st fertilizer application to seedlings is carried out 11 days after sowing seeds in the nursery, while transplanting occurs at 25–28 days. Further fertilizer applications are carried out 5–7 days after transplanting the seedlings, then 30 days later, and the last dose is applied at 60 days after transplanting. The fields are flooded with water from the 65th day and this can go on till the 100th day. Afterwards, the plots are drained in preparation for harvesting about 20 days later. Rice varieties at the commercial farm are the short-duration type (mainly K 23, K 85, and Pusa) that mature between 105 and 120 days. Dimethoate and Beam can be applied when required to control stem borers and leaf fungi, respectively. Harvesting mainly occurs during the June–July and December–January dry seasons and it is carried out by combine harvesters. Fallowing is rare but if any, it may be between 1 and 2 growing seasons.

The smallholder rice paddies (SHD), on the other hand, are rain-fed and largely tilled using hand hoes or ox-ploughs. The soil texture is clay loam (Table S3). The plots are small, some measuring about 50 ft by 100 ft, and a farmer may use one or more plots for rice farming. Slash-and-burn is commonly practiced for field preparation. Planting usually occurs during the rainy seasons of late February–early March and late August–early September and the crop is harvested during the June–July and December–January dry seasons after about  $3^{1}/_{2}$ –4 months. Land preparation, planting, weeding, and rice harvesting rely on family or hired human labor. Agrochemicals, such as fertilizers, herbicides, and pesticides are not used on these farms. After harvest, some farmers immediately hand-till their fields in preparation for the next season, although others may choose to fallow their rice fields for a season or two.

A third sampling site, an uncultivated natural wetland (Uncul), was adopted as a control. The Uncul soil texture is loam (Table S3). The natural wetland is fed by river water flowing into the reservoir that supplies the large-scale commercial farm. It had pristine wetland vegetation cover and was hardly influenced by human activities, like that in the rice paddies. The vegetation mainly comprised *Cyperus papyrus* L., *Cyperus latifolius* Poir., Cycads, *Impatiens tinctoria* A.Rich, *Phragmites australis* (Cav.) Trin and *Echinocloa pyramidalis* (Lam.) Hitch., and Chase.

All the three sites (COM, SHD, and Uncul) were similar by being wetland areas located close to each other within a 10 km radius and had been under the particular land use/cover type and climatic conditions for at least 30 years [42].

## 2.2. Soil Sampling and Macroinvertebrate Extraction

Soil sampling for macroinvertebrate and chemical characteristics was carried out between September 2018 and December 2019. For macroinvertebrate analysis, four replicate plots were randomly selected in the large-scale and small-scale rice paddies for each crop stage: young rice (about one month after transplanting), mature rice (at harvest), and fallow (unplowed rice fields after harvest for at least one season). This was aimed at capturing the variability in macroinvertebrate community within and between farming pathways. In each plot, 3 monoliths measuring  $25 \times 25 \times 15$  cm deep were randomly dug out along a diagonal transect. The exact position of the monoliths was marked out using a wooden quadrat of  $25 \times 25$  cm outside dimensions. In the first 2 days of invertebrate collection in September 2018, monoliths were initially dug to a depth of 30 cm [43,44]. However, because deep layers (15–30 cm) had barely any macroinvertebrates, sampling depth was adjusted to 0–15 cm in the later sampling. Similarly, soil replicates were collected from four randomly selected plots located in the natural wetland. However, since there were not crop stages in the natural wetland, 12 soil samples were collected from the 4 plots. Therefore, in total, 84 soil samples were collected. At each sampling point, litter within the 25 cm quadrat was initially hand-sorted for any surface macroinvertebrates before the monolith was dug out. Then, the monolith was carried to the sorting area, crumbled and all large invertebrates (>5 mm long) and visible under a magnifying glass were carefully hand-sorted and removed with forceps. The soil sample was subsequently mixed with water in a bucket and sieved over a 2 mm sieve to maximize extraction of macroinvertebrates. The collected macroinvertebrates were preserved in 10% formalin and

carried to the laboratory where they were stained with Rose Bengal to ease sorting and counting under a binocular dissecting microscope.

Soil samples for chemical analysis were also collected from the top 0–15 cm adjacent to the sampling locations for soil samples for fauna analysis. The samples were then mixed homogeneously, air-dried, their gravel, roots, and large organic residues were removed, and then they were passed through a 2 mm sieve. Generally, analysis of soil samples for chemical parameters was performed following procedures described in [45]. The parameters measured included pH, texture (%), soil organic matter (%), available phosphorus (ppm), available nitrogen (%), available potassium (cmol/Kg of soil), and available calcium (cmol/Kg of soil). Soil texture was classified following the USDA soil texture triangle classification [46].

