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Earthworm Population Response to Simplified Tillage and Shortened Crop Rotations in a Central Lithuanian Cambisol: A Five-Year Study

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Abstract: This five-year study examined the impact of simplified tillage practices and shortened crop rotations on soil physical attributes and earthworm populations as an important indicator of soil health in Central Lithuanian Cambisols. The experiment was set up following a split-plot design to compare conventional tillage and no-tillage systems across three rotation schemes (three-field, two-field, and monoculture). The experiment was carried out over a period of 5 years, from 2010 to 2014. Preliminary soil conditions revealed notable disparities in moisture content across tillage methods (20.0 ± 0.3% against 17.9 ± 0.3% at a depth of 5–10 cm; $p < 0.001$), although variations in bulk density were more evident in the deeper soil layer (1.42 ± 0.02 versus 1.47 ± 0.01 mg m^{-3} at 15–20 cm). Earthworm abundance exhibited a strong negative association with bulk density ($r = -0.612$, $p = 0.041$) and a positive correlation with total porosity ($r = 0.583$, $p = 0.044$) in the upper soil layer. Notably, this study revealed the unexpected resilience of earthworm populations to tillage practices, with no significant differences between conventional and no-till systems ($F_{1,108} = 1.414$, $p = 0.237$). Rotation effects showed more significance than tillage intensity, as both two-field and three-field rotations sustained comparable earthworm populations (127.5–131.2 ind. m^{-2}, 32.8–35.4 g m^{-2}), but monoculture exhibited markedly lower figures (105.0 ± 13.2 ind. m^{-2}, 25.6 ± 2.7 g m^{-2}; $p < 0.048$). Three-way ANOVA indicated substantial temporal effects ($F_{4,108} = 17.227$, $p < 0.001$), demonstrating that environmental influences gained prominence as systems evolved. These findings challenge traditional assumptions about tillage impacts on soil fauna and indicate that crop diversification within the rotation cycle, rather than tillage intensity or rotation duration, is the essential determinant for sustaining earthworm populations in agricultural systems. Soil structural factors proved to be a significant factor but played a less substantial role.

Keywords: soil fauna; soil structure; soil biodiversity; conservation agriculture; agroecosystem management

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1. Introduction

The increasing demand for agricultural production has resulted in a greater focus on sustainable management practices. These practices can help to minimize energy use and maintain soil fertility and crop yields at the same time [1]. Reduced tillage techniques have been identified as a potentially effective solution to achieve these objectives. These techniques are characterized by decreased tillage frequency and depth, often consolidating many operations into single passes. These techniques may play a significant role in conserving soil moisture and reducing erosion. However, their impact on crop yields may vary [2,3]. This improvement in productivity is often linked to enhanced soil carbon sequestration and improved soil structure [4].

Earthworms are an essential organism for maintaining soil health and fertility in agroecosystems. They act by providing several ecosystem services such as nutrient cycling, soil structure improvement, and organic matter decomposition [5]. Research has quantified these benefits, showing that earthworm activities can increase crop yield and above-ground biomass by up to 25% and 23%, respectively [6,7]. The presence and abundance of earthworms serve as key indicators of soil biodiversity in agricultural systems, as they occupy multiple trophic levels and provide diverse ecosystem services that support both soil health and agricultural productivity [8]. Their influence extends to improving soil microorganism activity and enhancing nutrient bioavailability [9]. Earthworm populations in agricultural settings are significantly influenced by land management practices, particularly tillage systems, crop rotation diversity, and organic matter contributions [10].

The incorporation of modern agricultural gear into European farming systems, particularly in countries like Lithuania, has enabled farmers to abandon traditional plowing techniques. The shift to simplified tillage systems provides several advantages. These advantages include reduced fuel consumption and decreased labor requirements and production costs, at the same time providing environmental benefits. Studies across European agricultural landscapes have shown that these benefits extend beyond production aspects to include enhanced biodiversity and ecosystem services [11]. The successful implementation of these systems in brief crop rotations requires careful assessment of soil agrochemical and agrophysical properties, weed pressure, disease occurrence, and the maintenance of soil biological communities, particularly earthworms as essential indicators of soil health.

Earthworms are typically classified into three distinct ecological categories based on their feeding habits, burrowing behavior, and soil habitat [12]. Epigeic species: These earthworms reside on the surface litter layer, ingesting undecomposed organic matter. They do not create permanent burrows and have a negligible impact on soil formation [13]. Endogeic species: These earthworms reside in and ingest organic matter within the upper mineral soil layers, creating horizontal burrows. They consume significant amounts of soil and its associated organic matter [13]. Anecic species: These are deep-burrowing earthworms that create enduring vertical tunnels. They ingest surface waste, which they convey into their burrows, integrating it with soil [13]. Previous research has shown that the functional diversity of these ecological groups significantly increases under reduced tillage practices, leading to enhanced soil ecosystem services [14].

In European agroecosystems, especially in temperate regions, earthworm ecosystems are typically dominated by endogeic and anecic species. In Lithuanian agricultural soils, commonly identified earthworm species are *Aporrectodea caliginosa* (endogeic species), *Lumbricus terrestris* (anecic species), *Allolobophora chlorotica* (endogeic species), and *Aporrectodea rosea* (endogeic species) [15]. These species may respond quite differently depending on management practices applied.

The relationship between earthworms and agricultural productivity is bidirectional—while earthworms enhance soil fertility and crop growth, agricultural practices significantly influence earthworm communities and their functional diversity [16]. The intensity of tillage is a key factor influencing earthworm populations and community composition. Conventional tillage can lead to a significant reduction in earthworm populations due to deep plowing, which is characteristic for this soil management technique. This reduction occurs through multiple mechanisms: destruction of existing burrow networks, alteration of soil physical conditions including temperature and moisture regimes, and direct mechanical damage to earthworms during tillage operations [17]. Studies indicate decreases of up to 70% in biomass and 80% in abundance in comparison to reduced tillage systems [18]. A recent comprehensive meta-analysis of 165 publications across 40 countries has shown even more dramatic effects, with reduced tillage practices increasing earthworm abundance by 137% and biomass by 196% compared to conventional plowing, with particularly strong responses in soils under reduced tillage for over 10 years [19]. Research in Lithuania indicates that shifting from conventional to conservation tillage practices can substantially increase the density and biomass of earthworm populations [20].

Earthworms contribute substantially to soil porosity and aggregate stability through their burrowing activities and cast production. Research indicates that earthworm channels can enhance water infiltration by up to 150% compared to soils without earthworm activity [21]. Earthworm burrows play a crucial role in soil structure development, particularly in compacted soil layers where their channels serve as essential pathways for root growth [22]. The heterogeneity created by earthworms through burrows and cast production is closely linked to local root adaptations and their tendency to proliferate in nutrient-rich sites [23]. Soil moisture content is a critical factor influencing earthworm population dynamics. Water retained in soil micropores (width 30–0.2 μm) represents the capillary or plant-available soil moisture. The soil's water-holding capacity is directly influenced by its pore structure, which includes pore size, shape, continuity, and tortuosity [24]. Soil texture, organic matter content, and agricultural management practices alter these characteristics by influencing soil aggregate structure [25].

