

Article



# Soil Macrofauna Abundance and Taxonomic Richness under Long-Term No-Till Conservation Agriculture in a Semi-Arid Environment of South Africa

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Abstract: Soil macrofauna are a critical component for ecosystem function and strongly affect production sustainability. The effect of fertiliser management practices, in different cropping systems, on soil macrofauna communities remains poorly understood in semi-arid to arid regions. The objective of this study was to investigate the effect of tillage practices i.e., no-till (NT), rotational tillage (RT) and conventional tillage (CT) and nitrogen fertiliser application rates (0, 100 and 200 kg/ha N) (urea) on abundance and order diversity of soil macrofauna in a maize continuous monocropping system. The sampling of macrofauna in the trial was conducted in April 2019, August 2019 and March 2020 using  $25 \text{ cm} \times 25 \text{ cm} \times 25 \text{ cm}$  steel monoliths in randomly selected positions, and this was replicated four times. The study found a significant (p < 0.05) increase in the abundance of soil macrofauna in NT (127 ind/m<sup>2</sup>) and RT (110 ind/m<sup>2</sup>) compared to CT (51 ind/m<sup>2</sup>) treatment. The abundance of macrofauna was also negatively affected by increasing the rate of fertiliser, with 0, 100 and 200 kg/ha resulting in 133, 94 and 62 ind/m<sup>2</sup>. Orders Haplotaxida and Diplopoda were found to be sensitive to increases in the fertiliser application rate. Haplotaxida (48.4%) was the most abundant order, followed by Coleoptera (18.2%). Other orders included Diplopoda (9.2%), Gastropoda (6.3%), Isoptera (4.7%), Chilopoda (4.7%), Araneae (4%), Hymenoptera (3.2%), Orthoptera (0.9%) and Dermaptera (0.3%). No-till (NT) and rotational tillage (RT) with mulch favoured the establishment of various macrofauna communities in the studied cropping system.

Keywords: conventional tillage; arthropods; termite; Shannon-Wiener index; earthworms

# 1. Introduction

Soil organisms are amongst the different factors that influence soil formation. The effects of these organisms in soil determine various quality characteristics such as soil structural formation. These organisms are highly diverse and can be classified into different groups based on size, i.e., macrofauna, mesofauna and microfauna, or based on whether they live in water-filled or air-filled pore spaces of the soil and litter [1]. Soil macrofauna consist of a large number of different organisms that have an average body width that is greater than 2 mm. On the other hand, soil mesofauna have an average body width of 100  $\mu$ m to 2 mm, while soil microfauna consists of organisms with an average body width



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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/). of less than 100  $\mu$ m. Soil macrofauna include organisms such as earthworms, millipedes, centipedes, ants, Coleoptera (adults and larvae), Isopoda, spiders, slugs, snails, termites, arthropods, etc. Earthworms, termites and ants form the most important macrofauna component of soils because they are the most abundant in the soil [2] and are considered ecosystem engineers because of their key function in re-working the soil [3–5]. These organisms exert physical, chemical and biological effects on the key quality parameters of the soil [6].

Macrofauna play important key roles in the soil, particularly in soil structural formation [2]. They are responsible for breaking down soil organic matter and also change organically bound nutrients into inorganic forms, making nutrients available for plant uptake [2,7,8]. Organisms such as earthworms [9] and termites [10], the "ecosystem engineers", helps in moving, mixing and aerating the soil through burrowing. Burrowing increases soil porosity, water-holding capacity and the infiltration of water into the soil [2,11,12]. In addition, it also reduces soil compaction, breaks the hard plough pan and promotes effective root penetration [13,14].

On the other hand, conventional tillage practices have been reported to cause physical and habitat disruptions in the soil, and this negatively affects the availability of soil macrofauna [15,16]. It creates stressful conditions for macrofauna settlement since the removal of residues from the soil exposes the soil macrofauna to extreme variation in temperature and humidity and also reduces soil organic matter as their food source [1]. This has, therefore, led to a renewed effort to adopt sustainable agricultural practices that have a minimal effect on soil disturbance, such as conservation agriculture (CA) [2]. With CA, there is little disturbance to the soil, and this helps to protect soil organisms from abrasion [4]. Conservation agriculture is based on four concepts, i.e., no-till and/minimum soil disturbance, permanent soil cover, diversified crop rotation and integrated nutrient management [17]. This, therefore, helps to protect the soil against degradation, provide macrofauna with unlimited food sources [8,18] and increase soil organic matter (SOM) and the recycling of nutrients [2,7,8], especially when leguminous crops that fix nitrogen are used in the rotation [4,19,20].