#### 2.3. Data Analysis

Macroinvertebrate data were collected in terms of abundance of invertebrate taxa per soil sample. Frequency of occurrence of each taxon was then derived by considering the number of soil samples in which a particular taxon was present against the total number of replicates over the entire study. Community indices, such as richness, evenness, and diversity were also obtained. Diversity and evenness of soil macroinvertebrates were, respectively, calculated using Equations (1) and (2) [47–50], as follows:

$$H' = -\sum_{i=1}^{s} p_i * In(p_i)$$
 (1)

$$J = H' / In(s) \tag{2}$$

where H' is the Shannon–Wiener diversity index, s is the total number of taxa collected,  $p_i$  is the proportion of individuals of taxon  $i(n_i)$  relative to all individuals (N) from all the collected taxa for a particular land use ( $p_i = n_i/N$ ), and J is Pielou's measure of evenness.

Rank–abundance curves were then produced to display the relative abundance of macroinvertebrate taxa for each farming pathway and natural wetland. Macroinvertebrate abundances, richness, evenness, and diversity were further compared across sites and crop stages using one-way ANOVA in R 4.0.4 [51]. This aimed to establish whether or not the differences in means varied among the categories. Count data were standardized to densities (ind./m<sup>2</sup>) [43], normality, and homoscedasticity evaluated using the Shapiro–Wilk test and Levene's test, respectively.

To visualize how farming pathway related with the macroinvertebrate community, non-metric multidimensional scaling (NMDS) was performed based on Bray–Curtis dissimilarity index [52,53]. The results were plotted in an NMDS ordination plane, in a two-dimensional space. Differences in the macroinvertebrate community among the different patterns visualized with NMDS were analyzed using a permutational multivariate analysis of variance (PERMANOVA) test. After a significant PERMANOVA test ( $p \le 0.05$ ), SIMPER (similarity percentages) analysis was performed to examine which invertebrate groups were driving the differences in community among the sites. The significance of contribution of each macroinvertebrate group to the differences and which of the soil environmental factors (pH, SOM, N, P, K, Na, Ca, and texture) were significantly correlated to the first two axes of the NMDS ordination plane were tested using the envfit function. NMDS ordination, PERMANOVA, SIMPER, and envfit function were performed with the vegan package (2.5–7) in R 4.0.4 [51,54].

## 3. Results

## 3.1. Macroinvertebrate Community Composition

A total of 18 macroinvertebrate taxa were observed in the soil samples. They belonged to 3 phyla Annelida, Mollusca, and Arthropoda at 61.3%, 17.9%, and 20.8%, respectively (Table S1). The arthropods were mainly larvae and nymphs. Oligochaeta (57.3%), Gastropoda (17.9%), Coleoptera (12.4%), Hirudinea (4.0%), Hymenoptera (2.1%), Diptera (1.6%),

and Trichoptera (1.1%) were the most abundant groups and constituted about 96% of the macroinvertebrates. Arachnida, Isopoda, Dermaptera, and Orthoptera comprised about 3% while the rest of the taxa Decapoda, Diplopoda, Chilopoda, Diplura, Ephemeroptera, Blattodea, and Odonata totaled less than 1%. In addition, Oligochaeta was the most ubiquitous taxon followed by Coleoptera, Gastropoda, and Hirudinea (Table S2). Macroinvertebrate occurrence frequencies across sites and crop stages did not follow a specific pattern. For instance, oligochaetes occurred in 100%, 97%, and 72% of the soil samples collected from the natural wetland, smallholder paddies, and large-scale commercial paddies, respectively; meanwhile, Coleoptera were mainly found in soil samples from the large-scale commercial paddies (66.7%), followed by natural wetland (58.3%), and least in the smallholder paddies (33.3%). Similarly, the majority of the Gastropods and Hirudinea were encountered in the large-scale commercial paddies (72.2% and 50.0%, respectively), followed by smallholder paddies (33.3% and 38.9%, respectively), and natural wetland (25.0% and 8.3%, respectively). Overall, eight of the taxa were common to all the three sites. One taxon (Isopoda) was common to both large-scale commercial paddies and natural wetland, while three taxa (Diptera, Odonata, and Trichoptera) were common to both large-scale commercial and smallholder paddies. Four taxa (Decapoda, Chilopoda, Diplopoda, and Blattodea) were exclusive to the natural wetland, while two taxa (Diplura and Ephemeroptera) were exclusive to the large-scale commercial paddies. No taxa were exclusive to smallholder paddies and none to both smallholder and natural wetland. Taxa such as Hirudinea, Decapoda, Trichoptera, Ephemeroptera, and Odonata which are not regular soil candidates, especially in terrestrial soils, were also encountered in the samples.