Research in Lithuania indicates that soil moisture content significantly affects earthworm population density, with variations based on soil type and texture [26]. Sandy loam soils typically support more earthworm populations than denser soils, mostly due to enhanced drainage and aeration characteristics. Studies in the United States on winter wheat rotations have demonstrated that the quality and quantity of crop residue, along with soil moisture, affect earthworm population density and species-specific biomass growth [27].

The variety of crop rotation significantly influences earthworm populations. Research demonstrates that earthworm populations decline consistently with the decrease in crop rotation systems—from four-field to three-field and two-field rotations, resulting in average counts of 64.3, 62.1, and 57.7 individuals per m^2, respectively [28]. The choice of the preceding crop significantly affects earthworm population density. The highest earthworm densities were seen after sugar beet (99 individuals m^{-2}), followed by barley and winter wheat (56 and 50 individuals m^{-2}, respectively), with the lowest densities recorded after peas (35 individuals m^{-2}) [29]. Another study conducted in Lithuania demonstrated that after increasing the cereal proportion in crop rotation from 50% to 100%, a decline of 33% in earthworm population density was observed [30].

Conservation agricultural practices, like reduced tillage or retention of crop residue, have shown positive effects on earthworm populations. The positive effects of earthworms in agricultural soils are particularly pronounced in conservation agriculture systems, where they enhance water infiltration, decrease runoff, and prevent erosion through their burrowing activities [5]. Global meta-analysis demonstrates that no-tillage and conservation agriculture can increase earthworm biomass by up to 196% and earthworm abundance by as much as 137% compared to conventional tillage [10].

Climate change poses an increasing challenge to earthworm populations because of changing ecological conditions across Lithuanian agroecosystems. The conservation of soil biodiversity through appropriate agricultural practices is becoming increasingly critical as climate change poses additional stress on agricultural ecosystems [31]. As temperature and precipitation patterns become more variable, significant effects on earthworm activity, reproduction, and survival may be expected. In terms of the climatic conditions of Lithuania and the surrounding region, warming trends can lead to an extended period of earthworm activity; on the other hand, there is an increased risk of drought stress during the summer months [32].

Currently, research is focused on evaluating the feasibility of simplified tillage regimes and shorter crop rotations, at the same time preserving the soil quality and beneficial soil fauna populations. This is especially relevant for cultivated Central Lithuanian soils, where it is crucial to understand the effects of simplified tillage methods and shortened crop cycles on crop yield, soil physical parameters, and biological health, including earthworm populations.

1.1. Hypothesis

Simplified tillage and shortened crop rotations in Central Lithuanian Cambisols will not negatively impact earthworm populations or their biomass compared to conventional tillage and traditional rotation systems.

1.2. Goal

To investigate and validate the feasibility of shortened crop rotation cycles and the possibility of winter wheat re-cropping without detriment to soil properties or negative impacts on earthworm populations, which are critical markers of soil health

1.3. Objectives

1. To evaluate the effects of diverse tillage methods on soil physical properties (bulk density, penetration resistance, porosity, moisture content) across different crop rotations.
2. To examine the impact of tillage methods on soil pedobiological characteristics, namely the biomass and prevalence of earthworm populations.
3. To analyze the relationships between soil physical properties, management practices, and earthworm population dynamics.
4. To evaluate the optimal combinations of tillage techniques and crop rotation sequences that maintain both agricultural productivity and soil health.

2. Materials and Methods

2.1. Study Site and Experimental Design

The research was conducted during 2010–2014 at the Lithuanian Research Centre for Agriculture and Forestry (55°23′38″ N, 23°51′35″ E). The experimental site was established on a Cambisol (Endocalcaric, Endogleyic) with two soil textures: sandy loam and loam. The experiment employed a split-plot design with two main factors: tillage system (Factor A) and crop rotation (Factor B). There were three complete blocks as replications, although only two blocks were used in 2010 due to the loss of data from replicate block 2 caused by adverse environmental conditions that hindered the collection of representative samples (Figure 1).

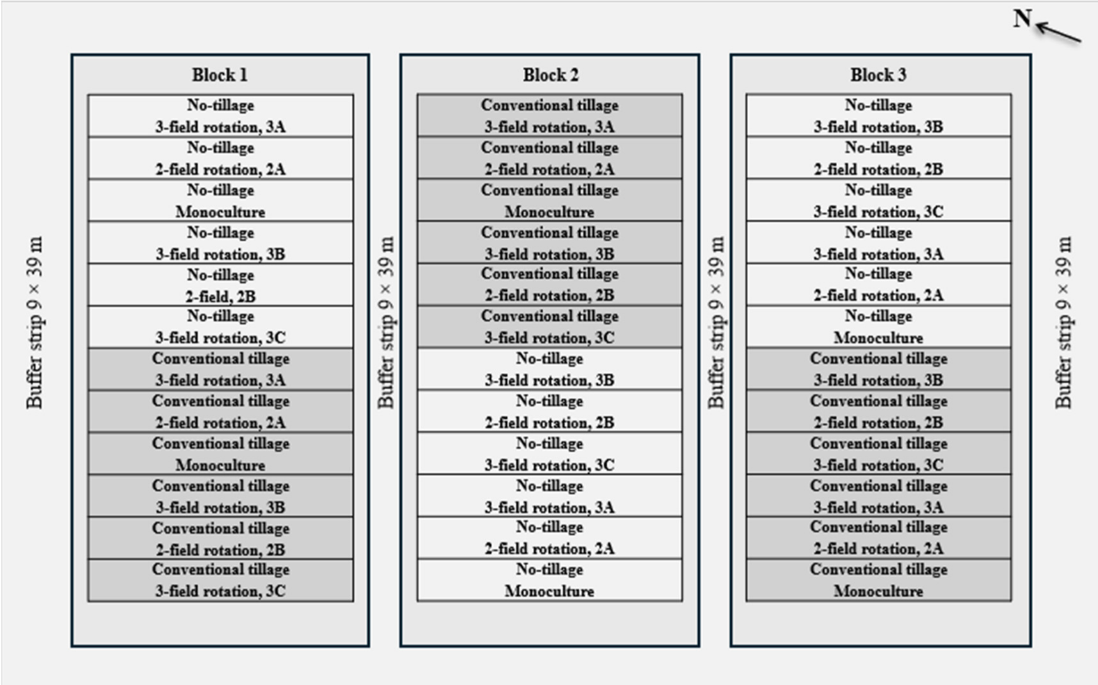

Figure 1. Experimental layout of long-term field study (2010–2014) showing systematic arrangement of tillage treatments (conventional tillage; no tillage) combined with rotation systems (3A, 3B, 3C—three-field rotation variants with different crop sequences; 2A, 2B—two-field rotation variants; M—monoculture) in three complete blocks. Individual plot size was 60 m^2 (3 m × 20 m) with harvest areas of 39.6 m^2 (2.2 m × 18 m) established to eliminate edge effects. Note that block 2 data from 2010 were unavailable due to technical issues.