Furthermore, macrofauna in the soil can also be decreased by the use of herbicides and pesticides and the type of inorganic fertiliser applied. According to [21], the effect of nitrogen fertilisers on soil macrofauna does not only depend on the rate of application but also on the type of fertiliser used and soil pH. The authors found that the abundance of earthworms was significantly affected by increasing amounts of applied nitrogen fertiliser. Furthermore, the earthworm population was reported to decrease at higher pH levels. This study concluded that the application of nitrogenous fertilisers such as ammonium sulphate and sulphur coated urea for long periods have deleterious effects on earthworms in the absence of liming. Similar findings were made by [22], where the population and the diversity of soil fauna increased at low levels in comparison with high levels of nitrogen fertiliser. The authors also reported that there was a decrease in diversity in response to fertilisation on fields that are about 9-20 years older. However, these observations were based on studies that were conducted in areas with high rainfall. Little has been reported on the effects of fertiliser application rates on other ecosystem engineers, particularly those found in semi-arid to arid tropical and sub-tropical regions characterized by lower rainfall. This information is important to make informed decisions on the management and sustainability of agroecosystems.

Thus the objective of the study was to evaluate the long-term effects of different tillage practices and nitrogen fertiliser application rates on the abundance and order diversity of soil macrofauna. This study is part of a long-term experiment that is being conducted in Winterton, Bergville, South Africa, where it forms a large part of the maize growing area in KwaZulu-Natal Province. Several reports have been published on this trial [5,12,23], and one included a study on macrofauna [5]. However, this study was based on lime ammonium nitrate (LAN) as the source on N fertiliser. Treatments with urea, therefore, were not taken into consideration. The current study is a comprehensive study assessing

the effects of different tillage treatments and different fertiliser application rates (urea) on soil macrofauna abundance and diversity in different sampling times.

#### 2. Materials and Methods

# 2.1. Study Area

The study was conducted in a long-term field trial based in Grouton farm ( $28^{\circ}55'26.83''$  S,  $29^{\circ}33'38.64''$  E), which is situated in Winterton, KwaZulu-Natal Province, South Africa (Figure 1). The area is about 35 km south of Bergville. This area forms the larger part of dryland maize commercial production in KwaZulu-Natal and has been managed under no-till since 1990. The experiment was established in the 2003–2004 growing season to investigate the long-term tillage effects on soil fertility and diseases. The annual rainfall, which falls primarily between October and March, is about 643 mm. The mean annual air temperature ranges from about 19.3 °C in June, the coolest month, to 27.9 °C in January, the hottest month. Table 1 represent the mean temperature and total rainfall received during the sampling time. The trial was established on clay loamy textured soil or Ferralsols Haplic soil according to the [24] classification and is planted with dryland maize in summer and left fallow in winter. The soil analysis of the 0–30 cm depth of the trial found that the SOC was 4.1, 3.9 and 1.8% in NT (no-till), RT (rotational tillage) and CT (conventional tillage), respectively. The soil pH (KCl) was 5.86 in NT and RT and 6.10 in CT, while the bulk density ranged from 1.35 g/cm<sup>3</sup> in tilled plots to 1.44 g/cm<sup>3</sup> in untilled plots.



**Figure 1.** The location of the study area  $(28^{\circ}55'26.83'' \text{ S}, 29^{\circ}33'38.64'' \text{ E}, 1038 \text{ m above sea level}), where ($ **a**) & (**b**) shows the aerial view at different scales.

Table 1. Average temperature and total rainfall received between April	l 2019 and l	March 2020
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Month	April	May	June	July	August	September	October	November	December	January	February	March
	2019	2019	2019	2019	2019	2019	2019	2019	2019	2020	2020	2020
Temp. (°C)	17.9	16.3	12.1	12.85	16.3	17.55	20.3	22.3	20.4	21.4	22.75	20.2
Rainfall (mm)	124	24.2	0	0	0.4	2.6	17.4	71.6	83	88.2	117.8	128

