

Original Research

Soil Phosphatase Activity and Phosphorus Content as Influenced by Catch Crops Cultivated as Green Manure

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Abstract

In a 3-year experiment, the effect of catch crop management [catch crop incorporated in autumn (A) or mulched (B) vs plots without a catch crop (C)] on soil acid (P_{AC}) and alkaline (P_{AL}) phosphatase activities as well as on the available phosphorus content (P_{AVAIL}) were investigated on typical *Alfisol*. The catch crops were sown at the beginning of August and ploughed in the autumn in 2008, 2009, and 2010, or left as mulch during the winter. Soil samples were taken four times a year from spring barley plots that were grown in 2009, 2010, and 2011. Generally, catch crop treatment significantly influenced the enzymatic activity as compared to the control. The P_{AC} activity was significantly greater with a mulched catch crop than in a ploughed one only in I and II sampling dates, whereas the P_{AL} activity was not influenced by the method and time of field pea management. The time of sampling significantly influenced the P_{AL} activity in 2011 and the P_{AC} activity in 2009 and 2011.

Keywords: catch crops, mulching, direct incorporation, phosphatase activity, available P content

Introduction

In the early 1990s, significant changes were implemented in the Polish agrarian structure. The changes consisted of growth in the share of cereal area while the area for other plants (root plants, papilionaceous, crops for green forage, etc.) decreased significantly over a 20-year period. At present in Polish agriculture, the percentage of cereals in the crop structure amounts to 73.8%, while plants having a high coefficient of soil organic matter reproduction (papilionaceous and grasses) occupy only an area of 7.5%. At the same time, a significant decrease in average livestock density has occurred. The number of livestock units (LUs) decreased from 65 LUs in 1990 to 45 LUs in 2000 [1].

As a result, the production and application of farmyard manure in Poland is too low [2]. Consequently, the successive depletion of soil with organic matter and a decrease in soil fertility and productivity has occurred. In order to limit the negative effects of simplified crop rotation and the excessive participation of cereals in the crop structure, which make the physicochemical and biological properties of soil worse, other sources of organic matter must be utilized. One of the management techniques that has been implemented for this purpose is catch crop treatment and the use of their biomass as green manure. Catch crops make it possible to use fertilizer compounds that were not utilized by the previous crop and thus limits their leaching into the ground water [3, 4]. Moreover, they protect the soil against erosion between the two main crops when incorporated, supply the soil with appreciable amounts of easily decomposable organic matter and therefore simultaneously

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improve soil properties. Legume crops are additionally valued due to their enrichment of soil with symbiotic nitrogen from the atmosphere [5]. Different forms of green manure, both fresh and processed, are usually applied. Mulching with fresh plant biomass, plant-derived biogas slurry, and compost have been tested as alternatives to the direct incorporation of the catch crop mass [6, 7]. Moreover, the effect of the time of incorporation has also been studied, indicating that incorporation time can be an important management tool [4]. Some studies have simply compared autumn incorporation to spring incorporation [8, 9], while in other experiments two or more incorporation dates have been compared in different seasons [e.g. 10, 11].

Some researchers have investigated the positive effect of the incorporation of catch crops for green manure on soil enzyme activity [e.g. 7, 12]. Soil enzymes may respond to changes in soil management more quickly than other soil variables and might be useful as early indicators of biological changes thereafter [13]. Soil enzymes play an essential role in the catalyzing reactions that are associated with organic matter decomposition and nutrient cycling [14]. Extracellular phosphomonoesterases (orthophosphoric monoester phosphohydrolases) are important enzymes that are involved in the P cycle of soil. These enzymes are of great agronomic significance because they hydrolyze compounds of organic P and transform them into inorganic P forms, which can be assimilated by plants and microorganisms [15]. The optimum pH of the activity of phosphomonoesterases are classified as acid (E.C. 3.1.3.2) with the optimum to be within a range of 4 to 6.5 and alkaline (E.C. 3.1.3.1) with an optimum pH of 8.5-11.0 [16, 17]. Acid phosphatase is produced in soil by bacteria and fungi, as well as in the form of plant-root exudates [18]. Alkaline phosphomonoesterase has not been detected in plants [17] and is mainly produced by soil bacteria and soil microorganisms other than arbuscular mycorrhizal fungi [19].