2.2. Experimental Treatments

Tillage systems (Factor A) consisted of two contrasting approaches, with operational protocols detailed in Table 1. All field operations were conducted at standard equipment speed of 8–10 km/h, with timing determined by soil conditions. Spring operations were initiated when soil reached physical maturity (35–60% of field capacity) and were friable without adhering to implements.

Table 1. Implementation protocols for tillage systems, showing detailed operational specifications, including equipment types, working depths, and timing of operations in the long-term field experiment (2010–2014). All field operations were conducted at standard equipment speed of 8–10 km/h, with spring operations initiated when soil moisture content reached 35–60% of field capacity to ensure optimal soil workability.

Treatment	Primary Operations	Secondary Operations	Pre-Sowing Operations	Sowing Method
Conventional Tillage	Stubble cultivation (Vaderstad Carrier CRL-425, 10–12 cm) followed by moldboard plowing (Kverneland ES-80/95, 23–25 cm) after 2–3 weeks	Residue incorporation during stubble cultivation	Combined pre-sowing implement (4–5 cm for cereals, 2–3 cm for rapeseed)	Vederstad Rapid 400C disc seeder
No Tillage	Stubble cultivation (10–12 cm)	Glyphosate application (4 L ha^{-1} in 200 L water) after 2–3 weeks	Direct drilling	Vederstad Rapid 400C disc seeder

Crop rotation systems (Factor B) included three distinct patterns—three-field rotation (with three variants), two-field rotation (with two variants), and monoculture, implemented as detailed in Table 2. Seeding rates were calculated assuming 100% seed viability and adjusted according to actual germination tests.

Table 2. Crop rotation systems and seeding specifications showing rotation sequences, crop-specific seeding rates, and recommended sowing periods. All seeding rates were calculated for 100% seed viability and adjusted according to actual germination tests.

Rotation Type	Sequence	Seeding Rates	Optimal Sowing Period
Three-field (3A)	Spring barley → winter wheat → spring oilseed rape	Spring barley: 4.0 million seeds ha^{-1} (400 plants m^{-2})	Spring barley: early spring
Three-field (3B)	Winter wheat → spring oilseed rape → spring barley	Winter wheat: 4.5 million seeds ha^{-1} (450 plants m^{-2})	Winter wheat: Sept 10–25
Three-field (3C)	Spring oilseed rape → spring barley → winter wheat	Spring oilseed rape: 6.0 kg ha^{-1} (110–120 plants m^{-2})	Spring oilseed rape: early spring
Two-field (2A)	Winter wheat → winter oilseed rape	Winter oilseed rape: 4.0 kg ha^{-1} (70–80 plants m^{-2})	Winter oilseed rape: Aug 5–15
Two-field (2B)	Winter oilseed rape → winter wheat		
Monoculture	Continuous winter wheat		

2.3. Soil Sampling and Analysis

Initial soil conditions were assessed in 2010 at the beginning of the experimental period. Undisturbed soil cores (100 cm^3) were collected using steel cylinders from two depths (5–10 cm and 15–20 cm) in spring when soil moisture was at field capacity. Three samples were taken from each experimental plot using random sampling within the harvest area, avoiding plot edges. Sample locations were determined by randomly throwing a sampling ring within the designated area. Samples were stored at +2 °C until analysis. These baseline measurements provided context for understanding the initial soil habitat conditions under different management treatments.

Soil moisture content was determined gravimetrically by drying samples at 105 °C to constant weight. Bulk density was calculated from the oven-dried weight of the undisturbed core samples. Total porosity was calculated from bulk density values using particle density of 2.65 g/cm^3. Aeration porosity was determined as the difference between total porosity and moisture-filled pore space at field capacity.

All samples were stored at +2 °C until analysis to preserve their physical condition. While these measurements represent initial conditions rather than temporal changes, they provide important baseline data for interpreting earthworm population responses to different management practices over the subsequent experimental period (2010–2014).

2.4. Earthworm Sampling and Analysis

Earthworm sampling was conducted post-harvest to avoid impact on crop yield assessment. Soil monoliths (25 cm × 25 cm in cross-section) were extracted to 25 cm depth, with one sample from each plot's harvest area. Sample locations were determined using random selection within the harvest area by throwing a sampling ring. Each monolith was carefully hand-sorted in the field on a 2 m × 2 m polyethylene sheet.

Collected earthworms were immediately transferred to containers with moistened filter paper and transported to the laboratory. Specimens were washed with distilled water, counted, and weighed to determine fresh biomass. Population density was calculated as individuals per m^2, and biomass was expressed as g per m^2. All measurements were conducted within 24 h of collection to ensure accurate biomass determination.

2.5. Meteorological Conditions

Weather data were collected from the Dotnuva Meteorological Station located near the research site. Mean monthly air temperature and precipitation data were recorded for the growing period (April–October), which was further divided into seasons for analysis: spring (April–May), summer (June–August), and autumn (September–October). The detailed weather conditions are presented in Table 3.

Table 3. Monthly weather conditions during the experimental period (2010–2014) recorded at Dotnuva Meteorological Station, Lithuanian Research Centre for Agriculture and Forestry. Data includes mean air temperature (°C), total monthly precipitation (mm), and number of days with precipitation ≥ 1 mm for each month of the growing season (April–October). Values are presented alongside long-term averages (LTA, 1924–2014) to contextualize experimental conditions within historical climate patterns.

Year	Month	Mean Air Temperature (°C)	Total Monthly Precipitation (mm)	Days with Precipitation ≥ 1 mm
2010	April	7.3	14.7	7
	May	13.7	31.4	15
	June	16.2	24.1	10
	July	21.7	47.3	11
	August	19.8	23.7	7
	September	11.9	17.4	9
	October	5.0	12.7	8
2011	April	8.8	7.8	6
	May	13.0	15.6	9
	June	18.1	15.0	8
	July	19.7	38.3	15
	August	17.4	34.6	11
	September	13.7	18.0	14
	October	7.6	8.0	7
2012	April	7.3	15.8	8
	May	13.3	14.0	8
	June	14.9	26.2	11
	July	18.9	40.1	10
	August	16.5	27.3	11
	September	13.2	14.3	8
	October	7.2	20.5	10
2013	April	4.8	15.4	10
	May	16.0	16.5	8
	June	18.6	15.5	10
	July	18.5	34.8	12
	August	18.0	14.1	7
	September	12.6	25.9	10
	October	8.5	9.6	5
2014	April	8.9	9.0	5
	May	13.0	25.6	11
	June	14.4	21.9	12
	July	20.0	23.0	6
	August	17.6	37.1	16
	September	13.3	7.2	3
	October	7.1	13.0	8
Long term average	April	5.9	36.9	8
	May	12.3	52.1	9
	June	15.7	62.3	10
	July	17.8	75.5	11
	August	16.7	74.2	11
	September	12.1	51.1	9
	October	6.8	49.6	9

For analytical purposes, the following seasonal patterns were examined:

- Spring (April–May): temperatures ranged from 6.1 °C to 12.4 °C, with precipitation varying from 11.7 mm to 34.6 mm.
- Summer (June–August): mean temperatures were 17.7–19.2 °C, with precipitation ranging from 24.4 mm to 37.0 mm.