#### 2.2. Experimental Design

The experiment was a split plot design with randomized tillage strips (no-till (NT), rotational tillage (RT), and conventional tillage (CT)) forming the whole plot with three replicates and nitrogen fertiliser sources (urea or LAN) and rates of application (0, 50, 100, 150, 200 kg/ha) forming the sub-plots, which were randomized within the whole plot (Table 2). In this study however, sampling was performed only in 0, 100 and 200 kg/ha,

in urea as the source of fertiliser. The sub-plots had  $12 \times 9.5 (144 \text{ m}^2)$  rows of maize and the whole area of the trial was 0.86 ha. Under NT, there were no disturbances in the soil; it involved direct seeding using a planter, and about 10–12 t of maize residues were left on the surface as permanent soil cover. Rotational tillage (RT) was managed under no-till for 4 years and then conventionally tilled on the 5th year, while CT was ploughed using a mouldboard plough to a depth of 30 cm each year and disking to a depth of 10 cm. The nitrogen source was top dressed 4 weeks after planting, while phosphorus and potassium were applied at planting in the band at a rate of 20 and 50 kg/ha, respectively. Lime was applied at a rate of 2 t/ha on the soil surface in NT plots and incorporated during ploughing in CT plots every second season. A combination of S-metolachlor, atrazine, 2,4-D and mesotrione chemicals was used to control weeds using a tractor-drawn ring equipped with an 18 m wide boom sprayer. Leaf fungal diseases were controlled using carbendazim plus flusilazole and azoxystobin.

**Table 2.** Field experimental layout. Coloured blocks represent sampled treatments. NT = no-till, CT = conventional tillage, RT = rotational tillage and LAN = lime ammonium nitrate.

	Rep 1			Rep 2		Rep 3			
NT	СТ	RT	СТ	NT	RT	RT	NT	СТ	
50 Urea	50 Urea	200 LAN	0 N *	200 LAN	150 Urea	150 Urea	200 Urea *	50 LAN	
100 Urea *	150 Urea	50 Urea	150 Urea	200 Urea *	0 N *	150 LAN	200 LAN	50 Urea	
150 LAN	0 N *	200 Urea *	100 Urea *	50 LAN	100 LAN	200 LAN	50 LAN	200 LAN	
50 LAN	100 Urea *	50 LAN	50 LAN	0 N *	200 LAN	50 LAN	50 Urea	200 Urea *	
200 Urea *	100 LAN	100 LAN	200 LAN	150 LAN	100 Urea *	100 LAN	100 Urea *	100 LAN	
150 Urea	200 LAN	150 Urea	50 Urea	100 Urea *	150 LAN	50 Urea	0 N *	150 LAN	
100 LAN	200 Urea *	150 LAN	200 Urea *	150 Urea	50 Urea	100 Urea *	150 LAN	0 N *	
200 LAN	50 LAN	100 Urea *	100 LAN	100 LAN	200 Urea *	200 Urea *	150 Urea	100 Urea *	
0 N *	150 AN	0 N *	150 LAN	50 Urea	50 LAN	0 N *	100 LAN	150 Urea	

\* Indicates the sub-plots that were sampled.

#### 2.3. Macrofauna Sampling

Macrofauna sampling was conducted in April 2019, August 2019 and March 2020 according to the method previously described by [25]. One soil monolith of 25 cm  $\times$  25 cm  $\times$  25 cm depth was excavated randomly in each replicate (n = 4) and hand-sorted to collect macrofauna. The same sampling method was employed in April 2019, August 2019 and March 2020. Thereafter, all macrofauna collected were preserved in glass bottles containing 70% alcohol for laboratory identification. Macrofauna included all organisms visible to the naked eye (2–80 mm) that spend most of their important lifecycle in the soil or on the soil surface. All of the organisms collected were identified at the order or family level and classified according to their typical ecological behaviours.

### 2.4. Data Analysis

The total abundance, Shannon–Wiener index and Pielou's evenness index were calculated at each sampling, so as to analyse the effects of treatments on the soil macrofauna communities. The total abundance was computed as the total number of macrofauna per plot converted to individual/m<sup>2</sup>. The Shannon–Wiener index (diversity) [26] was computed using the following formula:

$$H' = -\sum (pi \ln pi) \tag{1}$$

where:

pi = proportion of individuals found in species *i*. For a well-sampled community, the proportion can be estimated as:

$$pi = ni/N$$
 (2)

where:

ni = number of individuals in species *i* 

N = total number of individuals in the community.

By definition, *pi* will always be between zero and one; the natural log makes all of the terms of the summation negative, which is why the inverse of the sum is taken.

The Shannon–Wiener index takes into account the number of orders encountered, with i = 1 to s, where pi = the probability of meeting a taxon i on a plot and s = total number of taxa encountered on the plot [27]. H = 0 when there is only 1 taxon and is at maximum when all taxa are of equal abundance [27].

