

Article

# Enzymatic Activity of Soil Under Spelt Grown in An Organic Farming System in Poland's Temperate Climate

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**Abstract:** Wheat spelt is a very valuable plant, especially in organic farming. Its high nutritional values combined with low soil and climate requirements increase the interest in growing spelt in various soils. Spelt can be grown in different soil types, including sandy, wet, poor, non-draining and low-fertility soils. It is well adapted to cold climate. Compared to common wheat, it requires less nutrients and is more competitive against weeds. Activities of enzymes in soil under winter spelt have not been studied yet. We sought to determine whether the choice of varieties will also shape the enzymatic activities in different soil types and whether these activities will be the same under different climatic conditions of 2007, 2010 and 2013 year. The aim of this field experiment was to assess the impact of growing different winter spelt varieties (Oberkulmer Rotkorn, Franckenkorn, Schwabenkorn, Ostro) on the enzymatic activity on two different soil complexes. In the years 2007–2013, two three-year rotations of the experiment were carried out (the first ended in 2010 and the second in 2013). Spelt was cultivated in an organic system. Physicochemical properties of the two different types of soil after three and six years of the experiment were compared to the soil properties before the experiment. The catalase activity ranged between 3.33 and 6.75  $\mu\text{mol H}_2\text{O}_2 \text{ g}^{-1} \text{ d.m. min}^{-1}$ , the dehydrogenase activity ranged between 1.6 and 87.7  $\text{nmol TPF g}^{-1} \text{ d.m. 24 h}^{-1}$ , and the arylsulfatase activity ranged between 37.5 and 157  $\mu\text{mol pNF g}^{-1} \text{ d.m. h}^{-1}$ . The enzymatic activity in the soil depended on the type of soil (as a rule, the activity was higher in a stagnic luvisol than in a haplic cambisol) and on the spelt variety (although no variety had a clear beneficial effect on the activities of all enzymes). Spelt cultivation in an organic system led to an increase of enzymatic activity in a stagnic luvisol, but not always in a haplic cambisol soil type.

**Keywords:** stagnic luvisol; haplic cambisol; dehydrogenases; catalase; arylsulfatase; spelt wheat; soil enzymatic activity

## 1. Introduction

In addition to the physical and chemical properties of soil, biologic properties are important in determining soil fertility. Biologic parameters include enzyme activities and microbial biomass, as well as the composition and abundance of microorganisms. Many researchers believe that enzymes may be sensitive indicators of changes taking place in the soil [1–4]. The results obtained using the methods of measurements of soil enzyme activity inform about the intensity of the narrow range of transformations catalyzed by the tested enzymes. Oxidoreductases belong to enzymes frequently

used as biologic parameters. Dehydrogenase activity is an indicator of the intensity of respiration of soil microorganisms. Catalase is an enzyme that breaks down hydrogen peroxide into water and molecular oxygen. An important group of enzymes are also those which facilitate the use of nutrients from organic compounds. For instance, arylsulfatase is involved in hydrolysis of sulfur-containing organic compounds, which leads to sulfate sulfur formation. This process leads to soil enrichment in readily available sulfur compounds, which improves plant yield (in the case of wheat cultivation– it increases the content of exogenous sulfur amino acids and improves protein quality).

The abundance of microorganisms in the soil and the activities of the enzymes depend on many factors determined by soil management and environmental conditions (e.g., soil pH, water and air relationships and the content of organic compounds) [5,6]. Microorganisms and vegetation determine the direction and nature of biochemical processes and create balance in soil environments [7]. The aim of plant production is to obtain maximum weight and quality of the crop, typically achieved through appropriate cultivation practices in conjunction with selected types and doses of fertilizers and plant protection products. The usual effect is an increase in soil fertility. However, inadequate management can cause disturbances in the functioning of agroecosystems, including soil biologic activity [8,9].

Wheat, particularly winter wheat, is the main cereal species cultivated in Poland [10]. Intense and conventional production systems are based on the cultivation of high-yield species (which are characterized by appropriate quality parameters). Organic cultivation of wheat is an alternative [11], and older wheat species are becoming increasingly popular in organic production, including one of the oldest known, spelt (*Triticum spelta* L.). Spelt owes its revival to organic production, as it is one of the least demanding cereals with regard to cultivation conditions [12–14]. Spelt is resistant to adverse weather conditions and can be grown in poor soils. Spelt has high radiation use efficiency and does not require high doses of fertilizers. Used in bread production, spelt may have better nutritional properties than common wheat [15]. In Poland, 1,637 ha were used to cultivate spelt in 2010 [16].