The overall macroinvertebrate density was  $304.8 \pm 327.6$  ind./m<sup>2</sup> and ranged between  $284.0 \pm 210.3$  ind./m<sup>2</sup> (in the natural wetland),  $307.6 \pm 357.2$  ind./m<sup>2</sup> (in the large-scale commercial paddies), and  $308.9 \pm 336.0$  ind./m<sup>2</sup> (in the smallholder farms) (Table 1). Density per replicate ranged between 16 and 1584 ind./m<sup>2</sup> in the large-scale rice paddies, 32 and 1872 ind./m<sup>2</sup> in the smallholder paddies, and 32 and 736 ind./m<sup>2</sup> in the natural wetland. Across the crop stages, macroinvertebrate density was highest in the smallholder harvest ( $353.3 \pm 493$  ind./m<sup>2</sup>) and lowest in the commercial fallow ( $250.7 \pm 247.5$  ind./m<sup>2</sup>). There was high variability in density among replicates and it muffled any significant differences in total density between crop stages and sites. Nevertheless, some taxa showed significant differences in their density ( $253.8 \pm 345.2$  ind./m<sup>2</sup>) in the smallholder paddies ( $202.7 \pm 200.7$  ind./m<sup>2</sup>) and this variation among sites was significant (p < 0.05). Post hoc comparison indicated that the difference was mainly between the smallholder and large-scale pair (p < 0.05).

The second most abundant taxon was Gastropoda. It recorded significantly higher densities (p < 0.05) in the large-scale paddies ( $111.1 \pm 212.7 \text{ ind./m}^2$ ) compared with smallholder rice paddies ( $12.4 \pm 31.6 \text{ ind./m}^2$ ) and natural wetland ( $10.7 \pm 21.9 \text{ ind./m}^2$ ). A post hoc test showed that the difference was also mainly between the smallholder and large-scale paddies (p < 0.05). The other important taxon in terms of density was Coleoptera. Coleoptera had a relatively higher density in the large-scale commercial paddies ( $72.0 \pm 214.4 \text{ ind./m}^2$ ) compared with smallholder rice paddies ( $9.8 \pm 24.9 \text{ ind./m}^2$ ) and natural wetland ( $18.7 \pm 19.1 \text{ ind./m}^2$ ), but the difference among sites was not significant (p > 0.05). The rest of the taxa recorded relatively low densities and the differences were also not significant among sites and crop stages (p > 0.05).