Repeated ANOVA measurements using GenStat 17th Edition (VSN International, Hemel Hempstead, UK) were performed to evaluate the effects of treatments (NT, RT and CT), the sampling time (April 2019, August 2019 and March 2020), and their interaction on the indices on soil macrofauna community and abundance of taxonomic orders. To satisfy the normality of variance assumption, data were log (x + 1) transformed prior to analysis. Means were separated using Tukey's least significant difference (LSD) at 5% level of significance.

#### 3. Results

## 3.1. Soil Macrofauna Abundance

Significant differences (p < 0.05) were found in different tillage treatments, with NT having significantly more organisms than CT (Table 3 and Figure 2). However, significant differences were not found between NT and RT (Figure 2), although NT treatment was observed to have a higher number of organisms than RT. Highly significant (Table 3) differences were found amongst the different sampling times on soil macrofauna, with August 2019, which corresponded to winter, having the lowest soil macrofauna compared to March 2020 and April 2019, respectively, which corresponded to summer and the rainy season (Figure 3). Significant differences (p < 0.05) were also observed in soil macrofauna under different nitrogen application rates, with 0 kg/ha having the highest abundance of soil macrofauna compared to 100 kg/ha and 200 kg/ha, respectively (Figure 4).



**Figure 2.** The variation in abundance of soil macrofauna in three treatments of no-till (NT), rotational tillage (RT) and conventional tillage (CT) recorded after 17 years of trial establishment. T = treatment.



**Figure 3.** The variation in abundance of soil macrofauna across the three sampling times recorded after 17 years of trial establishment. S = sampling time.

The interaction between treatments (tillage practices), sampling time, and nitrogen levels on soil macrofauna was found to be insignificant (p > 0.05) (Table 3). Soil macrofauna in NT in April 2019 were significantly higher (p < 0.05) than in CT, whereas there were no differences (p > 0.05) between NT and RT (Table 4). The macrofauna abundance was 17% and 28% higher in NT than in RT and CT, respectively. In August 2019, there were no significant differences (p > 0.05) found between tillage treatments, although abundance in NT was 59% and 58% higher than in RT and CT treatments, respectively. In March 2020, RT had significantly (p < 0.05) higher numbers of soil macrofauna compared to CT, whereas when compared to NT treatment, there was no difference (p > 0.05). At this sampling time, RT had 18% more organisms than NT and 14% more than in CT treatment.

**Table 3.** The results of repeated ANOVA of the effect of tillage treatments (NT, RT and CT), sampling time (April 2019, August 2019 and March 2020), N levels and their interaction on the abundance of macrofauna.

Effects	df	F	р
Treatments	2	4.11	0.022 *
S. time	2	13.9	< 0.001 **
N-level	2	3.19	0.049 *
Treatment $\times$ S. time	4	1.84	0.136
N-level $\times$ treatment	4	0.49	0.742
S. time $\times$ N-level	4	1.13	0.351
S. $\times$ N level $\times$ treatment	8	0.83	0.577

t = treatment. N = nitrogen fertiliser application rate and S = sampling time. The symbols \*, \*\*, denote statistical significance at 0.05 and 0.01, respectively.

Significant differences (p < 0.05) were also found in NT at different sampling times, with August 2019 having significantly lower numbers of macrofauna than in April 2019, whereas April 2019 and March 2020 were not significantly different (p > 0.05). The RT treatment followed the same trend with lower numbers of soil macrofauna in August 2019. However, in this treatment, at the April 2019 and March 2020 sampling times, the numbers of macrofauna were similar (p > 0.05). In the CT treatment, on the other hand, there were no significant differences (p > 0.05) in soil macrofauna at the different sampling times, although their numbers were observed to be higher in April 2019 and March 2020 compared to the winter season in August 2019. No significant differences (p > 0.05) were

found in the tillage  $\times$  N fertiliser application rate. The general trend in most cases was that soil macrofauna decreased with increased nitrogen level.

**Table 4.** Abundance of soil macrofauna under different tillage treatments (NT = no-till, RT = rotational tillage and CT = conventional tillage), sampling times (April 2019, August 2019 and March 2020) and nitrogen fertiliser application rates (0, 100 and 200 kg/ha).