The aims of our study were to:

- (1) evaluate the activity of soil phosphatases and phosphorus content as influenced by the method and time of catch crop management, i.e. direct incorporation of a fresh catch crop into the soil and mulch left unincorporated during the winter until the following spring
- (2) to establish the relationship between phosphatase activities and soil chemical properties.

Material and Methods

Site Description and Experimental Design

The effect of catch crop management treatment (ploughed in the autumn or mulched during the winter) on soil acid and alkaline phosphatase activities (P_{AC} and P_{AL}) and the available phosphorus (P_{AVAIL}) content was investigated in a 3-year experiment. A one-factor, randomized block designed experiment was carried out at the Experimental Station in Mochełek near Bydgoszcz (17° 51' E; 53° 13' N) in Midwestern Poland. The soil of the field experiment was a typical *Alfisol* with a sandy loam texture [20].

The first factor of the experiment was catch crop (*Pisum sativum* L.) management, and therefore field pea green mass was ploughed in the autumn (A) or was left as a mulch during the winter (B). The control soil (C) was tilled without catch crop treatment. The time of soil sampling was the second experimental factor. Soil samples for enzymatic activity were collected four times a year under spring barley, before sowing (I date), during the spreading phase (II date), shooting (III date) and after harvesting (IV date). The chemical properties were determined twice a year, before the sowing of spring barley (March) and after it was harvested (August).

The previous crop that had been grown as a catch crop was winter wheat (*Triticum aestivum* L.). After the harvesting of winter wheat, the catch crops were sown at the beginning of August and harvested between 15 October and 3 November (2008-10). Three cycles of field experiments were conducted. Each cycle was performed in a different part of the experimental field. Every year after the harvesting of the catch crop, the soil in plots A and C was ploughed (at a depth of 27 cm). The green biomass of catch crops (mulch) in plots B was left on the soil surface through the winter. It was cut up and mixed with soil (at a depth of 10-12 cm) using a disc harrow the following spring. Spring barley (*Hordeum vulgare* L.) was sown between 2 and 8 April of the following years (2009-11). Phosphorus and potassium were applied in the spring at doses of 26.2 kg P·ha⁻¹ and 66.4 kg K·ha⁻¹, respectively. Nitrogen fertilization (90 kg N·ha⁻¹) was applied in two doses: 45 kg N·ha⁻¹ was applied before sowing spring barley (together with K and P fertilization) and 45 kg N·ha⁻¹ was applied during the shooting of spring barley. The same management procedures were repeated each year during the entire study period (2009-11).

The site is characterized by a moderate climate with an average annual temperature over the study period of about 12.3°C and an average annual rainfall of 432 mm (2009-11).

Soil Analysis

Soil samples were analyzed for chemical properties after air-drying at room temperature and sieving (< 2 mm). The chemical properties were assayed according to standard methods and each sample was analyzed in triplicate [21]. The total nitrogen (N_{TOT}) in the soil was determined using the Kjeldahl method. The soil organic carbon (C_{ORG}) content was determined using the dichromate oxidation procedure, while soil pH (1 M KCl) was measured using the potentiometric method in 1:2.5 soil:solution suspensions. The available phosphorus (P_{AVAIL}) was determined using the vanadium-molybdenum method.

The enzyme activities were performed on fresh, moist sieved soils of 1 g oven-dry equivalent. Field moist samples were sieved (< 2 mm) and stored at 4°C in a plastic box for not less than 2 days in order to stabilize microbial activity and then were analyzed for enzymatic activity within two weeks. In order to determine the acid (EC 3.1.3.2) and alkaline (EC 3.1.3.1.) phosphatase activities, an equivalent of 1 g dry soil was incubated for 1 h in a

Table 1. Chemical properties in studied soil.