Soil is the primary environmental factor determining wheat yield. Plant growth and development may depend on soil enzyme activities [17] and on the type of soil microflora, particularly in the vicinity of the roots (i.e., in the rhizosphere) [18], although (because of interactions of weather conditions, soil properties, plant species) relationships between plants and microorganisms may not always be possible to confirm [19]. The root system plays an important role in the life of plants and in soil fertility as it constitutes from 50% to 70% of the total weight of crop residue and it is a source of energy for soil microbes. An increased weight of the root system is one of the advantages of organic crop cultivation when compared to conventional cultivation [20].

The available literature lacks studies on the enzymatic activity in soil under wheat spelt. This was the reason for undertaking research in this field. The aim of the study was to analyze the impact of selected winter spelt varieties (Oberkulmer Rotkorn, Franckenkorn, Schwabekorn, Ostro) on the enzymatic activity of two different soil types under organic wheat spelt. Spelt was cultivated in an organic system. Catalase, dehydrogenase and arylsulfatase activities were tested in subsequent years of the experiment.

## 2. Materials and Methods

### 2.1. Field Site and Experiment Descriptions

The field experiment was established in 2007 at the Experimental Station of the Department of Agrotechnology and Agricultural Ecology in Kraków–Mydlniki, belonging to the University of Agriculture in Kraków, Poland (50°05'32.8"N 19°51'31.8"E). In 2007–2013, two 3-year rotation experiments were performed (the first rotation was completed in 2010 and the second in 2013). The experiment was in a randomized block design with four replications. In both rotations, tests were carried out in two types of soils [21]: stagnic luvisol (SL) and haplic cambisol (HC), classified in two suitability complexes: good wheat (2<sup>nd</sup>) and good rye (5<sup>th</sup>) in Polish agronomic nomenclature. Winter spelt was cultivated under an organic system. The forecrop for the four varieties of spelt (Oberkulmer Rotkorn, Franckenkorn, Schwabekorn, Ostro) was a mixture of grain and legumes (oat + spring

vetch) grown for seed. Spelt was sown in autumn in optimal agricultural time, 250 spikes m<sup>-2</sup>. The area of each harvest plot was 18 m<sup>2</sup>.

The analysis of soil properties was performed before starting the experiment in autumn 2007 on the pooled samples for each of the soil suitability complexes. The soil analysis was repeated after three and six years of the experiment (in 2010 and 2013), following the harvest. Soil samples were collected from the topsoil (0–20 cm). From each 18 m<sup>2</sup> plot, about 10–12 individual samples of approximately 100 g soil fresh matter were collected at random using Egner's stick. Individual samples from one plot were then mixed together and an average sample of about 500 g soil fresh matter was taken. Laboratory analyses were conducted on dried and sieved samples (1 mm mesh), excluding determinations of enzymatic activity. Those determinations were conducted on fresh and sieved samples (2 mm mesh) with moisture determined by weight method.

The weather conditions in the years of research were varied. Total precipitation and the average daily air temperature in the discussed period were different compared to the average from the multiyear period (1961–1990) adopted as the standard under the conditions of the experiment. Individual years of research also had different distribution of temperature (Table 1) and precipitation (Table 2) in individual months, which modified the development conditions of the studied plants.

**Table 1.** Mean temperatures (°C) during the course of study.

Year	Month												J.-D.
	J.	Fb.	Mr.	Ap.	M.	Jn.	Jl.	Ag.	Sp.	Oc.	Nv.	D.	
2007	3.2	1.2	6.0	8.5	15.2	18.4	19.4	19.0	12.4	7.7	0.8	-1.1	9.23
2008	1.2	2.2	3.8	9.1	13.6	18.4	18.7	18.2	12.6	9.6	3.3	-1.2	9.13
2009	-3.4	-1.2	2.7	11.4	13.6	16.0	19.9	18.6	12.9	9.8	5.7	-1.2	8.73
2010	-6.3	-2.2	3.3	9.0	12.8	17.5	20.7	18.4	12.3	5.3	5.9	-5.3	7.62
2011	-1.2	-2.6	3.6	10.3	13.5	18.2	17.7	19.0	14.1	8.6	2.1	1.6	8.74
2012	-1.2	-6.6	4.4	9.4	15.0	17.4	20.3	18.9	14.0	8.6	5.0	-2.9	8.53
2013	-2.4	-0.5	-0.9	8.9	14.3	17.5	19.4	19.1	12.0	9.9	4.9	1.2	8.62
2007–2013	-1.4	-1.4	3.3	9.5	14.0	17.6	19.4	18.7	12.9	8.5	4.0	-1.3	8.65
1961–1990 <sup>1</sup>	-3.3	-1.6	2.4	7.9	13.1	16.2	17.5	16.9	13.1	8.3	3.2	-1.0	7.73

