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Selected extracellular soil enzymes as impacted by long-term applications of manure

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Introduction

Extracellular soil enzymes are involved in numerous biogeochemical processes impacting soil organic matter transformations and nutrient cycling. The present study evaluated selected soil enzymes activities related to N, P, and C mineralization as influenced by 10 yr of manure type, rates, and different timing applications on timothy crop at two sites with different soil textures. Carbon, N and P were also measured.

Methodology

- Long-term application of manure : 1995-2004.
- Semisolid (beef) manure, surface applied and not incorporated.
- Rates of manure: 0, 75, 150 and 300 total kg N ha^{-1}.
- Application times: spring, summer, early fall (August-September) and late fall (October-November)
- Two sites with different soil textures; i) silty clay loam (Dykeland site), and sandy loam (Upland site).
- Following soil enzymes measured using a fluorometric microplate assay :

➢ Beta-glucosidase (BG) and cellobihydrolase (CB), both are part of cellulase enzymes acting on B-1,4 linkages in cellulose chain.

➢ Phosphomonoesterase (phospho) which acts on low molecular weight of P compounds with monoester bonds.

➢ Leucine aminopeptidase (LAP) involved in the degradation of proteins.

➢ Phenol oxidase and peroxidase enzymes which are involved in breaking down lignin and other aromatic compounds as well as the production of humic substances. Spectroscopic microplate assay was used for these two enzymes.

- Incubation time and temperature was the same for all enzymes : 1.5 hours at 25 ° C except for LAP that was incubated for 24 hours at 4 ° C .
- Selected soil parameters analyzed following the standard procedures: total nitrogen (N) and soil carbon (SOC) by dry combustion and phosphorus by Mehlich-3 extraction ($P\text{-}M_3$).
- Mixed procedure of SAS was used to analyze the effects of the main factors.

Results and discussion

- Results are presented under table 1.
- Site effect was significant with Dykeland site showing higher values than Upland for phospho but the opposite trend was observed for LAP, phenol oxidase and peroxidase. Site effects was significant on SOC and $P\text{-}M_3$ with higher values associated with Upland site.
- Time effect was significant on phospho and phenol oxidase with higher values associated with late fall application for phospho activity.

Results

Results

Table 1. Effects of site, manure application times and rates on selected parameters

Source of variation		BG	CB	PHOSPHO	LAP	Peroxidase	Phenol oxidase	SOC	N	pH	$P\text{-}M_3$
		μmoles h^{-1} g^{-1}						g kg^{-1}			mg kg^{-1}
Site	Upland	49.6	14.5	110.4b	6.1a	470,300b	229,236	25.4a	2.2	6.3	166.4a
	Dykeland	49.7	14.3	128.3a	3.2b	874,841a	364,943	22.4b	2.2	6.2	80.0b
N Rate											
	0	41.0c	11.7d	110.2b	3.4c	758,430a	404,672	20.0c	1.8c	6.1cd	81.8d
	75	56.4a	17.3a	138.4a	5.0ab	647,764ab	307,616	23.6b	2.2b	6.2c	100.8c
	150	54.5a	15.7a	132.3a	5.8a	694,877ab	285,560	23.8b	2.3b	6.3b	126.4b
	300	46.7b	12.8c	96.5c	4.5bc	562,209c	190,508	28.2a	2.6a	6.5a	184.1a
Time											
	Spring	48.1	13.9	116.4b	4.6	734,607	352,990	22.3b	2.0 b	6.3a	116.3b
	Summer	51.9	14.8	120.9ab	4.2	654,938	266,856	23.7b	2.2 ab	6.3a	129.9a
	Early fall	48.9	13.8	114.5b	5.3	690,653	267,941	25.8a	2.3 a	6.3a	137.7a
	Late fall	49.7	15.2	125.5a	4.5	610,085	300,571	23.8b	2.3 a	6.1b	108.7b
ANOVA (p values)											
	Site	NS	NS	***	***	***	***	***	NS	NS	***
	Time	NS	NS	*	NS	NS	***	**	*	***	***
	Site*time	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS
	Rate	***	***	***	***	**	***	**	***	***	***
	Site*rate	NS	NS	*	NS	NS	*	NS	NS	***	NS
	Time*rate	NS	NS	NS	NS	NS	NS	NS	NS	NS	**
	Site*time*rate	NS	NS	NS	NS	NS	*	NS	NS	NS	NS
Contrasts											
	Linear	NS	NS	***	NS	***	***	**	***	***	***
	Quadratic	***	***	***	***	NS	NS	NS	NS	NS	NS
	Cubic	**	**	**	NS	NS	NS	NS	NS	NS	NS

NS, *, **, ***, Not significant at 0.05, 0.01 and 0.001 probability level, respectively. Values followed by different letters within a group are statistically different.

- Higher values of phenol oxidase were associated with spring application.
- Time effects was significant on C, N, pH and $P\text{-}M_3$. Early fall application resulted in increased SOC, whereas delayed application showed increased N content compared with spring application. Values of $P\text{-}M_3$ were lower with spring and fall application and higher with summer and early fall application, implying P losses during spring and fall.
- A positive quadratic trend for BG, CB and LAP, and a negative linear trend for peroxidase and phenol oxidase was observed in response to increasing manure rate. Similar trends were observed for C, N, pH and $P\text{-}M_3$ showing linear trends.
- Soil pH increased with manure application rate and with summer and early fall applications. The late fall application was associated with lower soil pH.
- Opposite trends between $P\text{-}M_3$ and phospho, and between SOC with peroxidase and phenol oxidase were observed implying that certain soil enzyme activities releasing specific nutrients is inversely correlated with their availabilities (Ref. 1). A previous study reported that land use leading to SOM loss increase oxidative activities (Ref. 2) and in this study, phenol oxidase and peroxidase were higher without manure application. Phospho activity was reported to be repressed by high inorganic P (Ref. 3). Conversely, LAP increased with increasing manure rate showing a quadratic trend, revealing that it was enhanced by substrate availability.
- Higher LAP activity, SOC, and $P\text{-}M_3$ at Upland site is attributed to its light texture favoring SOM mineralization.

Conclusion

- Manure application time and rate impacted the nutrient accumulation and associated enzymes.
- This study indicated that extracellular enzymes are good indicators of soil management.

References

- Ref.1: Applied Soil Ecology (2008): 38:128-136.
- Ref. 2: Soil Biology and Biochemistry(2010) 42:391-404.
- Ref. 3: Biol. Fertil. Soil (1991)12:189-194.

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