Years	N_{TOT} ($g \cdot kg^{-1}$)		C_{ORG} ($g \cdot kg^{-1}$)		pH_{KCl}	
	Mean (\pm SD)	Range	Mean (\pm SD)	Range	Mean (\pm SD)	Range
2009	0.56 (\pm 0.05)	0.47-0.68	6.97 (\pm 0.91)	5.32-8.14	5.39 (\pm 0.71)	4.53-6.64
2010	0.64 (\pm 0.03)	0.59-0.71	8.02 (\pm 0.59)	6.17-8.79	4.79 (\pm 0.63)	4.32-6.65
2011	1.04 (\pm 0.08)	0.89-1.20	11.58 (\pm 0.59)	10.35-12.99	6.76 (\pm 0.22)	6.36-7.08

C_{ORG} – organic carbon; N_{TOT} – total nitrogen, pH_{KCl} – soil pH in 1M KCl

Table 2. Soil available phosphorus content ($mg \cdot kg^{-1}$) as influenced by the method and time of catch crop management (mean \pm SD).

Catch crop	2009		2010		2011	
	March	August	March	August	March	August
A*	88.2 (\pm 9.24)	76.9 (\pm 8.69)	72.0 (\pm 17.0)	102.4 (\pm 18.5)	102.0 (\pm 3.58)	107.2 (\pm 9.39)
B	86.2 (\pm 7.57)	87.6 (\pm 5.25)	77.4 (\pm 3.72)	108.6 (\pm 7.05)	104.0 (\pm 9.61)	109.8 (\pm 4.88)
C	88.2 (\pm 4.20)	92.4 (\pm 10.7)	69.3 (\pm 7.42)	91.0 (\pm 8.23)	96.3 (\pm 6.66)	110.3 (\pm 2.42)
Mean	87.5	85.6	72.9	100.7	100.8	109.1

*A – catch crop incorporated in autumn, B – mulched catch crop, C – control (without a catch crop)

rotating water bath at 37°C together with 4 ml of modified universal buffer (MUB) (pH 6.5 for the acid phosphatase and 11.0 for the alkaline one) and 1 ml of 0.115 M *p*-nitrophenylphosphate solution [22]. After incubation, 1 ml of 0.5 M $CaCl_2$ and 4 ml of 0.5 M NaOH were added to the samples followed by filtration. The concentration of *p*-nitrophenylphosphate was determined photometrically at 400 nm (Perkin Elmer Lambda 25 UV/VIS). The enzyme activity values were calculated on the basis of the oven-dry (105°C) weight of soil.

Statistical Analysis

Two-way analyses of variance was performed to examine the effect of catch crop management (direct incorporation in autumn and mulching) and the dates of sampling (seasonal variation) on the soil acid and alkaline phosphatase activities and the available phosphorus content. When significant treatment effects were found, Tukey's test was used to compare the treatment means. Means were considered significantly different at $P < 0.05$. The relations between the enzymatic activity and the chemical parameters (P_{AVAIL} , C_{ORG} , N_{TOT} , pH_{KCl}) were estimated using correlation analysis based on Pearson's correlation coefficients ($P < 0.05$, $P < 0.01$, $P < 0.001$). Statistical analyses were carried out using Statistica 8.1 for Windows software.

Results

Chemical Properties

Generally, the catch crop management did not significantly influence the chemical properties ($P < 0.05$).