<b>T</b> ( )	N Level		Sampling Time						
Ireatment	(kg/ha)	April 2019	August 2019	March 2020	Mean				
	0	325 <sup>a,b</sup>	11.0 <sup>a</sup>	114 <sup>a,b</sup>	150 <sup>a</sup>				
NT	100	203 <sup>a,b</sup>	71.0 <sup>a,b</sup>	162 <sup>a,b</sup>	145 <sup>a</sup>				
	200	165 <sup>a,b</sup>	21.0 <sup>a</sup>	71.0 <sup>a,b</sup>	85.7 <sup>a</sup>				
	Mean	231.0 <sup>b</sup>	34.3 <sup>a</sup>	116 <sup>a,b</sup>	127 <sup>a</sup>				
	0	210 <sup>a,b</sup>	5.00 <sup>a</sup>	348 <sup>b</sup>	188 <sup>a</sup>				
RT	100	107 <sup>a,b</sup>	4.00 <sup>a</sup>	124 <sup>a,b</sup>	78.3 <sup>a</sup>				
	200	121 <sup>a,b</sup>	9.00 <sup>a</sup>	59.0 <sup>a,b</sup>	63.0 <sup>a</sup>				
	Mean	146 <sup>a,b</sup>	6 <sup>a</sup>	177 <sup>b</sup>	110 <sup>a</sup>				
	0	130 <sup>a,b</sup>	7.00 <sup>a</sup>	37.0 <sup>a,b</sup>	58.0 <sup>a</sup>				
СТ	100	98.0 <sup>a,b</sup>	7.00 <sup>a</sup>	69.0 <sup>a,b</sup>	58.0 <sup>a</sup>				
	200	71.0 <sup>a,b</sup>	7.00 <sup>a</sup>	34.0 <sup>a,b</sup>	37.3 <sup>a</sup>				
	Mean	99.7 <sup>a</sup>	7.0 <sup>a</sup>	46.7 <sup>a</sup>	51.1 <sup>a</sup>				
I	$SD_{T} = 10$	57							

$$CV_T \vee N \vee c = 106$$

 $CVT \times N \times S = 100$ 

NT = no-till, RT = rotational tillage and <math>CT = conventional tillage. T = treatments, N = nitrogen fertiliser application rate, and S = sampling time. Numbers in the table not sharing the same letter differ significantly at LSD (p = 0.05).



**Figure 4.** Variation in the abundance of soil macrofauna in three treatments of N fertiliser at different application rates (0, 100 and 200 kg/ha) recorded after 17 years of trial establishment. N = nitrogen fertiliser application rate.

## 3.2. Taxonomic Groups

A total of 1457 individuals were recorded, belonging to 10 orders and 3 phyla (Table 5). Eight orders (Araneae, Chilopoda, Coleoptera, Dermaptera, Diplopoda, Hymenoptera, Isoptera and Orthoptera) belonged to the *Arthropoda* phylum. Single orders under Mollusca (Gastropoda) and Annelida (Haplotaxida) were also reported. The most dominant orders were Haplotaxida (48.4%), followed by Coleoptera (18.2%). Other orders included

Diplopoda (9.27%), Gastropoda (6.31%), Isoptera (4.67%), Chilopoda (4.74%), Araneae (3.98%), Hymenoptera (3.23%), Orthoptera (0.89%) and Dermaptera (0.34%) (Figure 5).

**Table 5.** The list of taxa, mean abundance (ind.m<sup>-2</sup>) of soil macrofauna collected from different tillage treatments and at different sampling times.