- Autumn (September–October): temperatures averaged 8.5–10.7 °C, with precipitation between 10.1 mm and 22.8 mm.

The experimental period exhibited significant divergences from long-term averages (1924–2014), especially in precipitation patterns. The seasonal divisions were crucial for examining earthworm population dynamics and their responses to climatic conditions during the growing season.

2.6. Statistical Analysis

The Shapiro–Wilk test was used to evaluate data normality. To check homogeneity of variance, Levene's test was employed. Square root transformation was utilized for earthworm abundance and biomass analysis when necessary to satisfy the assumptions of parametric statistical analyses.

The impact of tillage system, crop rotation, and year on earthworm abundance and biomass was assessed by three-way ANOVA, designating tillage and crop rotation as fixed factors and year as a repeated measure.

Two categories were utilized for rotation analysis: broad rotations (three-field, two-field, and monoculture) and detailed rotations (particular crop combinations within each rotation classification).

Multiple comparisons tests were performed using Tukey's HSD test at $p \leq 0.05$.

Mixed-effects modeling was performed to assess temporal patterns, using tillage system and crop rotation as fixed factors and block as a random effect. Soil parameters were included as covariates. Year-by-year analyses using two-way ANOVA were performed in case significant time interactions were detected.

Soil parameters were recorded in winter wheat plots at two depths (5–10 cm and 15–20 cm) from 2010 to 2012. Repeated-measures ANOVA was used to investigate treatment effects over time. Relationship between soil parameters and earthworm populations was analyzed using Pearson correlation analysis.

Principal component analysis was conducted to investigate significant correlations between measured variables, specifically soil physical factors and earthworm populations. Weather impact was examined by correlation analysis between climatic variables and both soil conditions and earthworm populations.

Statistical analyses were conducted using SPSS version 30.0.0. Graphs were generated using SigmaPlot version 16.0.0.28. Results are expressed as means ± standard error unless specified otherwise, and effects were deemed significant at $p < 0.05$.

3. Results

3.1. Initial Soil Physical Properties and Their Relationship with Earthworm Populations

Physical properties sampled at two depths (5–10 cm and 15–20 cm) in winter wheat plots in 2010 showed differences between tillage systems (Table 4). Conventional tillage exhibited higher soil moisture content in the upper layer, as compared to no tillage. This difference was also significant ($p < 0.001$) in the deeper layer. Bulk density showed minimal variation between tillage systems in the upper layer, though differences became more pronounced at 15–20 cm depth.

The correlation between physical properties and earthworm populations was significant in the upper soil layer, revealing a negative correlation between earthworm abundance and bulk density ($r = -0.612$, $p = 0.041$) and a positive correlation between earthworm abundance and total porosity ($r = 0.583$, $p = 0.044$). Relationships were less evident in the deeper soil layer. This suggests that surface conditions significantly influenced earthworm habitat quality.

Table 4. Soil physical parameters under different tillage systems in winter wheat plots (2010). ** indicates highly significant difference between tillage systems ($p < 0.001$).

Tillage	N	Depth (cm)	Bulk Density ($Mg\ m^{-3}$)	Soil Moisture (%)	Total Porosity (%)	Aeration Porosity (%)
Conventional tillage	15	5–10	1.40 ± 0.02	20.0 ± 0.3 **	47.2 ± 0.8	16.8 ± 1.3
	15	15–20	1.42 ± 0.02	19.9 ± 0.4 **	46.4 ± 0.8	15.0 ± 1.1
No tillage	15	5–10	1.42 ± 0.02	17.9 ± 0.3 **	46.3 ± 0.9	18.2 ± 1.2
	15	15–20	1.47 ± 0.01	17.8 ± 0.3 **	44.4 ± 0.5	15.8 ± 0.6

3.2. Earthworm Population Dynamics

Data revealed a strong correlation between abundance and biomass (r = 0.927, $p = 0.023$). This indicates a stable community structure across treatments. Mixed-effects modeling indicated significant temporal variation ($F_{3,141} = 15.965$, $p < 0.001$) across populations.

Population data were analyzed using two approaches:

Broad Rotation Effects—examining rotations by generalized categories:

- Three-field rotation (combining all three variants A, B, and C).
- Two-field rotation (combining variants A and B).
- Monoculture.

Detailed Rotation Analysis—examining particular rotation sequences:

Variants of three-field rotations:

- 3A: spring barley → winter wheat → spring oilseed rape.
- 3B: winter wheat → spring oilseed rape → spring barley.
- 3C: spring oilseed rape → spring barley → winter wheat.

Two-field rotation variants:

- 2A: winter wheat → winter oilseed rape.
- 2B: winter oilseed rape → winter wheat.

Three-field rotation yielded the highest earthworm populations (abundance: 131.2 ± 7.0 individuals m^{-2}; biomass: 35.4 ± 3.2 g m^{-2}), followed by two-field rotation (abundance: 127.5 ± 9.4 individuals m^{-2}; biomass: 32.8 ± 2.9 g m^{-2}), whereas monoculture exhibited significantly lower values (abundance: 105.0 ± 13.2 individuals m^{-2}; biomass: 25.6 ± 2.7 g m^{-2}; $p < 0.048$ for abundance, $p < 0.036$ for biomass). Earthworm population dynamics patterns are represented graphically in Figures 2 and 3.

Among specific sequences, both abundance and biomass showed considerable variation across treatments. The three-field B rotation (winter wheat → spring oilseed rape → spring barley) demonstrated the highest values under no till, peaking in 2014 for abundance (260.0 ± 34.9 individuals m^{-2}) and in 2011 for biomass (57.63 ± 22.24 g m^{-2}). The lowest values for both parameters were recorded in the monoculture system under no till in 2013 (abundance: 30.7 ± 5.3 individuals m^{-2}; biomass: 8.53 ± 0.96 g m^{-2}).

The strong correlation between abundance and biomass (r = 0.927, $p = 0.023$) indicated a consistent population structure across treatments, though temporal patterns differed slightly between these parameters.