Additionally, since the values of the properties studied were similar in March and August, the mean and range values for both sampling dates are presented in Table 1. The total N content ranged from 0.47 to 1.20 $g \cdot kg^{-1}$, with a mean value of 0.75 $g \cdot kg^{-1}$, while C_{ORG} ranged from 5.32 to 12.99 $g \cdot kg^{-1}$ with a mean value of 8.86 $g \cdot kg^{-1}$ when the whole experimental period was examined. The range of pH_{KCl} in 2009 and 2010 was similar (4.32-6.65), while in 2011 the range was higher and amounted to 6.36-7.08. The differences in soil pH between particular years were high because every year the experiment was carried out on different parts of the experimental field. The available P content was comparable between March and August in 2009 and 2011, while in March 2010 the available-P was on average 38% lower than in August. According to Polish Norm PN-R-04023, a very high content of available-P (over 89 $mg \cdot kg^{-1}$) was noted in August 2010 and on both sampling dates in 2011, while a high content (between 67 and 88 $mg \cdot kg^{-1}$) was generally observed on both sampling dates in 2009 and in March 2010 (Table 2).

Acid and Alkaline Phosphatase Activities

Enzymatic activity is presented as a function of catch crop management and seasonal variation for the individual years of the study (Tables 3-5, Fig. 1 a, b). Catch crop treatments significantly stimulated enzymatic activity with the exception of the P_{AL} in 2009 ($P < 0.001$ and $P < 0.01$). The time of sampling significantly influenced the P_{AL} activity in 2011 and the F_{AC} activity in 2009 and 2011 (Table 5). No significant effects of catch crop treatment and seasonal variation interactions were observed for either enzyme (Table 5). The results of enzymatic activity are additionally shown as the average for sampling dates

Table 3. Acid phosphatase activity (mM pNP·kg⁻¹·h⁻¹) as dependent on catch crop management and sampling dates.

Catch crop management	*I	II	III	IV
2009				
A	0.870aB#	1.302 b^A	1.333 abA	1.427 aA
B	0.947aB	1.590 aA	1.445 aA	1.525 aA
C	0.880aB	1.213 bA	1.177 bA	1.203 bA
Mean	0.899	1.368	1.318	1.385
2010				
A	1.690abA	1.785bA	1.710abA	1.723abA
B	1.860aA	1.940aA	1.825aA	1.855aA
C	1.450bA	1.623cA	1.505bA	1.493bA
Mean	1.667	1.783	1.68	1.69
2011				
A	1.350 bB	1.737 aA	1.765 aA	1.417aB
B	1.478 aB	1.842 aA	1.680 aAB	1.335aB
C	1.203 cB	1.555 bA	1.328 bB	1.307aB
Mean	1.343	1.712	1.591	1.353
2009-11				
A	1.3 bB	1.608 bA	1.603 aA	1.522 aA
B	1.428 aC	1.791 aA	1.650 aAB	1.572 aB
C	1.18 cB	1.464 cA	1.337 bA	1.334 bA
Mean	1.3	1.621	1.53	1.476

A – catch crop incorporated in the autumn, B – mulched catch crop, C – control (without a catch crop);

*sampling dates: I – before sowing of spring barley, II – during the period of barley tillering, III – during the period of barley shooting, IV – after harvesting of barley.

Values followed by the same low-case letter within each column are not significantly different at $P < 0.05$. Values followed by the same capital letter within a line are not significantly different at $P < 0.05$.

^ Different small letters indicate comparison between catch crops (within the same sampling date).

Different capital letters indicate a comparison among sampling dates within the same catch crop.

(I-IV) in order to make the data clearer and to easily determine whether there was any general tendency in the enzymatic activities as influenced by catch crop treatments (Fig. 1 a, b).