<sup>1</sup> multiyear period 1961–1990.

**Table 2.** Sum of precipitation (mm) during the course of study.

Year	Month												J.-Dc.
	J.	Fb.	Mr.	Ap.	M.	Jn.	Jl.	Ag.	Sp.	Oc.	Nv.	Dc.	
2007	100.6	42.2	61.1	15.4	51.7	72.1	71.0	76.4	179.8	48.3	90.4	21.4	830.4
2008	31.4	18.1	70.1	35.2	28.7	26.7	142.6	41.6	96.7	52.0	43.2	46.8	633.1
2009	28.0	40.1	66.5	4.7	106.6	122.1	82.7	53.3	61.5	42.7	27.6	36.2	672.0
2010	44.2	31.5	31.0	39.9	299.0	135.1	105.2	127.5	112.8	11.2	47.8	35.1	1020.3
2011	25.9	8.1	15.2	77.7	48.0	33.0	186.4	73.1	14.4	31.5	0.30	38.1	551.7
2012	51.6	27.9	17.3	49.0	17.5	143.8	70.6	54.6	43.7	96.3	22.1	27.2	621.6
2013	62.0	22.1	32.3	20.1	98.8	213.1	27.2	25.7	86.1	13.7	70.8	21.6	693.5
2007–2013	49.1	27.1	41.9	34.6	92.9	106.6	98.0	64.6	85.0	42.2	43.2	32.3	717.5
1961–1990 <sup>1</sup>	34.0	32.0	34.0	48.0	83.0	97.0	85.0	87.0	54.0	46.0	45.0	41.0	686.0

<sup>1</sup> multiyear period 1961–1990.

Comparing the temperature distribution and total temperature in individual months and years of research, large variations were observed. In all the years of research, the average temperature during the wheat spelt growing season was higher than the average from the multiyear period (7.73 °C) except for 2010 (7.62 °C). The characteristics of humidity conditions (Table 2) are presented on the basis of monthly precipitation totals for each year of the experiment. The amount of precipitation for individual years (Table 2) was characterized according to the Kaczorowska criterion [22]. Each year was considered average or in one of three aridity levels or one of three levels of excess rainfall.

Based on the humidity characteristics, four of the years of research were average according to the classification. The four years in question were 2008, 2009 (when precipitation accounted for 92% and 98% of the standard, respectively), 2012 (with 91% of the precipitation standard) and 2013 (101%

of the standard). The year 2007 was characterized as wet (121% of the standard), 2010 as very humid (149% of the standard) and 2011 as dry—the recorded rainfall was within 80% of the standard.

## 2.2. Laboratory Procedures

The soil pH was determined by potentiometry in a 1-mol L<sup>-1</sup> potassium chloride suspension (m/v 1:2.5) [23]. The content of total carbon, nitrogen and sulfur in the soil was determined using a vario MAX cube CNS elemental macroanalyzer (Elementar Analysensysteme GmbH) [24]. Sulfate sulfur was extracted from the soil using 0.03-mol dm<sup>-3</sup> acetic acid (30 min, 40 rpm, m/v 1:10) [22]. Sulfur content in the resulting solutions was determined using inductively coupled plasma optical emission spectrometry (ICP-OES) on an Optima 7300DV (PerkinElmer).

The activities of dehydrogenases and arylsulfatase was determined by a colorimetric method using a UV-Vis Beckman DU 640. To determine the dehydrogenase activity, the soil was incubated for 24 h at 37 °C with 2,3,5-triphenyl tetrazolium chloride (TTC), which was enzymatically reduced to triphenylformazan (TPF) [25]. The reaction product was extracted with methyl alcohol; TPF content was determined at a wavelength of 485 nm. To determine the arylsulfatase activity, the soil was incubated with p-nitrophenyl sulfate (1 h, 37 °C) [26]. The content of the resulting p-nitrophenol (pNF) was determined at a wavelength of 400 nm. Determination of catalase activity was performed using the manganometric method [25]. Reactions of the aqueous suspensions of soil with 0.3% hydrogen peroxide solution were performed at room temperature (30 rpm, 20 min). Excess hydrogen peroxide was titrated with an aqueous solution of potassium permanganate (VII) in the environment of sulfuric acid (VI).