	Crop Stages						Sites			
Таха	Com-You	Com-Har	Com-Fal	Shd-You	Shd-Har	Shd-Fal	Com	Shd	Natural Wetland	Overall
Oligochaeta	$176\pm313.8$	$58.7\pm77.3$	$24.0\pm33.1$	$186.7\pm260.4$	$313.3\pm503.4$	$261.3 \pm 221.8$	$86.2 \pm 193.7$	$253.8\pm345.2$	$202.7\pm200.7$	$174.7\pm278.7$
Hirudinea	$10.7\pm19.7$	$13.3\pm19.1$	$18.7\pm16.5$	$14.7\pm27.7$	$4.0\pm9.9$	$22.7\pm22.1$	$14.2\pm18.3$	$13.8\pm22.0$	$1.3\pm4.6$	$12.2\pm19.2$
Gastropoda	$140\pm300$	$49.3\pm74.9$	$144.0\pm205.1$	$22.7\pm48.9$	$13.3\pm22.5$	$1.3\pm4.6$	$111.1\pm212.7$	$12.4\pm31.6$	$10.7\pm21.9$	$54.5 \pm 148.3$
Arachnida	-	$2.7\pm9.2$	$6.7\pm12.7$	$2.7\pm 6.2$	$1.3\pm4.6$	-	$3.1\pm9.2$	$1.3\pm4.5$	$5.3\pm14.2$	$2.7\pm8.5$
Isopoda	-	$2.7\pm9.2$	-	-	-	-	$0.9 \pm 5.3$	-	$9.3\pm27.7$	$1.7\pm11.1$
Decapoda	-	-	-	-	-	-	-	-	$1.3\pm4.6$	$0.2\pm1.7$
Hymenoptera	$1.3\pm4.6$	$1.3\pm4.6$	-	$8.0\pm19.9$	$12.0\pm32.8$	$5.3 \pm 18.5$	$0.9\pm3.7$	$8.4\pm24.0$	$17.3\pm50.3$	$6.5\pm24.9$
Coleoptera	$6.7\pm8.2$	$164\pm356$	$45.3\pm69.2$	$9.3\pm12.7$	$2.7\pm 6.2$	$17.3\pm40.6$	$72.0\pm214.4$	$9.8\pm24.9$	$18.7\pm19.1$	$37.7 \pm 143.5$
Diptera	$5.3\pm14.2$	$9.3\pm18.6$	$1.3\pm4.6$	$13.3\pm22.5$	$4.0\pm7.2$	$1.3\pm4.6$	$5.3\pm13.8$	$6.2\pm14.5$	-	$5.0\pm13.1$
Dermaptera	-	$6.7\pm10.7$	-	-	$1.3\pm4.6$	-	$2.2\pm 6.8$	$0.4\pm2.7$	$5.3\pm7.9$	$1.9\pm5.5$
Diplura	$1.3\pm4.6$	$1.3\pm4.6$	-	-	-	-	$0.9\pm3.7$	-	-	$0.4\pm2.5$
Chilopoda	-	-	-	-	-	-	-	-	$2.7\pm9.2$	$0.4 \pm 3.5$
Diplopoda	-	-	-	-	-	-	-	-	$1.3\pm4.6$	$0.2\pm1.7$
Ephemeroptera	-	-	$1.3\pm4.6$	-	-	-	$0.4\pm2.7$	-	-	$0.2\pm1.7$
Orthoptera	$1.3\pm4.6$	-	$5.3\pm14.2$	$1.3\pm4.6$	-	$4.0\pm9.9$	$2.2\pm8.7$	$1.8\pm 6.4$	$6.7\pm12.7$	$2.7\pm8.5$
Blattodea	-	-	-	-	-	-	-	-	$1.3\pm4.6$	$0.2\pm1.7$
Odonata	-	$1.3\pm4.6$	$1.3\pm4.6$	-	$1.3\pm4.6$	-	$0.9\pm3.7$	$0.4\pm2.7$	-	$0.6\pm3.0$
Trichoptera	-	$18.7\pm35.3$	$2.7\pm 6.2$	$1.3\pm4.6$	-	-	$7.1\pm21.8$	$0.4\pm2.7$	-	$3.2\pm14.6$
Overall	$342.7\pm401.7$	$329.3\pm404$	$250.7\pm275.5$	$260.0\pm247.5$	$353.3\pm493$	$313.3\pm223.7$	$307.6\pm357.2$	$308.9\pm336.0$	$284.0\pm210.3$	$304.8\pm327.6$

<b>Table 1.</b> Mean density $\pm$ SD (ind.m <sup>-2</sup> )	of soil macroinvertebrate taxa by site and crop stage.

Abbreviations: Com—large-scale commercial paddies; Shd—smallholder paddies; You—young; Har—harvest/mature; Fal—fallow.

## 3.1.1. Richness, Evenness, and Diversity

Macroinvertebrate richness ranged between  $3.47 \pm 1.38$  in the large-scale paddies,  $3.42 \pm 2.07$  in the natural wetland, and  $2.64 \pm 1.22$  in the smallholder paddy soil replicates (Table 2). The difference in richness among sites was significant (p < 0.05) and a post hoc test indicated that it was significant between the smallholder and large-scale paddies pair (p < 0.05). Similarly, the diversity of macroinvertebrates was  $0.85 \pm 0.48$ ,  $0.72 \pm 0.65$ , and  $0.56 \pm 0.44$  in the large-scale commercial paddies, natural wetland, and smallholder paddies, respectively. It significantly varied among sites (p < 0.05) and was also between the smallholder and large-scale commercial paddies pair (p < 0.05). On the other hand, variation in evenness among sites was not significant (p > 0.05).

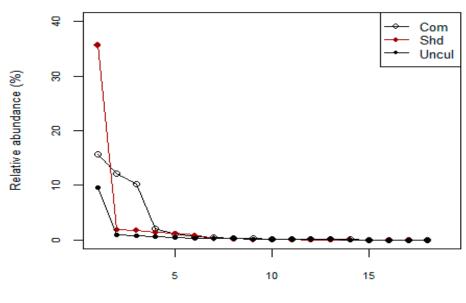
Table 2. Mean  $\pm$  SD richness, evenness and diversity of macroinvertebrates across crop stages and sites.