Generally, the acid phosphatase activity was significantly higher when the catch crop was applied to the soil as compared to the control (Tables 3, 5, Fig. 1a). The exception was the activity of the enzyme noted on the first date of sampling in 2009 and the last one (IV) in 2011. Mulching with field pea positively influenced the P_{AC} activity compared to the activity when catch crop was ploughed in the autumn. When the entire experimental period was considered (2009-11), P_{AC} activity was significantly higher in the mulched plots compared to the ploughed catch crop in sampling periods I and II. In the samples taken at sampling times III and IV, there was no significant influence of catch crop management on P_{AC} activity. The soil P_{AC} activity showed seasonal variability

but there was not a clear trend for these changes. In 2009 the P_{CA} activity on sampling date I was clearly reduced (mean of 0.899 mM pNP·kg⁻¹·h⁻¹) as compared with the data obtained at the II-IV sampling times (means between 1.318 and 1.385 mM pNP·kg⁻¹·h⁻¹). The P_{AC} activity in 2010 revealed similar values on all of the sampling dates with means ranging from 1.667 to 1.783 mM pNP·kg⁻¹·h⁻¹, and there was no statistical difference. In the last year of the experiment, the P_{AC} activity was the lowest before the spring barley was sown (1.343 mM pNP·kg⁻¹·h⁻¹) compared to the data obtained for further sampling periods (1.353-1.712 mM pNP·kg⁻¹·h⁻¹).

The soil alkaline phosphatase activity (Table 4, Fig. 1 b) was clearly higher in 2011 (1.199-1.552 mM pNP·kg⁻¹·h⁻¹) than in 2009 and 2010 (0.222-0.351 mM pNP·kg⁻¹·h⁻¹). Generally, the activity of P_{AL} was higher in the green manure treatment than in the control soil in 2010 and 2011, although these differences were not always statistically sig-

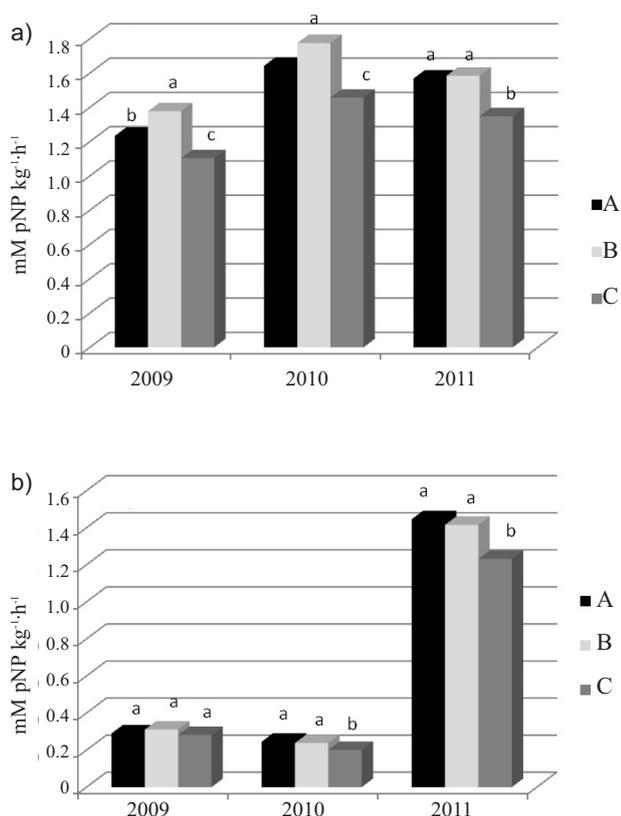


Fig. 1. The activity of (a) acid phosphatase, (b) alkaline phosphatase as dependent on the method and time of catch crop management; A – catch crop incorporated in the autumn, B – mulched catch crop, C – control (without a catch crop); Different small letters indicate comparison between catch crops (within the same year); Values followed by the same small letter are not significantly different at $P < 0.05$.

nificant. In 2009, however, there were no differences in the P_{AL} activity between catch crop management and the control or between the time and the method of field pea management. In contrast to P_{AC} , the alkaline phosphatase activity was not statistically differentiated by the time and the method of catch crop management. A seasonal variation of P_{AL} activity was observed only in 2011 and when the experimental times were considered together (2009-11). The P_{AL} activity was significantly higher before sowing and during the tillering of spring barley compared to the activity that was detected during the shooting and after the harvesting of spring barley.