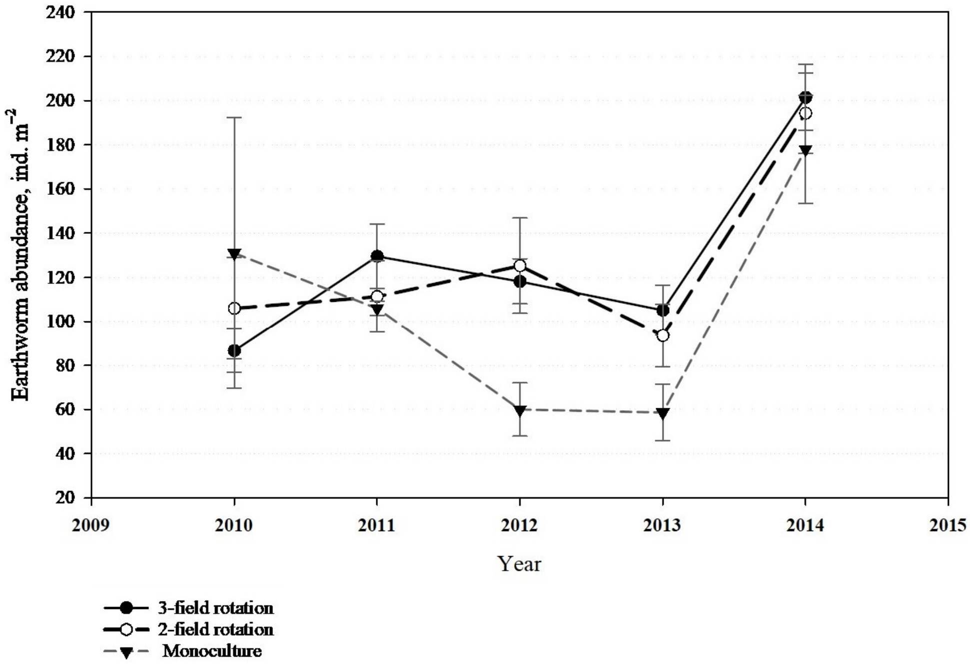

Figure 2. Earthworm population dynamics under different crop rotation systems—abundance (ind. m^{-2}) (2010–2014). Points and bars represent means ± standard error (n = 36 in 2011–2014; n = 24 in 2010). Rotation compositions: three-field (spring rape–spring barley–winter wheat), two-field (winter wheat–winter rape), and monoculture (continuous winter wheat). Samples were collected annually at post-harvest from 25 × 25 cm soil monoliths at 0–25 cm depth.

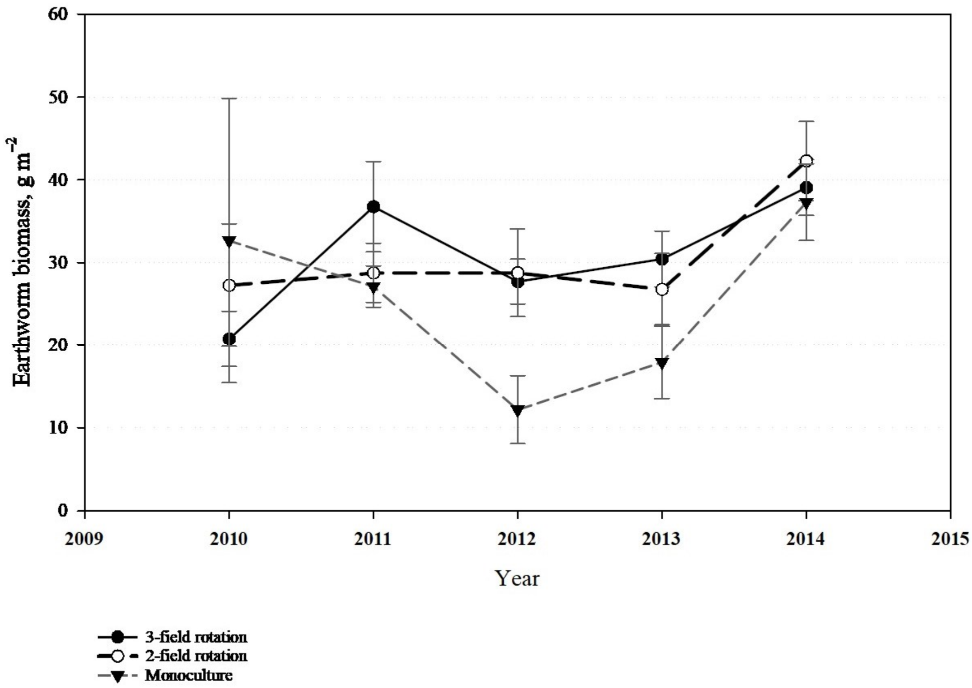

Figure 3. Earthworm population dynamics under different crop rotation systems—biomass (g. m^{-2}) (2010–2014). Points and bars represent means ± standard error (n = 36 in 2011–2014; n = 24 in 2010). Rotation compositions: three-field (spring rape–spring barley–winter wheat), two-field (winter wheat–winter rape), and monoculture (continuous winter wheat). Samples were collected annually at post-harvest from 25 × 25 cm soil monoliths at 0–25 cm depth.

3.3. Treatment Effects over Time

Three-way ANOVA demonstrated that temporal variation was the dominant factor influencing earthworm populations, with significant year effects observed for both abundance ($F_{4,108}$ = 17.227, $p < 0.001$, partial η^2 = 0.390) and biomass ($F_{4,108}$ = 6.438, $p < 0.001$, partial η^2 = 0.193). The impact of crop rotation showed variable effects on population parameters. Significance was marginal for abundance ($F_{5,108}$ = 2.116, p = 0.069, partial η^2 = 0.089) but significant for biomass ($F_{5,108}$ = 2.479, p = 0.036, partial η^2 = 0.103). Notably, the tillage type demonstrated no significant effect on either abundance ($F_{1,108}$ = 1.414, p = 0.237) or biomass ($F_{1,108}$ = 0.220, p = 0.640), suggesting that earthworm populations were more responsive to rotation practices than to tillage regimes over the study period. The main findings of the three-way ANOVA results for earthworm parameters are shown in Table 5 below.

Table 5. Three-way ANOVA results for earthworm parameters (* indicates significant effects ($p < 0.05$); † indicates marginally significant effects ($0.05 \leq p < 0.10$)).

Source of Variation	Response Variable	F-Value	df	*p*-Value	Partial η^2
Year	Abundance	17.227	4, 108	<0.001 *	0.390
	Biomass	6.438	4, 108	<0.001 *	0.193
Crop Rotation	Abundance	2.116	5, 108	0.069 †	0.089
	Biomass	2.479	5, 108	0.036 *	0.103
Tillage Type	Abundance	1.414	1, 108	0.237	0.013
	Biomass	0.220	1, 108	0.640	0.002

3.4. Management Effects over Time

Conventional tillage maintained slightly higher populations (abundance: 129.9 ± 7.4 individuals m^{-2}; biomass: 32.5 ± 2.8 g m^{-2}) compared to no till (abundance: 121.3 ± 7.3 individuals m^{-2}; biomass: 30.8 ± 2.6 g m^{-2}), though these differences were not statistically significant (p = 0.052 for abundance, p = 0.640 for biomass) (Table 5).

Year-by-year analysis (two-way ANOVA) on specific sequences shows significant rotation effects emerging in 2012 for both abundance ($F_{5,24}$ = 6.940, $p < 0.001$, partial η^2 = 0.591) and biomass ($F_{5,24}$ = 7.624, $p < 0.001$, partial η^2 = 0.614). Post hoc Tukey tests showed that three-field B and two-field A rotations significantly outperformed other treatments for both parameters ($p < 0.05$). The main findings of the year-by-year two-way ANOVA results are shown in Tables 6 and 7 below.

Table 6. Two-way ANOVA results by year—earthworm count (* indicates significant effects ($p < 0.05$); † indicates marginally significant effects ($0.05 \leq p < 0.10$); T × R represents Tillage × Rotation interaction).