Index	Crop Stages						Sites		
	Com-You	Com-Har	Com-Fal	Shd-You	Shd-Har	Shd-Fal	Com	Shd	Uncul
Richness per replicate	$2.83 \pm 1.27$	$4.08\pm0.90$	$3.50\pm1.68$	$3.17 \pm 1.47$	$2.25\pm1.29$	$2.50\pm0.67$	$3.47 \pm 1.38$	$2.64 \pm 1.22$	$3.42\pm2.07$
Pileou's Evenness (J')	$0.53\pm0.36$	$0.80\pm0.16$	$0.65\pm0.35$	$0.61\pm0.30$	$0.35\pm0.37$	$0.54\pm0.31$	$0.66\pm0.31$	$0.50\pm0.34$	$0.53\pm0.35$
Shannon's Diversity (H')	$0.59\pm0.46$	$1.10\pm0.32$	$0.87\pm0.53$	$0.77\pm0.46$	$0.39\pm0.48$	$0.52\pm0.31$	$0.85\pm0.48$	$0.56\pm0.44$	$0.72\pm0.65$

Abbreviations: Com—large-scale commercial paddies; Shd—smallholder paddies; You—young; Har—harvest/mature; Fal—fallow.

## 3.1.2. Rank Abundance Relationship

The macroinvertebrate community of the sampled sites was characterized by a few abundant taxa (Figure 2). The steepest slope was for smallholder rice paddies with one dominant taxon, followed by natural wetland with a less steep slope than for smallholder paddies, and also dominated by one taxon, and then the largescale commercial paddies with a shallower gradient but with three relatively more evenly abundant taxa. The majority of the taxa fell in the rare category, arbitrarily defined in this study as a relative abundance of 0.5% or less in a given farming pathway or natural wetland over the entire study.

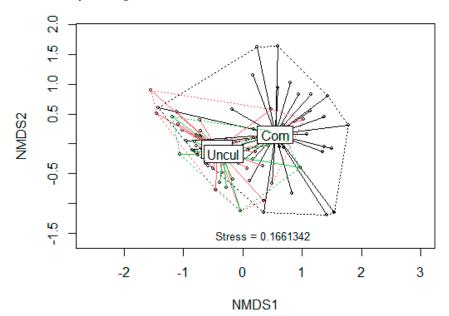


Abundance Rank

**Figure 2.** Rank abundance curves for the soil macroinvertebrates observed in the paddy rice farming pathways in Mpologoma catchment. Abbreviations: Com—large-scale commercial paddies; Shd—smallholder paddies; Uncul—natural wetland.

### 3.1.3. Community Ordination

The macroinvertebrate community in the large-scale commercial rice paddies (COM), smallholder rice paddies (SHD), and natural wetland (Uncul) showed some overlap (Figure 3), especially between SHD and Uncul, and 15% of the variation in communities was due to management differences as confirmed by PERMANOVA ( $p \le 0.001$ ,  $R^2 = 0.151$ ). A subsequent pairwise SIMPER analysis to examine the average contribution of the different macroinvertebrate groups to the overall dissimilarity between site pairs revealed that Oligochaeta, Gastropoda, and Coleoptera were the most influential taxa (Table 3). Oligochaetes contributed largest to the variation in each site pair. In addition, the first 2 invertebrate taxa accounted for over 60% of the dissimilarity in each pairwise site comparison, while with the third taxon, Coleoptera, over 75% of the dissimilarity between sites could be explained. Further analysis of the intrinsic taxa indicated that Oligochaeta, Gastropoda, Coleoptera, and Odonata were the only taxa that had significant contribution to the overall dissimilarity among sites (Table 4).



**Figure 3.** Bray-Curtis based NMDS plot of soil macroinvertebrate community composition. Points represent macroinvertebrate samples condensed on the two axes of NMDS plot. Spider diagrams connect the points to the respective land use type: Com (black), Uncul (green), and Shd (red).

**Table 3.** Cumulative percentage contribution of most influential taxa to the overall similarity between site pairs.

Macroinvertebrate Community Pair	Oligochaeta	Oligochaeta and Gastropoda	Oligochaeta, Gastropoda, and Coleoptera
Com-Shd	47	69	82
Com–Uncul	43	65	78
Shd-Uncul	59	67	74

Abbreviations: Com—large-scale commercial paddies; Shd—smallholder paddies; Uncul—natural wetland.