Correlation between the Studied Properties

According to linear regression analysis, the soil alkaline phosphatase activity was significantly correlated with the chemical properties (Table 6), while the P_{AC} activity was only significantly correlated with pH_{KCl} in the mulched plots ($r = -0.573$). The highest coefficients of correlation were noted between P_{AL} and C_{ORG} , and N_{TOT} and pH_{KCl} ($P < 0.001$), while the P_{AVAIL} content was positively correlated with P_{AL} at $P < 0.05$ in the mulching and control plots.

Discussion

The application of green manure to the soil increases enzymatic activity because the added materials may stimulate the soil microorganisms that degrade organic matter through the production of diverse extracellular enzymes [23, 24]. In this study, the activity of soil phosphatases were generally higher under catch crop treatments than in the control soil, although these differences were not always statistically significant (Tables 3, 4). Similarly, in the study of Roldán et al. [25], higher values of acid phosphatase activity in soil were found with the addition of crop residue compared to the control.

Some research has shown that catch crop treatments may enhance P content in the soil [e.g. 26, 27]. Plants used as catch crops improve the P supply of the following main crops directly by increasing the plant available P amount in the soil or indirectly when used as green manures [28, 29]. Catch crops have been found to mobilize P in soil in different experiments. They substantially increased the P content in soil and soil solution in P poor soils and the P uptake of main crops was enhanced [30]. In a pot experiment, green fertilization with phacelia increased the P uptake of the following maize crop more than a mineral P fertilizer or manure [27]. In this study however, the P_{AVAIL} content was not significantly influenced by catch crop management and sampling time. This can be explained by the fact that P accumulated in the spring barley yield, which was significantly higher in the catch crop treatments compared to the control (data not presented). In turn, the differences in the P available between the years of investigation are shown (Table 2). The highest yield of catch crop biomass, amounting to 27.5 Mg·ha⁻¹, was noted in 2008, compared to 17.9 Mg·ha⁻¹ and 19.6 Mg·ha⁻¹ in 2009 and 2010, respectively (data not presented). This resulted in the highest yield of spring barley in 2009 and, as a result, caused a decrease in the soil P_{AVAIL} content, which probably accumulated in the yield of spring barley. Inversely, the higher P content in 2010 and 2011 could be associated with the lower yield of the catch crop in 2009 and 2010 and the lower yield of spring barley in 2010 and 2011. That explains why the influence of the catch crop on the available P content in the soil was not significant. Additionally, the possible effect of catch crops can be inferred by the relatively high P reserve in soil (Table 2) and the addition of P with fertilizers each year.

In this study the catch crop treatments, both mulching and direct incorporation in the autumn, tended to change the acid phosphatase activity, while alkaline phosphatase was not statistically differentiated. The changes in the soil enzyme activity are dependent on the chemical composition of the plants biomass applied as the catch crops and their C/N ratio, that was incorporated in the soil. This may be due to the fact that crop residue can modify the microbial population, which is the main source of enzyme activity in soil [31]. Differences in response to the phosphatase activity may be due to their different origins. The higher acid phosphatase activity in mulch treatment may be caused by the fact that the year-round crop cover in the mulch treat-

Table 4. Alkaline phosphatase activity (mM pNP·kg⁻¹·h⁻¹) as dependent on catch crop management and sampling dates.

Catch crop management	*I	II	III	IV
2009				
A	0.265aA	0.323aA	0.297aA	0.275aA
B	0.325aA	0.360aA	0.287aA	0.270aA
C	0.285aA	0.370aA	0.258aA	0.215aA
Mean	0.292	0.351	0.281	0.253
2010				
A	0.245aA	0.245aA	0.232aA	0.258aA
B	0.232aA	0.245aA	0.237aA	0.240aA
C	0.228aA	0.190bA	0.198bA	0.188bA
Mean	0.235	0.227	0.222	0.229
2011				
A	1.6aA	1.617aA	1.277aB	1.292aB
B	1.555aA	1.583abA	1.265aB	1.260aB
C	1.325bA	1.455bA	1.055bB	1.095aB
Mean	1.493	1.552	1.199	1.216
2009-11				
A	0.703 aA	0.728aA	0.602 aB	0.608 aB
B	0.704 aA	0.729aA	0.596 aB	0.590 aB
C	0.613 bA	0.672aA	0.504 bB	0.499 bB
Mean	0.673	0.71	0.567	0.566