## 2.3. Data Statistical Analysis

The enzymatic activity under the different treatments was statistically compared for each rotation of the experiment. A two-way analysis of variance (factor 1: variety, factor 2: soil type) was performed. The significance of variation among mean values was determined using Tukey's test at a significance level  $\alpha \leq 0.05$ . The statistical analysis of the results was performed with Statistica v12 software (StatSoft, Inc.). Determinations of the soil basic chemical properties, complementing the determinations of biologic parameters, were also subjected separately to statistical analysis for each rotation of the experiment. A one-way analysis of variance was performed (factor: soil type) while maintaining the other previously mentioned parameters of the analysis.

## 3. Results

### 3.1. Chemical Soil Properties

Table 3 shows the results of selected physicochemical properties of the soils before the experiment (2007), after the first rotation (2010), and after the second rotation (2013). Prior to the experiment, the SL soil was slightly acidic. Total carbon content was 7.14 g kg<sup>-1</sup> d.m., total nitrogen 0.595 g kg<sup>-1</sup> d.m., total sulfur 55.0 mg kg<sup>-1</sup> d.m. and sulfate sulfur 6.03 mg kg<sup>-1</sup> d.m. The C:N:S ratio was 130:11:1. Before the experiment, the HC soil was acidic. Total carbon content was 8.98 mg kg<sup>-1</sup> d.m., total nitrogen 0.945 mg kg<sup>-1</sup> d.m., total sulfur 75.0 mg kg<sup>-1</sup> d.m. and sulfate sulfur 6.20 mg kg<sup>-1</sup> d.m. The C:N:S ratio was 120:13:1.

**Table 3.** Means and SDs of selected soil properties under different soil types

Soil Type	pH <sub>KCl</sub>	C <sub>total</sub> (g kg <sup>-1</sup> d.m. ± SD)	N <sub>total</sub>	S <sub>total</sub> (mg kg <sup>-1</sup> d.m. ± SD)	S <sub>SO4</sub>	S <sub>SO4</sub> in S <sub>og</sub> (%)	C:N:S
<b>2010</b>							
Stagnic luvisol	5.28 a	8.47 b ± 0.23	1.006 b ± 0.051	106.3 b ± 4.79	7.69 b ± 0.79	7.23	80:9:1
Haplic cambisol	5.59 b	6.40 a ± 0.44	0.688 a ± 0.070	85.0 a ± 7.07	5.13 a ± 0.83	6.04	75:8:1

2013							
Stagnic luvisol	5.11 a	7.37 a ± 0.33	0.955 b ± 0.053	97.5 b ± 6.45	7.90 a ± 0.95	8.10	76:10:1
Haplic cambisol	5.78 b	6.98 a ± 0.39	0.756 a ± 0.017	76.3 a ± 11.81	6.83 a ± 1.54	8.95	92:10:1
2007 (before starting the experiment)							
Stagnic luvisol	5.58	7.14 ± 0.09	0.595 ± 0.007	55.0 ± 0.1	6.03 ± 0.07	10.96	130:11:1
Haplic cambisol	4.99	8.98 ± 0.02	0.945 ± 0.007	75.0 ± 0.1	6.20 ± 0.04	8.27	120:13:1

Mean values of soil properties in columns under different soil types for a given experiment rotation (2010 or 2013) marked with the same letters are not significantly different according to Tukey's test at a significance level  $p \leq 0.05$ ; single-factor analysis (factor: soil type: stagnic luvisol and haplic cambisol); SD—standard deviation.

In 2010, the HC soil was characterized by a significantly lower content of total carbon, nitrogen and sulfur and of sulfate sulfur, compared to the SL soil (Table 3). Acidification of the HC soil was significantly lower than that of the LS soil (slightly acidic and acidic reaction, respectively). The same relations were observed in 2013, in relation to the total content of nitrogen and sulfur and pH.