Table 4. Taxa driving the overall distribution pattern across sites.

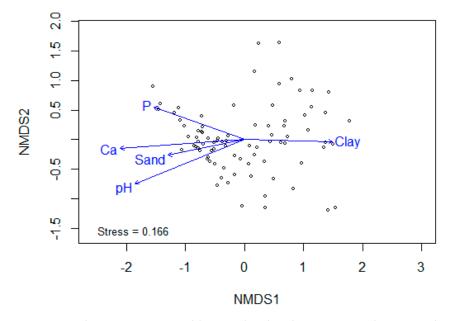
Taxa	Squared Correlation Coefficient, R <sup>2</sup>	<i>p</i> -Value	
Oligochaeta	0.5245	0.001 ***	
Gastropoda	0.3677	0.001 ***	
Coleoptera	0.1871	0.001 ***	
Odonata	0.1477	0.002 **	

Significance codes: \*\* *p* < 0.01, \*\*\* *p* < 0.001.

## 3.2. Soil Environment and Its Relationship with Macroinverterbate Community

Generally, soil characteristics varied across the sites (Table S3). The overall average pH was  $5.73 \pm 0.06$  and ranged between  $5.54 \pm 0.11$  in the large-scale commercial paddies,  $5.81\pm0.08$  in smallholder paddies, and  $6.03\pm0.11$  in natural wetland. The difference in pH among sites was significant (p < 0.05) and the post hoc test showed that it was mainly from the natural wetland and large-scale paddies pair (p < 0.05). Similarly, soil organic matter (SOM), nitrogen (N), phosphorus (p), and calcium (Ca) contents were, respectively,  $37.53 \pm 3.74\%$ ,  $0.46 \pm 0.06\%$ ,  $0.24 \pm 0.02$  ppm, and  $51.64 \pm 11.48$  cmol/Kg in the natural wetland, 26.36  $\pm$  2.5%, 0.46  $\pm$  0.02%, 0.18  $\pm$  0.01 ppm, and 30.63  $\pm$  5.21 cmol/Kg in the smallholder paddies, and  $17.7 \pm 1.38\%$ ,  $0.43 \pm 0.02\%$ ,  $0.13 \pm 0.01$  ppm, and  $8.17 \pm 1.44$  cmol/Kg in the large-scale commercial rice paddies. There were significant differences (p < 0.001) in the variation of phosphorus and calcium among sites. Post hoc test indicated that the differences in *p* and Ca were between the pairs: natural wetland and large-scale paddies (p < 0.001 and p < 0.001, respectively), smallholder and large-scale paddies (p < 0.01 and p < 0.01)p < 0.01, respectively), and natural wetland and smallholder paddies (p < 0.01 and p < 0.1, respectively). Similarly, the percentage of clay varied (p < 0.001) such that it was highest in the large-scale paddies (41.39  $\pm$  1.57%), followed by smallholder paddies (32.56  $\pm$  1.73%), and lowest in the natural wetland (24.50  $\pm$  3.42%). On the other hand, sand also varied (p < 0.01) being highest in the natural wetland (44.25 ± 1.56%), followed by smallholder paddies (40.64  $\pm$  2.31%), and large-scale paddies (33.89  $\pm$  1.56%). Post hoc test showed that the differences in sand and clay composition were between the pairs: smallholder and large-scale commercial paddies (p < 0.05 and p < 0.01, respectively) and natural wetland and large-scale commercial paddies (p < 0.05 and p < 0.001, respectively). No significant differences were observed for SOM, sodium (Na), nitrogen (N), potassium (K), and silt.

Soil environment variables were further correlated with the NMDS ordination scores of the macroinvertebrate community. Soil pH, Ca, P, and sand had a negative correlation while clay content had a positive correlation (Figure 4). Basing on R<sup>2</sup> and the *p*-values to discriminate the strength of the relationship, it was shown that there was a strong correlation for pH and calcium (p < 0.001), moderate intensity correlation for phosphorus (p < 0.01), and a low intensity correlation for clay and sand (p < 0.05) (Table 5). SOM, N, Na, K, and silt were not significantly correlated with the NMDS ordination pattern of the macroinvertebrates in this study.



**Figure 4.** Soil environment variables correlated with macroinvertebrate samples in an ordination system. Points represent samples. Arrows show the significant vectors while their direction represents the gradient direction of the environmental driver.