For symbol explanations see Table 3.

ment might have promoted higher rates of enzyme activity since plants are an important source of P_{AC} activity in soil. Additionally, as stated by Haynes [32], continuous crop cover can enhance microbial biomass and enzyme activity as a result of increased organic C content in soil. Although the total C_{ORG} content was not significantly changed by catch crop management in this study, the quality of available fractions may have changed and influence soil enzymatic activity. Contradictory results were obtained by Elfstrand et al. [7], who showed that soil enzyme activities (acid phosphatase, arylsulphatase, protease) responded similarly to fresh green manure that was directly incorporated or applied as a mulch, although the highest enzyme activities were observed in the direct incorporation treatments. As stated by the authors, a larger surface area for microorganisms to attack and a more favorable environment for decomposition might have been beneficial for microorganisms in direct incorporation compared with the mulch treatment.

Both direct incorporation in the autumn and mulching of the catch crop promoted a higher P_{AC} activity until the last sampling after the harvest of spring barley (except in 2011), which indicates that fresh field pea was able to promote the long-lasting stimulation of soil phosphatases.

The seasonal changes in enzymatic activity can be explained by the intensive rhizosphere microbial population that accompanies the rapid development of the crop root system during the growth period [33]. The release of nutrients into the soil from the biomass of catch crops could account for the continuing increase or maintenance of enzymatic activity at the same level during the season [4], which could also have been the case for the phosphatase activity in this study. The lower acid phosphatase activity in the treatment with the catch crop incorporated in autumn than in spring could be related to a deeper and earlier mixing of biomass with soil. The autumn incorporation of the catch crop could contribute to the increase of the rate of the biomass mineralization. The study by Thorup-Kristensen and Dresbøll [11] showed that incorporation of the catch crop biomass to the soil in early autumn and large amount of precipitation in winter can result in a rapid mineralization and leaching nutrients released in this process from the soil before sowing of succeeding crops.

The differentiation of acid and alkaline phosphatase activities responses over the season can be explained by the fact that acid phosphatase activity can be of plant origin [17], and thus the crop might have contributed to the increased phosphatase activity that was observed during the

Table 5. Statistical differences (F-values and significance level) between means of variables by two-way ANOVA with factors catch crops management and sampling dates (2009-11).

Enzymes	Year	Catch crops (d.f. = 2)	Sampling dates (d.f. = 3)	Interactions: Catch crops management × Sampling dates (d.f. = 6)
P _{AL}	2009	0.770 NS	4.178 NS	0.621NS
	2010	0.652 **	0.095 NS	0.297 NS
	2011	8.939 ***	12.731 ***	1.166 NS
P _{AC}	2009	35.748 ***	81.050 ***	1.466 NS
	2010	38.528***	2.551 NS	0.188 NS
	2011	18.615***	25.105 ***	0.155 NS

P_{AL} – alkaline phosphatase activity; P_{AC} – acid phosphatase activity; NS, not significant; ** P < 0.01; *** P < 0.001; d.f., degree of freedom

intensive crop growth and at the end of the cropping season (II-IV dates). A high seasonal differentiation of acid phosphatase activity was confirmed by the study of Elfstrand et al. [7], who showed that the activity was lower in June (298 µg pNP·g⁻¹·h⁻¹) than in September (614 µg pNP·g⁻¹·h⁻¹).