Year	Source	F-Value	df	*p*-Value	Partial η^2
2010	Tillage	0.468	1, 30	0.623	0.015
	Rotation	0.297	2, 30	0.745	0.019
	T × R	0.384	2, 30	0.684	0.025
2011	Tillage	0.382	1, 30	0.541	0.013
	Rotation	0.468	2, 30	0.630	0.030
	T × R	0.294	2, 30	0.747	0.019
2012	Tillage	1.873	1, 30	0.181 †	0.059
	Rotation	4.047	2, 30	0.028 *	0.213
	T × R	0.582	2, 30	0.565	0.037
2013	Tillage	0.004	1, 30	0.949	<0.001
	Rotation	2.623	2, 30	0.089 †	0.149
	T × R	0.847	2, 30	0.439	0.053
2014	Tillage	0.262	1, 30	0.771	0.009
	Rotation	0.198	2, 30	0.821	0.013
	T × R	0.156	2, 30	0.856	0.010

Table 7. Two-way ANOVA results by year—earthworm biomass (* indicates significant effects ($p < 0.05$); † indicates marginally significant effects ($0.05 \leq p < 0.10$); T × R represents Tillage × Rotation interaction).

Year	Source	F-Value	df	*p*-Value	Partial η^2
2010	Tillage	0.524	1, 30	0.475	0.017
	Rotation	0.183	2, 30	0.834	0.012
	T × R	0.291	2, 30	0.749	0.019
2011	Tillage	0.428	1, 30	0.518	0.014
	Rotation	0.387	2, 30	0.682	0.025
	T × R	0.276	2, 30	0.761	0.018
2012	Tillage	1.762	1, 30	0.195 †	0.056
	Rotation	4.302	2, 30	0.023 *	0.223
	T × R	0.673	2, 30	0.518	0.043
2013	Tillage	0.183	1, 30	0.672	0.006
	Rotation	2.760	2, 30	0.079 †	0.155
	T × R	0.928	2, 30	0.406	0.058
2014	Tillage	0.247	1, 30	0.623	0.008
	Rotation	0.183	2, 30	0.834	0.012
	T × R	0.173	2, 30	0.842	0.011

3.5. Temporal Patterns

Mixed-effects analysis suggests three roughly distinct phases in system development over the five-year period of this study. The establishment phase (2010–2011) was characterized by high variability in abundance (CV > 60%) and biomass (CV > 58%). No significant effects for either broad rotation categories ($p > 0.297$) or specific sequences ($p > 0.315$) were observed. During this initial period, population levels averaged 115.4 ± 12.3 individuals m^{-2} and 28.7 ± 3.1 g m^{-2} in three-field rotations, 109.2 ± 11.8 individuals m^{-2} and 26.4 ± 2.9 g m^{-2} in two-field rotations, and 95.3 ± 10.9 individuals m^{-2} and 23.2 ± 2.8 g m^{-2} in monoculture.

The year 2012 showed significant rotation effects on both abundance ($F_{2,30} = 4.047$, $p = 0.028$, partial $\eta^2 = 0.213$) and biomass ($F_{2,30} = 4.302$, $p = 0.023$, partial $\eta^2 = 0.223$). For this reason, it was regarded as the differentiation phase. This phase showed clear separation between rotation treatments, with the three-field B rotation demonstrating the highest values (128.3 ± 8.7 individuals m^{-2}, 32.4 ± 2.6 g m^{-2}) and monoculture showing the lowest values (100.4 ± 7.9 individuals m^{-2}, 25.1 ± 2.3 g m^{-2}). Furthermore, during this period significant tillage effects ($F_{1,24} = 4.113$, $p = 0.054$) were observed for the first time.

The 2013–2014 period could be considered a stabilization phase, as variability reduced substantially across all treatments (CV ≈ 32%). During this phase, environmental correlations became more evident, with spring temperature effects showing significant influence ($r = 0.890$, $p = 0.043$) and precipitation patterns demonstrating increased importance. Peak populations were achieved during this period in 2014, with the three-field B rotation under no till reaching 260.0 ± 34.9 individuals m^{-2}, while the system-wide mean stabilized at 195.2 ± 8.9 individuals m^{-2}. This development pattern was consistent across both broad rotation categories and detailed sequence analyses, though specific sequences showed varying rates of stabilization.

3.6. Preceding Crop Effects

Preceding crop effects showed a clear hierarchy in their influence on earthworm populations, with break crops consistently supporting higher abundances and biomass compared to cereals. Winter oilseed rape as a preceding crop supported the highest earthworm populations (144.6 ± 13.2 individuals m^{-2}, 32.2 ± 2.6 g m^{-2}), followed closely by spring oilseed rape (142.7 ± 12.1 individuals m^{-2}, 33.9 ± 2.8 g m^{-2}). Spring barley maintained intermediate population levels (138.9 ± 11.2 individuals m^{-2}, 32.5 ± 2.9 g m^{-2}), while winter wheat showed substantially lower support for earthworm communities (117.5 ± 8.7 individuals m^{-2}, 29.6 ± 2.2 g m^{-2}). The absence of a preceding crop (initial year of the study) resulted in the lowest population parameters (100.5 ± 13.1 individuals m^{-2}, 24.9 ± 4.0 g m^{-2}), demonstrating the importance of crop residues and root systems in maintaining earthworm populations. Graphic representations of the preceding crops' impact on earthworm counts and biomass are provided in Figures 4 and 5 below.

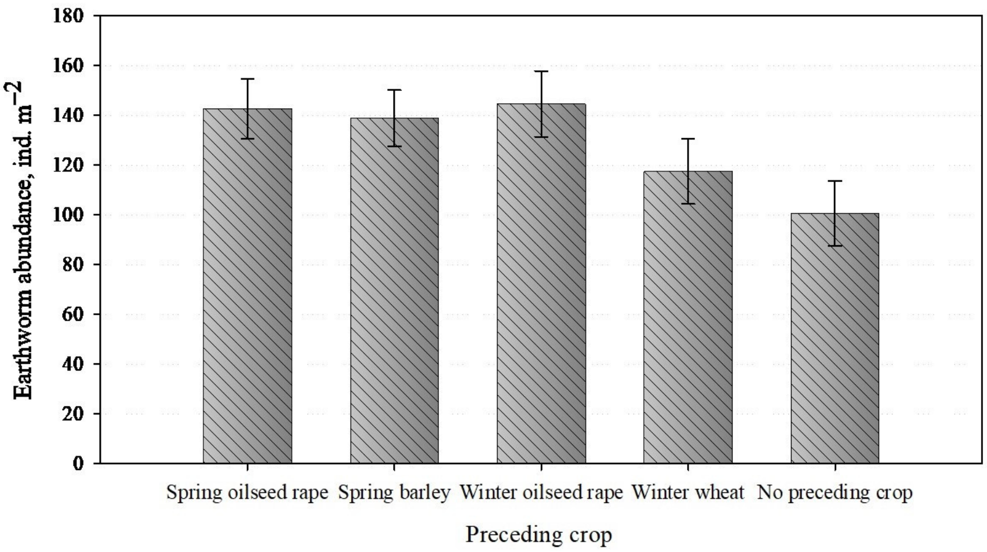

Figure 4. Effect of preceding crops on earthworm populations—abundance (ind. m^{-2}) (2010–2014). Bars represent means ± standard error (n = 36 in 2011–2014; n = 24 in 2010). Samples were collected from 25 × 25 cm soil monoliths at 0–25 cm depth at post-harvest.