Variable	<b>R-Squared</b>	<i>p</i> -Value
pH	0.1954	0.001 ***
SOM <sup>1</sup>	0.0231	0.394
N	0.0291	0.305
Р	0.1256	0.007 **
K	0.0398	0.193
Na	0.0397	0.181
Са	0.2156	0.001 ***
Sand	0.0855	0.027 *
Clay	0.1121	0.012 *
Silt	0.0094	0.694

Table 5. Correlation analysis of soil characteristics with NMDS scores.

 $\overline{1}$  SOM—soil organic matter. R-squared—squared correlation coefficient. Significance codes: \* *p*-value < 0.05, \*\* *p*-value < 0.01, \*\*\* *p*-value < 0.001.

# 4. Discussion

The study has shown that macroinvertebrate taxa responded differently to the paddy rice farming pathways in Mpologoma catchment during the 2018–2019 period. In particular, the most ubiquitous oligochaetes were encountered in 100% of the samples from the natural wetland but the frequency reduced in the large-scale commercial and smallholder rice paddies. Similar studies have observed higher or lower frequencies of some oligochaete species in the natural grassland and/or hardwood forest than in the cultivated areas [55]. In the current study, however, oligochaetes were not identified to the lower levels; hence, differences that could arise at species levels were not captured. Nevertheless, a similar trend would be obtained with some oligochaete species showing higher or lower frequencies in the natural wetland compared with the cultivated paddies. In the rice paddies, oligochaete density also varied: it was significantly higher in the smallholder than large-scale commercial paddies. Variation in oligochaete frequency and density among sites could be attributed to the differences in soil conditions. Generally, the magnitude of physical soil disturbance through tillage and chemical disturbance through agrochemical use increases with farming intensity from the pristine natural wetland condition where the soil is largely at rest to the cultivated condition where the soil is regularly worked for farming. Such disturbance has been reported to affect the distribution of soil invertebrates within the soil column and across habitats [21,32,35]. At the large-scale commercial farm, pesticides and inorganic fertilizers are heavily used coupled with high mechanization and associated soil compaction. Conversely, the smallholder paddy rice farmers in this area do not use inorganic fertilizers; they rely on the natural fertility of the alluvial soil deposited by moving water. Soil compaction is also minimal. Therefore, these factors could have made the soil environment at the large-scale commercial farm less conducive to oligochaetes compared with that in the smallholder rice paddies and natural wetland [32,34,56–58]. In addition, the lowest oligochaete density observed in the commercial fallow stage was attributed to the farm management dynamics whereby plots that were not in active agricultural use were not supplied with irrigation water. Consequently, this could have increased the concentration of agrochemical residues in those plots which jeopardized oligochaete survival [59].

Gastropod density also varied among sites, being highest in the large-scale commercial rice paddies and lowest in the natural wetland. Gastropods are largely herbivorous organisms and some are avid feeders on young rice stems [60–62]. Therefore, the stable food supply at the large-scale farm where rice is grown throughout the year coupled with reduced predation from diplopods and chilopods [63] could have been major drivers for the high density of gastropods in the large-scale rice paddies. Additionally, the scaring away of birds by the bird chasers who are employed by the commercial farm could also have significantly contributed to the reduced predation on snails by the predatory birds, such as the African Jacana (Actophilornis africanus) and ducks, among others [41].

Densities for individual arthropod taxa were low except coleoptera and did not significantly vary among sites. Nevertheless, some arthropods showed habitat exclusiveness. In this category were the detritivorous diplopods which were exclusive to the natural wetland. Their exclusivity could be attributed to litter availability because the short fallow periods (usually one season or none) in the cultivated large-scale and smallholder rice paddies coupled with the routine soil disturbance may not have allowed sufficient time for litter to accumulate in these fields compared with the natural wetland where the soil had negligible perturbations. Similarly, Diplurans were only found in the large-scale commercial rice paddies, as previously documented [64]. This preference could be related to high prey abundances, such as insect larvae (e.g., Coleopteran nymphs and Trichopteran larvae), in the large-scale paddies compared with the natural wetland and smallholder rice paddies. Furthermore, some studies have documented high Acari and Collembola abundance in vineyards [20,65] relative to other arthropods, but in our study these taxa had low densities and insignificantly contributed to the dissimilarity among sites. This implies that it is difficult to make generalizations about macroinvertebrate abundances and diversity in varying agricultural systems because the agricultural practices vary and are complex, yet the response of macroinvertebrates might be a habitat-specific occurrence [66].