### 3.2. Activities of Enzymes

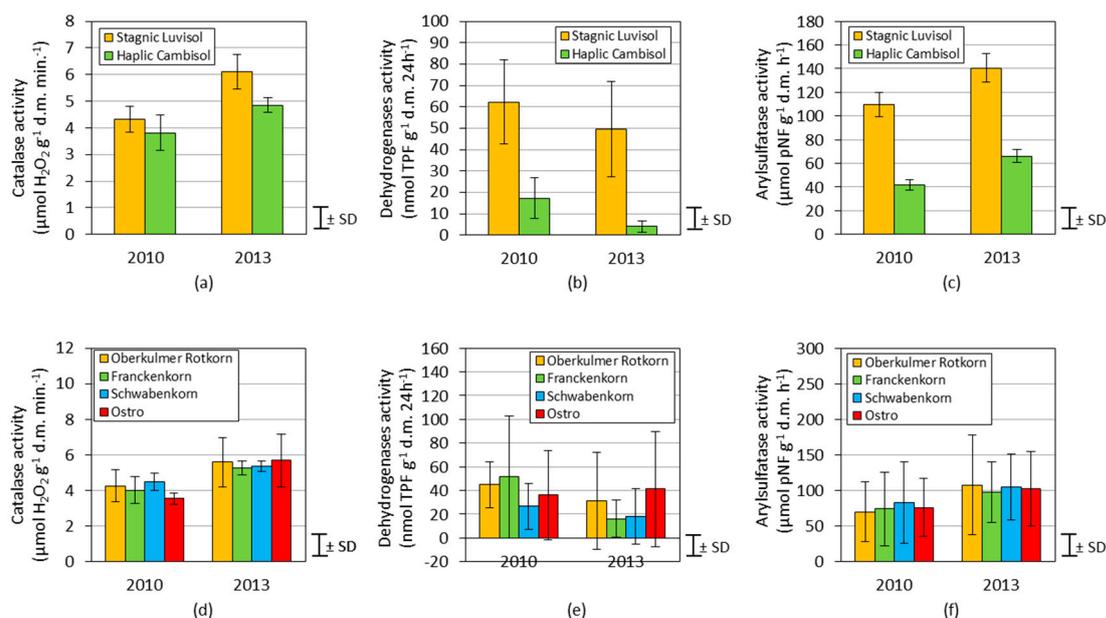
The activities of catalase, dehydrogenase and arylsulfatase in the tested soil samples depended on the soil type (usually significantly higher in the SL) and the cultivated spelt variety (Table 4).

**Table 4.** Means and SDs of activities of catalase, dehydrogenases and arylsulfatase in soil.

Year	Soil Type	Spelt Variety	Catalase Activity ( $\mu\text{mol H}_2\text{O}_2 \text{ g}^{-1}$ d.m. $\text{min}^{-1} \pm$ SD)	Dehydrogenases Activity ( $\text{nmol TPF g}^{-1}$ d.m. $24 \text{ h}^{-1} \pm$ SD)	Arylsulfatase Activity ( $\mu\text{mol pNF g}^{-1}$ d.m. $\text{h}^{-1} \pm$ SD)
2010	Stagnic luvisol	Oberkulmer Rotkorn	4.89 e ± 0.07	58.6 cd ± 11.9	100.0 c ± 4.8
		Franckenkorn	4.54 d ± 0.05	87.7 e ± 23.1	110.4 d ± 6.1
		Schwabenkorn	4.12 c ± 0.13	40.2 bc ± 0.8	123.6 e ± 2.0
		Ostro	3.75 b ± 0.18	62.5 d ± 3.8	104.7 cd ± 4.2
	Haplic cambisol	Oberkulmer Rotkorn	3.64 ab ± 0.18	31.1 ab ± 1.8	40.1 ab ± 0.1
		Franckenkorn	3.49 ab ± 0.06	15.2 a ± 3.0	37.5 a ± 1.9
		Schwabenkorn	4.81 de ± 0.28	13.1 a ± 1.1	42.7 ab ± 0.2
		Ostro	3.33 a ± 0.03	9.6 a ± 1.1	47.7 b ± 2.6
2013	Stagnic luvisol	Oberkulmer Rotkorn	6.57 d ± 0.17	60.4 e ± 3.8	157.0 d ± 11.9
		Franckenkorn	5.54 c ± 0.04	27.3 c ± 1.2	127.9 c ± 1.9
		Schwabenkorn	5.55 c ± 0.21	34.9 d ± 3.4	137.7 c ± 2.6
		Ostro	6.75 d ± 0.32	75.3 f ± 2.8	139.9 c ± 7.3
	Haplic cambisol	Oberkulmer Rotkorn	4.61 a ± 0.01	2.2 ab ± 0.2	58.8 a ± 3.9
		Franckenkorn	4.99 ab ± 0.15	5.0 ab ± 0.6	67.4 ab ± 4.9
		Schwabenkorn	5.16 bc ± 0.11	1.6 a ± 0.1	72.3 b ± 2.6
		Ostro	4.64 a ± 0.16	7.0 b ± 0.6	65.4 ab ± 2.9
2007 (before starting the experiment)	Stagnic luvisol	—	3.95 ± 0.05	7.8 ± 1.1	28.5 ± 1.3
	Haplic cambisol	—	4.53 ± 0.10	11.3 ± 1.4	66.4 ± 2.9