When the entire experimental period was considered, the alkaline phosphatase activity was significantly and positively correlated with the C_{ORG}, N_{TOT}, and pH_{KCl} values

(Table 6). Soil organic carbon and nitrogen are the most important soil properties that can significantly influence the activities of soil enzymes since they reflect the level of organic matter [34]. That is why soil enzymatic activity is expected to be positively related to the soil organic matter. Catch crop treatment can enhance the soil microbial biomass content and activity by increasing the available organic matter content in soil. The other expected relationship was that between pH_{KCl} and phosphatase activities, since each enzyme shows a characteristic pH-dependent activity profile, an optimum pH for its maximum activity and specific stability, which is related to soil pH [35]. The soil pH values were in the range of near-optimal for the acid phosphatase, and that is why the enzyme did not correlate with this property. Alkaline phosphatase, however, functioned below the pH optimum for its activity and therefore reacted positively to changes of this feature, showing the huge activity in 2011 connected with the highest pH value (1.7 units) as compared with former years of the study. This close relationship was confirmed by high coefficients of correlation (Table 6).

Our study found a significant and positive, although low, correlation between the P_{AL} activity and the available P content (Table 6), while a lack of relationship between the P content and acid phosphatase activity was found. Generally, the relationship between the available P content and the phosphatase activity in soil is complex, and both positive and negative as well as no relationships can be observed between these properties. When no relationship is

Table 6. Correlation matrix between soil properties studied in experimental period (2009-11) (n = 24, means for years and sampling dates).

Catch crop	Properties	P _{AL}	pH _{KCl}	P _{AVAIL}	N _{TOT}	C _{ORG}
A	C _{ORG}	0.877	0.877	0.482	0.428	X
	N _{TOT}	0.892	0.892	0.633	X	***
	P _{AVAIL}	NS	NS	X	***	*
	pH _{KCl}	0.675	X	NS	**	*
	P _{AL}	X	**	NS	***	***
B	C _{ORG}	0.866	0.890	0.787	0.962	X
	N _{TOT}	0.880	0.927	0.775	X	***
	P _{AVAIL}	0.691	0.767	X	***	***
	pH _{KCl}	0.931	X	***	***	***
	P _{AL}	X	***	**	***	***
C	C _{ORG}	0.897	0.812	0.621	0.954	X
	N _{TOT}	0.906	0.837	0.654	X	***
	P _{AVAIL}	0.651	0.680	X	**	**
	pH _{KCl}	0.850	X	**	***	**
	P _{AL}	X	***	**	***	***

A – catch crop incorporated in the autumn, B – mulched catch crop, C – control (without a catch crop); C_{ORG} – organic carbon; N_{TOT} – total nitrogen; P_{AVAIL} – available phosphorus; P_{AL} – alkaline phosphatase; *P < 0.05, **P < 0.01, ***P < 0.001; NS – not significant

observed, P may not limit the studied system and some other factors may influence the production and activity of the enzyme [35]. A significant and positive relationship between phosphatase activity and P availability is obtained in soils that have not been fertilized and/or in those with a low abundance of nutrients, where a P deficiency occurs [36]. The uptake of available P by plants may additionally complicate this relationship.

Conclusions

Catch crops that are ploughed in the autumn or used as mulch significantly increased the soil phosphatase activities as compared to the control. Therefore, it can be concluded that catch crop treatments may be considered to be a good practice in helping to increase soil enzymatic activity, which is a measure of soil biological activity. As regards the method and time of the application of green manure, the acid phosphatase activity was more enhanced by mulching with field pea than by the indirect incorporation of the catch crop in the autumn. Alkaline phosphatase, however, was not significantly affected by catch crop management. The acid phosphatase activity was more sensitive to the method and time of catch crop management as compared to the alkaline form of the enzyme. That is why the acid phosphatase activity appeared in the specific soil studied to be a better indicator of the influence of catch crop treatment. It is reasonable to assume that in an alkaline soil alkaline phosphatase might be a better indicator. Additionally, it is important to highlight that soils with different origins and properties may show different phosphatases activity patterns.

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