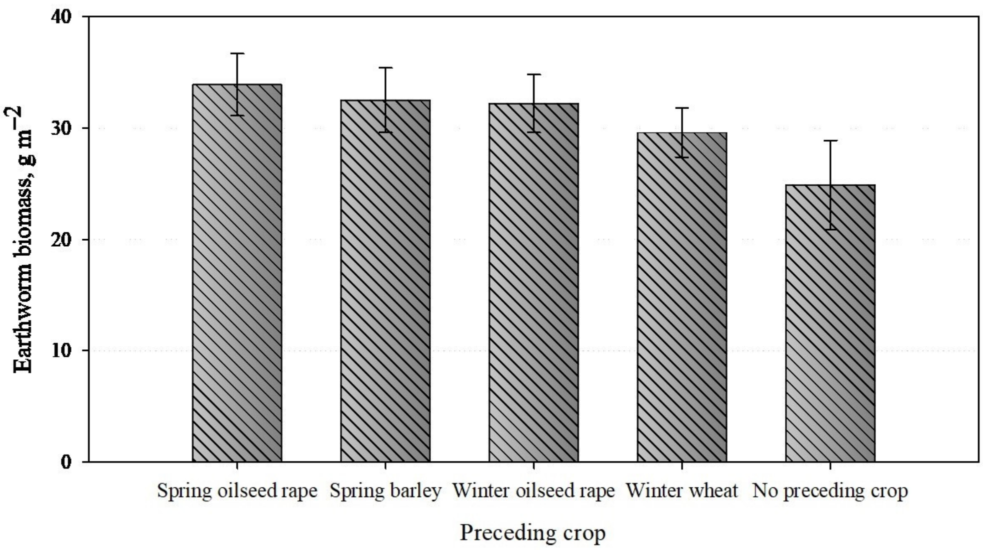

Figure 5. Effect of preceding crops on earthworm populations—biomass (g m^{-2}) (2010–2014). Bars represent means ± standard error (n = 36 in 2011–2014; n = 24 in 2010). Samples collected from 25 × 25 cm soil monoliths at 0–25 cm depth at post-harvest.

3.7. Weather Effects and Population Response

Spring temperature showed strong positive correlations with both abundance (r = 0.867, p = 0.048) and biomass (r = 0.890, p = 0.043), while autumn precipitation showed a negative correlation with both parameters (abundance: r = −0.871, p = 0.054; biomass: r = −0.901, p = 0.037). More details on correlations between factors are presented in Table 8.

Table 8. Correlation coefficients between weather parameters and earthworm population characteristics (2010–2014). (**: significant at $p < 0.05$; *: approaching significance at $p \sim 0.05$.)

Weather Parameter	Period	Abundance		Biomass	
		r	p-Value	r	p-Value
Temperature	Spring Summer Autumn	0.867 ** 0.534 0.423	0.048 0.112 0.183	0.890 ** 0.512 0.445	0.043 0.124 0.171
Precipitation	Spring Summer Autumn	0.645 −0.534 −0.871	0.087 0.112 0.054 *	0.678 −0.556 −0.901	0.082 0.107 0.037 **
Days with precipitation >1 mm	Spring Summer Autumn	0.712 −0.445 −0.823	0.073 0.171 0.058 *	0.734 −0.467 −0.845	0.068 0.162 0.056 *

3.8. System Stability

Analysis of system stability indicated strong similarities in population variability across different rotating systems (Table 9). The three-field rotation demonstrated the strongest stability, with coefficient of variation values of 49.2% and 51.4% for abundance and biomass, respectively. Two-field rotation systems showed intermediate stability levels (CV = 55.1% for abundance, 57.3% for biomass), while monoculture displayed the largest variability in both population metrics (CV = 66.7% for abundance, 68.9% for biomass). Regarding various rotation sequences, three-field B rotation emerged as the most stable system with the lowest variability (CV = 45.3%), followed by the two-field A rotation showing moderate variability (CV = 52.8%). All other rotation sequences revealed larger variability in population characteristics, with a coefficient of variation ranging from 54.2% to 66.7%. This stability gradient shows that higher crop diversity within rotation sequences contributes to more consistent earthworm population maintenance over time.

Table 9. Coefficient of variation (%) for earthworm parameters under different management regimes (2010–2014). CV—coefficient of variation. Three-field rotation sequences: A (spring barley → winter wheat → spring oilseed rape), B (winter wheat → spring oilseed rape → spring barley), and C (spring oilseed rape → spring barley → winter wheat). Two-field rotation sequences: A (winter wheat → winter oilseed rape) and B (winter oilseed rape → winter wheat).

Management Regime	CV for Abundance (%)	CV for Biomass (%)
Rotation type		
Three-field	49.2	51.4
Three-field A	55.3	57.8
Three-field B	45.3	48.2
Three-field C	52.4	54.6
Two-field	55.1	57.3
Two-field A	52.8	55.1
Two-field B	57.3	59.4
Monoculture	66.7	68.9
Tillage system		
Conventional tillage	54.2	56.5
No tillage	52.1	54.3
Development phase		
Establishment phase (2010–2011)	60.3	62.8
Differentiation phase (2012)	45.	47.9
Stabilization phase (2013–2014)	32.4	34.7

4. Discussion

This five-year study reveals complex dynamics in earthworm populations in response to crop rotation and tillage practices. The strong correlation between abundance and biomass (r = 0.927, p = 0.023) across treatments suggests consistent population structure despite varying management conditions, indicating robust community development.

4.1. Soil Physical Properties and Soil–Earthworm–Weather Relationships (2011–2012)

The baseline soil physical parameters assessed in 2010 provide essential background for comprehending earthworm habitat conditions across various management regimes. Although conventional tillage preserved higher soil moisture levels, the inverse relationship between bulk density and earthworm population indicates that soil structural features may be more pivotal in assessing habitat quality than moisture availability alone. This corresponds with findings that soil moisture content is a critical factor influencing earthworm population dynamics [15], while earthworms require specific soil physical conditions for optimal growth and reproduction [5,33].

The depth-specific nature of these interactions suggests that management effects on earthworm habitat are predominantly focused within the surface layer. Nevertheless, the comprehensive dynamics of the earthworm population over the five-year span indicate that initial soil physical conditions most likely diminish in significance as systems mature, while other factors, notably management practices (crop diversity in rotations) and environmental conditions, become more critical in influencing earthworm abundance and distribution.