Таха					April 2019 August			ugust 201	2019 March 2020			
Phylum	Class	Order	Family	NT	RT	СТ	NT	RT	СТ	NT	RT	СТ
Arthropoda	Arachnida	Araneae	Salticidae	10.7 <sup>a</sup>	2.37 <sup>a</sup>	2.37 <sup>a</sup>	1.78 <sup>a</sup>	1.19 <sup>a</sup>	0 <sup>a</sup>	9.48 <sup>a</sup>	4.74 <sup>a</sup>	1.19 <sup>a</sup>
1	Myriapoda	Chilopoda		11.3 <sup>a</sup>	5.93 <sup>a</sup>	0 a	0 a	0 a	0 a	1.78 <sup>a</sup>	8.89 a	5.33 <sup>a</sup>
		Diplopoda		32.6 <sup>a,b</sup>	21.9 <sup>a,b</sup>	1.78 <sup>a</sup>	2.37 <sup>a</sup>	0.59 <sup>a</sup>	0 <sup>a</sup>	15.4 <sup>a,b</sup>	4.74 <sup>a</sup>	1.19 <sup>a</sup>
	Insecta	Coleoptera		10.7 <sup>a</sup>	14.2 <sup>a,b</sup>	20.1 <sup>a,b</sup>	5.93 <sup>a</sup>	1.19 <sup>a</sup>	6.52 <sup>a</sup>	42.7 <sup>a,b</sup>	40.1 <sup>a,b</sup>	24.9 <sup>a,b</sup>
		Dermaptera		0.59 <sup>a</sup>	0 a	0 a	0.59 <sup>a</sup>	0 a	0.59 <sup>a</sup>	0.59 <sup>a</sup>	0.59 <sup>a</sup>	0 a
		Hymenoptera		10.1 <sup>a</sup>	7.11 <sup>a</sup>	6.52 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	3.56 <sup>a</sup>	5.33 <sup>a</sup>
		Isoptera	Termitidae	1.19 <sup>a</sup>	2.37 <sup>a</sup>	4.15 <sup>a</sup>	21.9 <sup>ab</sup>	1.19 <sup>a</sup>	0 <sup>a</sup>	0.59 <sup>a</sup>	2.37 <sup>a</sup>	6.52 <sup>a</sup>
		Orthoptera	Gryllidae	2.37 <sup>a</sup>	0.59 <sup>a</sup>	2.37 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	2.96 <sup>a</sup>	0 <sup>a</sup>	1.19 <sup>a</sup>
Mollusca		Gastropoda	2	16.6 <sup>a,b</sup>	17.2 <sup>a,b</sup>	20.7 <sup>a,b</sup>	0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>
Annelida	Clitellata	Haplotaxida		135 <sup>c</sup>	74.7 <sup>b,c</sup>	40.3 <sup>a,b</sup>	1.78 <sup>a</sup>	1.78 <sup>a</sup>	0 <sup>a</sup>	42.1 <sup>a,b</sup>	111 <sup>c</sup>	0.59 <sup>a</sup>
$LSD_{T \times S} = 28.8$												

 $CV_{T \times S} = 325$ 

T = treatments and S = sampling time. Numbers in the table not sharing the same letter differ significantly at LSD (p = 0.05).



**Figure 5.** Variation in abundance of the different taxonomic groups observed after 17 years of trial establishment. O = orders.

Significant differences (p < 0.05) were observed in the interaction between nitrogen fertiliser application rates and macrofauna orders (Table 6 and Figure 6). Most notably, the orders Haplotaxida and Diplopoda were more sensitive to the increase in the fertiliser application rate from 100 kg/ha to 200 kg/ha (Figure 6). All other orders were found to be less sensitive (p > 0.05) to the increase in the application rate of fertiliser. This may be also due, perhaps, to a lack of material to show the presence of an effect. Furthermore, the results showed clear differences in the number or orders between the winter (August 2019) and the other sampling periods (April 2019 and March 2020) (Figure 7) during the summer rainy season. Fewer orders and numbers of individuals (macrofauna) were recorded during winter (August 2019) compared to those in the April 2019 and March 2020 datasets. Order Haplotaxida, which represented a significantly higher number of individuals, was found to me more sensitive to lower temperature and rainfall observed in winter months, while order Isoptera was less sensitive to these conditions.

Effects	df	F	p
Treatments	2	4.46	0.012 *
Order	9	15.76	<0.001 ***
N level $\times$ Order	18	2.37	0.001 ***
S. time $\times$ Order	18	5.21	<0.001 ***
$Treatment \times S \ time \times Order$	36	1.58	0.019 *

**Table 6.** The results of repeated ANOVA of the effect of treatments (NT, RT and CT), sampling time (April 2019, August 2019 and March 2020), N-levels and their interaction on macrofauna diversity.

t = treatment. N = nitrogen fertiliser application rate and S = sampling time. The symbols \*, \*\*\* denote statistical significance at 0.05 and 0.001, respectively.

Most of the following macrofauna orders were found in the litter: Araneae (e.g., spiders), Gastropoda (mainly snails and slugs), Dermaptera (earwigs), Chilopoda (centipedes), Hymenoptera (e.g., ants), Orthoptera (e.g., crickets) and Diplopoda (e.g., millipedes). Coleoptera (e.g., grubs and meal worms), Haplotaxida (e.g., earthworms) and Isoptera (e.g., termites) were mainly found under the soil surface.



**Figure 6.** Variation in abundance of the different taxonomic groups in three treatments of N fertiliser application rate (0, 100 and 200 kg/ha) after 17 years of trial establishment. O = orders and N = nitrogen fertiliser application rate.



**Figure 7.** Variation in abundance of the different taxonomic groups across the three sampling times (April 2019, August 2019 and March 2020) after 17 years of trial establishment. O = orders and S = sampling time.