Land use conversion from natural wetland to cultivated rice fields, especially conversion to the large-scale commercial paddy rice farming, was also associated with increased richness and diversity of macroinvertebrates. Indeed, the rank–abundance relationship confirmed a higher macroinvertebrate evenness in the large-scale rice paddies than other sites. This contrasts with similar studies where diversity of macroinvertebrates decreased with increasing land use intensification [55,67]. The high diversity at the large-scale commercial farm could be attributed to the near-mosaic farm setup, where the bunds of natural vegetation that separate plots, strips, and blocks, and the road network which provides access to the plots, could have created refugia that enhanced macroinvertebrate diversity [68].

From the ordination system, macroinvertebrate communities of smallholder paddies and natural wetland seemed to be highly similar. This was attributed to the high proximity of the smallholder paddies to the natural wetland which could have easily allowed fauna mobility between the two adjacent sites in the landscape [20,69]. Furthermore, the NMDS regression showed that soil pH, calcium, phosphorus, clay, and sand were the major paddy soil variables that had significant relationships with the ordination pattern. Similar studies [65] have reported factors such as soil pH, SOM, and temperature to be important soil environment variables that drive invertebrate distribution, especially arthropods, in vineyards. They found texture to be insignificant; however, we found that texture, especially clay and sand content, were significantly correlated with macroinvertebrate distribution in rice paddies. This implies that the soil environment of rice paddies could be influenced by some unique factors unlike terrestrial croplands, and these would need further exploration. Nevertheless, this study has highlighted that changes in soil properties could play a role in driving the community characteristics of macroinvertebrates in rice paddies.

This study recorded some macroinvertebrate taxa that are not regular soil residents. These included Hirudinea, Decapoda, Ephemeroptera, Trichoptera, and Odonata. Of these, Hirudinea have previously been observed in some studies [70]. What could be driving such taxa to wetlands and paddies in particular is not certain, but could be attributed to the wetland environment itself, as some taxa have to return to water to breed, making flooded wetlands suitable habitats. Additionally, the changing climate could be impacting the breeding behavior of these taxa in ways that are not currently known. Our study used an unbalanced design where some samples especially from the natural wetland were less than those from other sites. We think that this could have somehow affected these results, though some authors (e.g., Milliken and Johnson [71]) argue that lack of balance does not usually affect results in a single-factor ANOVA.

# 5. Conclusions

This study aimed to evaluate the response of soil macroinvertebrates to paddy rice farming pathways during the 2018–2019 period. A total of 18 macroinvertebrate taxa were observed, dominated by Oligochaeta. Three taxa: Oligochaeta, Gastropoda, and Coleoptera accounted for most of the dissimilarity among sites. Some taxa showed habitat exclusivity: Decapoda, Chilipoda, Diplopoda, and Blattodea were exclusive to the natural wetland, while two taxa (Diplura and Ephemeroptera) were exclusive to the large-scale commercial paddies. No taxa were exclusive to smallholder paddies and none to both smallholder and natural wetland. However, three taxa (Diptera, Odonata, and Trichoptera) were common to both large-scale commercial and smallholder paddies. Considering richness and diversity, large-scale commercial paddies generally had higher richness and diversity than natural wetland and smallholder paddies. Among the soil environmental variables, pH, phosphorus, calcium, clay, and sand seemed to most influence the macroinvertebrate community in the study area. We also observed some taxa that are not usually in soils. These included Hirudinea, Decapoda, Ephemeroptera, Trichoptera, and Odonata. Further studies are needed to extend experiments to other paddy rice farming contexts, monitor macroinvertebrate diversity over a longer term in a balanced design, and explore the potential of using such taxa in monitoring soil quality in rice paddies. The studies would identify indicator macroinvertebrate species that are tolerant or intolerant to chemical and physical soil disturbance in the paddies. It would also be necessary to identify any other apparently new macroinvertebrate taxa in paddies that could be using wetlands during some of their growth stages to respond to the changing global climate. Rice farmers, especially the smallholders, should also mosaic their paddy rice fields with natural areas to create refugia, increasing macroinvertebrate biodiversity.

**Supplementary Materials:** The following are available online at: https://www.mdpi.com/article/ 10.3390/agronomy12020312/s1, Table S1: Percentage composition of soil macroinvertebrate taxa by site and crop stage, Table S2: Proportion of soil samples where macroinvertebrate taxa were encountered per site and crop stage, Table S3: Soil characteristics (Mean  $\pm$  SE) per site and crop stage.

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