4.2. Rotation Effects and System Complexity

Both broad and detailed rotation analyses revealed important patterns. At the broad level, three-field rotations supported higher and more stable populations (131.2 ± 7.0 ind. m^{-2}, 35.4 ± 3.2 g m^{-2}) compared to monoculture (105.0 ± 13.2 ind. m^{-2}, 25.6 ± 2.7 g m^{-2}). This finding aligns with evidence that crop diversification enhances soil biodiversity through improved resource availability [5] and provides better conditions for soil fauna development [9]. Analysis at the detailed level indicated that specific rotation sequences (notably three-field B—winter wheat → spring oilseed rape → spring barley) created optimal conditions for earthworms, and this aligns with prior research indicating that diversified crop rotations promote varied organic inputs and enhance soil stability [34].

Multiple factors may contribute to the enhanced performance of the three-field B rotation [35,36]:

- Rotation of winter and spring crops to ensure balanced disturbance patterns.
- A combination of cereals and break crops enhances resource variety.
- Varied root systems create complex soil habitats.

4.3. System Development

The significant year effect observed for both abundance and biomass ($F_{4,108}$ = 17.227 and 6.438, respectively, p < 0.001) highlights the necessity for long-term research aimed at understanding soil ecological processes. The system exhibited several well-expressed phases, evolving from significant variability (CV > 60%) in its early years to enhanced stability (CV ≈ 32%) by 2014, indicating effective ecosystem maturation [37]. This temporal trend corresponds with research indicating that diverse cropping systems improve soil ecosystem resilience [8] and bolster the stability of soil biological communities [38].

4.4. Relationship Between Weather and Earthworm Population Dynamics

Strong correlation between spring temperature and earthworm parameters (abundance: r = 0.867, p = 0.048; biomass: r = 0.890, p = 0.043) align with previous findings about temperature thresholds for earthworm activity [39]. These patterns are supported by research that indicates that earthworm activity is significantly affected by temperature and moisture conditions [16], and these environmental factors can supersede management influences in shaping earthworm populations [38]. The negative impact of autumn precipitation suggests potential stress from excess moisture, an important consideration which might provide some insights on fluctuations in earthworm abundance.

The thorough analysis of earthworm abundance and biomass dynamics over a five-year period provides several key insights for agricultural management.

The clear progression through establishment, differentiation, and stabilization phases suggests that management expectations should be adjusted according to system age. Initial establishment periods (1–2 years) exhibited high variability (CV > 60%) and no significant management responses ($p > 0.297$) neither for abundance nor for biomass. The emergence of significant treatment effects in year three (abundance: $F_{5,24} = 6.940$, $p < 0.001$; biomass: $F_{5,24} = 7.624$, $p < 0.001$) suggests this as a critical evaluation point for management adjustment.

Statistical evidence consistently indicates that crop diversification effects supersede tillage impacts. While tillage showed only marginal effects (abundance: $F_{1,162} = 1.550$, $p = 0.215$; biomass: $F_{1,162} = 0.627$, $p = 0.430$), rotation impacts were significant, particularly through preceding crop effects. The strong performance of break crops (142.7–144.6 ind. m^{-2}, 37.5–38.2 g m^{-2}) compared to cereals (117.5 ind. m^{-2}, 28.4 g m^{-2}) suggests that crop sequence planning may be more crucial than tillage intensity reduction.

4.5. Unexpected Resilience to Management Practices

A key finding of this investigation was the absence of considerable adverse effects from cropping practices on earthworm populations. Conventional knowledge and substantial research indicate that intensive farming practices generally diminish earthworm populations, with studies revealing reductions of up to 70% in earthworm biomass and 80% in abundance relative to less intensive tillage systems [10,18], especially due to plowing [15]. Our data indicate significant resilience in earthworm groups across various management regimes.

This unforeseen result may be explained by multiple factors: Firstly, the well-established nature of the experimental areas, where soil ecosystems may have acclimated to prolonged management. Secondly, the rather moderate environment of Central Lithuania may mitigate potential adverse consequences of management practices. Thirdly, maintaining adequate soil moisture levels throughout treatments, highlighted by [5] as essential for earthworm survival irrespective of management intensity.

This finding is notably important as it contests prevalent convictions regarding the susceptibility of soil fauna to agricultural methods and indicates that earthworm populations may exhibit greater resilience to agricultural intensification than previously assumed, provided that essential habitat requirements are met. The preservation of earthworm populations under varying management intensities suggests that farmers may possess more flexibility in selecting agricultural approaches without compromising soil biological health.

5. Conclusions

This five-year study partially rejects the initial hypothesis that simplified tillage and shortened crop rotations negatively impact earthworm populations in agricultural systems. Most notably, this study reveals an unexpected resilience of earthworm populations to agricultural management practices. The findings demonstrate the following:

1. Initial soil physical properties (measured in 2010) differed between tillage systems, with conventional tillage showing higher moisture content (19.9% vs. 17.9%) but

lower aeration porosity (15.4% vs. 17.1%) compared to no tillage. However, restriction of soil measurements only to the initial year of this study is limiting deeper insights into long-term impacts on soil parameters.

2. Strong correlation between soil physical properties and earthworm populations demonstrates a strong relationship, especially within upper soil layer conditions ($r = 0.823$, $p = 0.034$). Environmental factors, notably spring temperature ($r = 0.890$, $p = 0.043$) and autumn precipitation ($r = -0.901$, $p = 0.037$), emerge as increasingly critical determinants of earthworm populations as systems mature.
3. Tillage intensity has less impact on earthworm populations than initially hypothesized, with no significant differences between conventional and no-till systems ($F_{1,108} = 1.414$, $p = 0.237$ for abundance; $F_{1,108} = 0.220$, $p = 0.640$ for biomass). This is somewhat contradictory to traditional assumptions about reduced tillage benefits for soil biological health. Nonetheless, as seen by the temporal patterns in system stability increasing, the influence of tillage regime may become significant over an extended duration, which has not been addressed in this study.
4. Rotation effects depend more on crop diversity than length. Both two-field and three-field rotations maintain similar population levels (127.5–131.2 ind. m^{-2}, 32.8–35.4 g m^{-2}), while only monoculture shows significantly lower populations (105.0 $\pm$ 13.2 ind. m^{-2}, 25.6 $\pm$ 2.7 g m^{-2}; $p < 0.048$). The three-field B rotation (winter wheat $\rightarrow$ spring oilseed rape $\rightarrow$ spring barley) could be considered as particularly effective in terms of supporting healthy earthworm populations.

The findings of this study infer that crop diversity and sequence optimization could be equally effective or in some instances even surpass tillage simplification. The demonstrated resilience of earthworm populations across different management regimes suggests greater flexibility in agricultural practice selection than previously thought. Specific focus on including break crops should be included. Shortened rotations can maintain earthworm populations at sustainable levels; however, maintaining crop diversity is an essential component to ensure healthy soil conditions.

Future research should address the following:

- Long-term monitoring of soil physical parameters to understand their evolution under different management regimes.
- Species-specific earthworm responses to management practices.
- Weather and land management interactions for climate-resilient agricultural systems.
- Economic analysis of rotation diversity benefits versus operational simplification.
- Integration of these findings with other soil quality indicators.

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