The repeated measures ANOVA results (Table 7) indicated that all indices of soil macrofauna communities responded significantly to treatments (p < 0.001), N fertiliser application rates (p < 0.001), time of sampling and their interactions (p < 0.001). In CT treatment, there were no significant (p > 0.05) differences found in macrofauna diversity (H') (Figure 8) and evenness (E) (Figure 9) across the three sampling times, and soil macrofauna community or diversity was not affected by the N fertiliser application rate. Contrarily, in the NT and RT treatments, H' and E of macrofauna were highly affected by sampling time (Figures 8 and 9). Nitrogen fertiliser in these treatments, i.e., 100 kg/ha, significantly reduced the presence of certain species. However, the types of macrofauna communities found in April 2019 and March 2020, which correspond to the summer and rainy season, respectively, were similar (p > 0.05) compared to the August 2019 sampling period, which corresponds to winter with no rain.



**Figure 8.** Variation in the Shannon–Weiner index of soil macrofauna communities relative to tillage treatments, N fertiliser application rates, and sampling times. April-19 = April 2019, Aug-19 = August 2019 and Mar-20 = March 2020. T = treatments, N = nitrogen fertiliser application rate and S = sampling time.



**Figure 9.** Variation in the Evenness index of soil macrofauna communities in different tillage treatments, N fertiliser application rates, and sampling times. April-19 = April 2019, Aug-19 = August 2019 and Mar-20 = March 2020. T = treatments, N = nitrogen fertiliser application rate and S = sampling time.

	Sha	annon–W	einer (H')	Evenness Index (E)			
Effects	df	F	р	df	F	р	
Treatments	2	18.17	< 0.001 ***	2	18.2	< 0.001 ***	
S. Time	2	40.6	< 0.001 ***	2	40.6	< 0.001 ***	
N level	2	16	< 0.001 ***	2	16.04	< 0.001 ***	
Treatment $\times$ S time	4	14.4	< 0.001 ***	4	14.4	< 0.001 ***	
N level $\times$ treatment	4	6.78	< 0.001 ***	4	6.78	< 0.001 ***	
Treatments $\times$ S time $\times$ N level	8	6.31	< 0.001 ***	8	6.31	< 0.001 ***	

**Table 7.** The results of repeated ANOVA of the effect of tillage treatments, N fertiliser application rate, and sampling time on Shannon–Weiner and Evenness indices.

t = treatment. N = nitrogen fertiliser application rate and S = sampling time. The symbols \*\*\* denote statistical significance at 0.001.

# 4. Discussion

The objective of this study was to evaluate the long-term effects of different tillage practices and nitrogen fertiliser application rates on the abundance and order diversity of soil macrofauna. The results of the study found that the abundance and order diversity of soil macrofauna were enhanced under no-till treatment compared to the rotational and conventional tillage treatments. The findings of this study were consistent with those of [5,28–33] which provided evidence of a positive contribution from no-till with residue mulch on soil macrofauna abundance. Crop residue mulch serves as a source of food and energy for the macro and microorganisms [1,32,34] in soil, and leaving maize residues as permanent soil cover under no-till and rotational tillage helped in improving habitat for soil macrofauna with important ecological functions in soil such as Haplotaxida (earthworms), Araneae (spiders), Chilopoda (centipedes), Coleoptera (beetles), Isoptera (woodlice), Hymenoptera, Orthoptera (grasshoppers), Diplopoda (millipedes) and Dermaptera (earwigs). Arthropods are responsible for breaking down added residues; thus, they are highly favoured by mulching [28], and hence, were found to be the most dominant phylum under no-till and rotational tillage this study. On the other hand, conventional tillage had a detrimental effect on macrofauna by physical breaking down their soil habitats and increasing the oxidation of physically protected soil organic matter by exposing it to microbial attack.

The highest numbers of earthworms (Haplotaxida) were recorded under no-till and rotational tillage with permanent soil cover mulch (Figure 5). On the other hand, almost no earthworms were found under conventional tillage, especially during the winter season (Table 6). Similar findings were observed by [1,35-37]. Earthworms are susceptible to very high and very low temperatures [38]. The mean temperatures in June, July and August 2019 were 12.1, 12.8 and 16.3 °C, respectively, and the total rainfall during these months was 0, 0 and 0.4 mm, respectively. This could have reduced the number of earthworms in these months, whereas the respective 17.9 and 20.2 °C temperatures and rainfall amounts of 124 and 128 that were observed in April 2019 and March 2020 could have favoured increases in the numbers of earthworms during the summer rainy season. Mulch prolongs the active periods for earthworms by slowing down the rate at which the soil dries during extremely hot conditions, thus allowing them to feed and reproduce much more [1]. Hence, the highest numbers of earthworms were recorded during the April 2019 and March 2020 sampling periods compared to winter, August 2019. Lower temperatures in winter with no rainfall, combined with soil disturbance every year in conventional tillage and lack of food sources due to the incorporation of residues into the soil, had a negative impact on the population of earthworms [1,39]. Earthworms help in increasing water infiltration and drainage through burrowing while forming stable aggregates that reduce erosion and nutrient losses [40], at the same time increasing root growth [1,39]. They are also responsible for mixing or incorporating residues into the soil especially under no-till practices where there is no mechanical mixing by farm implements [1,39]. In addition, earthworms and millipedes were observed to be sensitive to increases in the fertiliser application rate (Figure 6). These results agree with those of [21,22], found a decrease in