Mean values in columns for a given experiment rotation (2010 or 2013) marked with the same letters are not significantly different according to Tukey's test at a significance level  $p \leq 0.05$ ; two-factor analysis (factor 1: variety, factor 2: soil type); SD—standard deviation.

The effect of the soil type on the enzyme activities was visible in both rotations of the experiments (years 2010 and 2013). The activities of dehydrogenases and arylsulfatase were always significantly higher in the SL than in the HC (Table 4, Figure 1). Catalase activity was also significantly higher in the SL than in the HC or no statistical differences were recorded. Due to the strong influence of the soil type (further in this study), the activities of the analyzed enzymes were described separately for each type of soil.



**Figure 1.** Activities of catalase, dehydrogenases and arylsulfatase in soil: (a–c) Means for soil types; (d–f) means for spelt variety; SD—standard deviation.

In the initial 2007 soil sample of the SL, catalase activity was  $3.95 \mu\text{mol H}_2\text{O}_2 \text{ g}^{-1} \text{ d.m. min}^{-1}$  (Table 4). After 3 years of the experiment, in 2010, this activity had increased on average to  $4.33 \mu\text{mol H}_2\text{O}_2 \text{ g}^{-1} \text{ d.m. min}^{-1}$  and in 2013, it had increased to an average  $6.10 \mu\text{mol H}_2\text{O}_2 \text{ g}^{-1} \text{ d.m. min}^{-1}$  (i.e., 54% more than in 2007). In 2010, the highest catalase activity was recorded in the SL soil where Oberkulmer Rotkorn was cultivated, followed by soils used in the cultivation of Franckenkorn, Schwabenkorn and Ostro. The differences of the catalase activity in SL resulting from spelt variety were statistically significant. In 2013, the differences resulting from the cultivation of different varieties of spelt were smaller. Significantly higher values were recorded after the cultivation of Ostro and Oberkulmer Rotkorn than in the case of Franckenkorn and Schwabenkorn.

In the initial 2007 HC samples, catalase activity was  $4.53 \mu\text{mol H}_2\text{O}_2 \text{ g}^{-1} \text{ d.m. min}^{-1}$ . After 3 years of the experiment, in 2010, it had decreased on average to  $3.82 \mu\text{mol H}_2\text{O}_2 \text{ g}^{-1} \text{ d.m. min}^{-1}$  and in 2013, it had again risen to an average  $4.85 \mu\text{mol H}_2\text{O}_2 \text{ g}^{-1} \text{ d.m. min}^{-1}$ —a 27% increase from 2010 and approximately a 7% increase from 2007. Significantly the highest catalase activity (especially in 2010) was recorded in the HC soil samples used for cultivation of Schwabenkorn spelt.

In the initial 2007 soil samples of the SL, dehydrogenase activity was  $7.8 \text{ nmol TPF g}^{-1} \text{ d.m. 24 h}^{-1}$  (Table 4). After 3 years of the experiment, in 2010, average dehydrogenase activity was  $62.3 \text{ nmol TPF g}^{-1} \text{ d.m. 24 h}^{-1}$  and in 2013 it was  $49.5 \text{ nmol TPF g}^{-1} \text{ d.m. 24 h}^{-1}$ , i.e., 6 times higher than in 2007. In 2010, the significantly the highest value of the dehydrogenase activity was recorded in the soil used for growing Franckenkorn spelt. In 2013, significantly the highest value was recorded after cultivation of Ostro spelt, then Oberkulmer Rotkorn, Schwabenkorn and Franckenkorn variety (the

differences of the dehydrogenase activity in SL resulting from spelt variety were statistically significant).