earthworm populations and other macrofauna groups with an increase in the fertiliser application rate.

The second most abundant group of macrofauna was Coleoptera (beetles, grubs and meal worms) (18.2%) (Figure 5). These soil macrofauna were highly abundant during the April 2019 and March 2020 sampling periods. The lowest numbers were recorded in winter (August 2019), and there were no significant differences in different tillage treatments in all three sampling periods (Table 6). These findings are similar to those of [5], where this group was similarly abundant under no-till rotational tillage and conventional tillage treatments with lime ammonium nitrate (LAN) as the fertiliser source. Taxa in the Coleoptera order are highly mobile, and they have the potential to reproduce and multiply in large numbers, making their populations less sensitive to changes in tillage practices [5,41]. Moreover, the experiment showed that millipedes (Diplopoda) were the third largest order of macrofauna (9.27%), mostly found under no-till and rotational rather than conventional tillage practices (Figure 5). This experiment yielded results similar to those reported by other authors [5,27,36]. No-till and rotational tillage treatments yielded a significantly higher population during the April 2019 and March 2020 sampling periods. Little to no millipedes were recorded in winter. Millipedes play a good role in decomposing vegetation (plant debris) and the cycling of nutrients and carbon, hence their preference to live under mulch. It has been reported that these organisms can become a pest to cultivated crops such as maize, sweet potatoes, carrots, etc. [42] if their food source (mulch or plant litter) is depleted [5,27,43]. Therefore, it is important that that such organisms be provided with enough food supply at all times [5].

The highest numbers of termites (Isoptera) were recorded in winter (Figure 5). Termites were the most abundant macrofauna of all in winter, mostly found under no-till treatment. Termites are important for breaking down dry organic materials; thus, they were recorded in winter where maize straw (mulch) left on the surface (NT treatment) was dry. Under a dryland cropping system, adding more organic residues can be used as a strategy to grow the population of termites [5]. Termites are important in soil structural formation; they are responsible for the formation of both macro and microaggregates, recycling of nutrients and improved porosity, especially in regions where earthworms are not common [40]. Winter adversely affected earthworms but favoured the abundance of termites (Figure 5). These findings were similar to those of [38] where the removal of residues decreased the numbers of termites by 90%. It has been reported that the use of machinery under conventional tillage causes the destruction of termite nests and burrows, which then reduces the size of termite populations [1].

Other macrofauna orders such as Araneae (spiders), Chilopoda (centipedes), Dermaptera (earwigs) and Hymenoptera (ants) were available but in small population numbers. These soil organisms feed on other arthropods and sustain the predator–prey relationship. In this manner, the ecosystem is balanced, and pests are controlled biologically, which helps in eliminating the chemical approach to controlling pests since it is expensive and pests tend to become resistant to continuous chemical applications [5,44].

### 5. Conclusions

This study found that no-tillage and rotational tillage with permanent soil cover favours the increased abundance of soil macrofauna as compared to conventional tillage treatment. This finding in turn has important positive implications for soil structural formation and the accumulation of soil organic carbon, a key indicator of soil quality. The sampling times and fertiliser application rates were also found to affect the abundance of soil macrofauna. Increasing the application of urea as an inorganic fertiliser was observed to decrease the abundance of soil macrofauna in general, and in this group, Haplotaxida and Diplopoda were the most sensitive. Macrofauna abundance was also affected during the sampling period that corresponded to winter, where there was no rainfall for 3 months, and during the summer rainy season, which was accompanied by high temperatures. The study found that macrofauna could vary greatly depending on the sampling time, with winter months reducing the availability of these organisms compared to the summer months when there is rainfall. Macrofauna taxa were also influenced by the type of tillage, with Coleoptera less sensitive to tillage practices and Haplotaxida more sensitive to tillage and temperature.

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