In the initial 2007 soil samples of the HC, dehydrogenase activity was 11.3 nmol TPF g<sup>-1</sup> d.m. 24 h<sup>-1</sup>. After 3 years of the experiment, in 2010, it was on average 17.3 nmol TPF g<sup>-1</sup> d.m. 24 h<sup>-1</sup>, and in 2013 it had decreased by 77% to 4.0 nmol TPF g<sup>-1</sup> d.m. 24 h<sup>-1</sup>. In 2010, no statistically significant effect of spelt variety on dehydrogenase activity was recorded, while in 2013 it was for Ostro (as a rule, however, the activity did not depend on the spelt variety). Neither in 2010 nor in 2013 was there a clear effect of the variety on dehydrogenase activity of HC.

In the initial 2007 in SL samples, arylsulfatase activity was 28.5 μmol pNF g<sup>-1</sup> d.m. h<sup>-1</sup> (Table 4). After the first rotation of the experiment, in 2010, the activity increased to 109.7 μmol pNF g<sup>-1</sup> d.m. h<sup>-1</sup> and in 2013, following the next rotation, it was 140.6 μmol pNF g<sup>-1</sup> d.m. h<sup>-1</sup>, almost 5 times higher than in 2007. In 2010, statistically significantly the highest arylsulfatase activity was recorded in the soil used for cultivation of Schwabenkorn spelt and in 2013– in the soil used for cultivation of Oberkulmer Rotkorn spelt.

The initial level of arylsulfatase activity in the HC soil was 66.4 μmol pNF g<sup>-1</sup> d.m. h<sup>-1</sup>. After 3 years, in 2010, arylsulfatase activity dropped to an average 42.0 μmol pNF g<sup>-1</sup> d.m. h<sup>-1</sup>, and in 2013, it had increased back to the initial level of 66.0 μmol pNF g<sup>-1</sup> d.m. h<sup>-1</sup>. The impact of spelt variety on arylsulfatase activity (and on the activity of dehydrogenases) in the HC soil was not significant.

Dehydrogenases activity was most positively correlated with the content of total sulfur and total nitrogen in the soil, while arylsulfatase activity was correlated with sulfate sulfur and total nitrogen in the soil (Table 5).

**Table 5.** Values of correlation coefficient between soil properties.

	Catalase Activity	Dehydrogenase Activity	Arylsulfatase Activity	H <sup>+</sup>	C <sub>total</sub>	N <sub>total</sub>	S <sub>total</sub>
Dehydrogenase act.	0.316						
Arylsulfatase act.	0.701 **	0.687 **					
H <sup>+</sup>	0.629 **	0.735 **	0.835 ***				
C <sub>total</sub>	0.199	0.613 *	0.620 *	0.425			
N <sub>total</sub>	0.470	0.736 **	0.864 ***	0.707 **	0.898 ***		
S <sub>total</sub>	0.104	0.758 ***	0.575 *	0.609 *	0.737 **	0.778 ***	
S–SO <sub>4</sub>	0.595 *	0.610 *	0.714 **	0.526 *	0.618 *	0.672 **	0.472

\*  $p < 0.05$ , \*\*  $p < 0.01$  and \*\*\*  $p < 0.001$ ;  $n = 16$ .

#### 4. Discussion

Soil physical and chemical properties can depend not only on the species, but also on the variety of the crop [20]. Selection of varieties is extremely important due to the differences in genotypic features of individual varieties. The varieties used in this research differed morphologically (height, length of the root system, yield structure components). The length of the root system as well as the mass and composition of postharvest residues determined the physical (water–air conditions) and chemical (particularly organic carbon content) properties of the soil, which may have considerably diversified the conditions of development of soil microorganisms.

Catalase is an enzyme responsible for the decomposition of hydrogen dioxide (a compound with strong oxidizing properties). Borowska et al. [27] found that in soils fertilized with manure and mineral fertilizers for winter wheat cultivation, catalase activity ranged from 3- to 30-μmol H<sub>2</sub>O<sub>2</sub> g<sup>-1</sup> d.m. min<sup>-1</sup>. The highest activity was recorded in the soil fertilized with manure stored under anaerobic conditions and mineral nitrogen, and the lowest in the soil fertilized with manure stored under aerobic conditions. The levels recorded in this experiment ranged between 3.33- and 6.75-μmol H<sub>2</sub>O<sub>2</sub> g<sup>-1</sup> d.m. min<sup>-1</sup>. The difference in catalase activity may be due to the different states of aeration of the soil. This is a consequence of belonging to different complexes of agricultural suitability, and to different decomposition rates of organic compounds present in the soil [28]. Kucharski et al. [29]

showed that the number and activity of microorganisms was associated more with the soil suitability complex than with the fertilizer dose, whereas high doses of mineral nitrogen changed the composition of soil microflora and reduced enzymatic activity. Joško et al. [30] and Nakatani et al. [31] also showed that not only the kind of soil additives and herbicides, but also soil properties and cultivar type influence microbial parameters. In this study, spelt was grown in an organic system, hence with a greater mass of crop residues and lack of mineral nitrogen fertilization than in conventional system.

Dehydrogenase activity is a good indicator of soil biologic activity, and more precisely of microbial aerobic metabolism, mainly bacteria and actinomycetes [32]. Determination of the activities of these enzymes in the soil is commonly used to assess factors adversely affecting soil microorganisms. It has been shown that a change in the oxygenation of the soil significantly alters dehydrogenase activity [33]. Literature data indicate that the level of soil enzyme activities is also closely related to soil organic matter content [34]. The proliferation of microorganisms and dehydrogenase activity may be stimulated by a supply of fresh organic matter from crop residues or the forecrop (a mixture of cereals and legumes in this study). In this study, the increase in dehydrogenase activity in both suitability complexes compared to the initial 2007 samples, and especially in 2010, may have been due to an increase in carbon content and total nitrogen in the SL soil and a reduction in soil acidity in the HC soil. These results indicate that organic cultivation of spelt is beneficial for soil microorganisms.

Many authors report that the conventional production system leads to a deterioration of biologic and physicochemical properties of the soil, while organic cultivation of plants counteracts these adverse changes [35,36]. This finding was partly confirmed in this study. An increase in soil dehydrogenase activity in organic cultivation of wheat was also shown by Fraç et al. [37], indicating that this may be the result of an increased supply of organic matter and a pH more favorable for development of microbes. Roberto et al. [38] also showed that organic matter increased the soil enzymatic activity.

Sulfur content in most arable soils in Poland is low, insufficient for plant growth [39]. Such levels were also recorded in the soils used in this experiment. Sulfur deficiency in soil—especially sulfate available to plants—reduces biomass yield and deteriorates its biologic value [40–44]. Sulfur cycle in the environment occurs through chemical processes, with the central role played by biochemical processes. Arylsulfatase hydrolyzes organic compounds (aromatic sulfate esters (R–O–SO<sub>3</sub>)) to phenols (R–OH) and inorganic sulfates (SO<sub>4</sub><sup>2-</sup>), which are available to plants [45]. The arylsulfatase activity in the SL soil was almost twice higher compared to that in the HC soil.

In the authors' own research, positive correlation coefficients were observed between the concentrations of hydrogen ions in the soil and the activities of all three enzymes. Such a relationship was also observed by Filipek-Mazur et al. [46] for arylsulfatase activity, but not for dehydrogenase or catalase activities. Liang et al. [47] found no negative impact of long-term use of soil-acidifying mineral fertilizers on the activity of dehydrogenases. In the authors' own research, soil acidification was high, that is why it may have not led to a decrease in enzyme activity. The considerable mass of postharvest residues (which constitutes a source of carbon and energy) may also have been beneficial for the activity of microorganisms.

## 5. Conclusions

For all the examined spelt varieties (Oberkulmer Rotkorn, Franckenkorn, Schwabenkorn, Ostro) and for both soil types (stagnic luvisol and haplic cambisol), the activities of catalase and arylsulfatase in the soil after six years of organic cultivation were higher than after three years, showing a beneficial effect of organic cultivation on soil biologic properties (the considerable mass of postharvest residues that enter the soil under such cultivation may have been beneficial for the activity of microorganisms).

The enzymatic activity in the soil depended on the type of soil. As a rule, the activity was higher in a stagnic luvisol than in a haplic cambisol. The soil enzymatic activity depended also on the spelt variety, although no variety had a clear beneficial effect on the activities of all enzymes. The obtained results point to the need to expand research in this field.

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