Agricultural Science; Vol. 2, No. 1; 2020
ISSN 2690-5396 E-ISSN 2690-4799
https://doi.org/10.30560/as.v2n1p109

Enzyme Activity Variability and Comparison in Soils under Medicinal versus Crop Plants of Anguo City, China

Wenke Liu[1]

[1] Institute of Environment and Sustainable Development in Agriculture, Chinese Academy of Agricultural Sciences, Key Lab of Energy Conservation and Waste Management of Agricultural Structures, Ministry of Agriculture, Beijing, China

Correspondence: Wenke Liu, Institute of Environment and Sustainable Development in Agriculture, Chinese Academy of Agricultural Sciences, Key Lab of Energy Conservation and Waste Management of Agricultural Structures, Ministry of Agriculture, Beijing, China. E-mail: liuwke@163.com

Received: March 25, 2020 Accepted: April 6, 2020 Online Published: April 16, 2020

Abstract

Long-term continuous cultivation of different plant species in a similar agroecosystem intensively may result in divergent variability in soil fertility, particularly soil biochemical properties. In this study, an investigation was conducted to clarify the variability of five soil enzyme activities (urease, protease, catalase, polyphenol oxidase and alkaline phosphatase) of croplands under medicinal plants (herbal fields) and food crops (crop fields) in Anguo city, a traditional cultivation base for Chinese medicinal plants in China. The results showed that five soil enzyme activities were similar between herbal and crop fields. However, soil urease and alkaline phosphatase activities of herbal and crop fields decreased significantly with soil depth (0-60 cm), while protease, catalase, polyphenol oxidase activities were similar in all soil layers for two kinds of fields. There were largely variation scenes at linear correlation analysis between soil physicochemical traits and enzymatic activities under medicinal plant versus crop fileds although extensively significant correlations were presented. In conclusion, soil enzyme activities were similar in two type of farmlands, and soil urease and alkaline phosphatase activities decreased with soil depth for both fields. Inconsistent linear correlations between soil physicochemical traits and enzymatic activities under medicinal plant versus crop fields were presented, so soil enzymatic activity variation was subjected to soil physicochemical traits dominated by agronomic managements designed for specific plant species.

Keywords: enzyme activity, herbal field, crop field, soil fertility, soil depth

1. Introduction

Soil enzymes are potential indicators of soil fertility because they did show us the status of soil biology and soil nutrient cycling in agroecosystems (Dick 1994; Dick et al.1996). They also indicate soil biochemical characteristics to understand the status and evolution of soil fertility and monitor the effects of soil management on long-term productivity (Doran and Parkin 1994; Zeng et al.2008). Nitrogen (N) and phosphorus (P) are essential macronutrients, in form of inorganic compounds and organic matter in soil. In soil, transformation and cycle of soil organic N and P are mainly catalyzed and driven by urease, protease and phosphatase. Soil urease and protease promote soil hydrolytic processes of urea and protein materials into inorganic N, respectively (Guan 1986). Phosphatase controls the mineralization of soil esters of P to produce inorganic phosphate (Speir and Ross 1978). In addition, there still some important soil enzymes involved in soil redox processes. Plant roots and microflora are sources of soil enzymes, dominating soil enzyme activities and the soil biology and biochemistry.

Soil enzyme activity responds rapidly to changes in soil management, and is affected by long-term agronomical management practices (Dick et al. 1996, Dick 1994), soil type, cropping system and so on. Anna (1999) had showed that soil enzyme activities were generally higher in continuous grass fields than cultivated fields, and added organic amendments increased soil enzyme activities in cultivated systems. Masciandaro (2004) showed that compost application increased agronomic yield due to release of nutrients for plant nutrition and soil metabolism. Also, long-term application of organic fertilizer was more conducive to conserve the soil biochemical characters and environment quality (Wang et al. 2007). Under long-term fertilization conditions, organic manure plus chemical fertilizers increased soil enzyme activities significantly, and the effect of fertilization on black soil was higher than dark brown soil (Jiao et al. 2011). Compared with soybean and maize rotation or intercropping, continuous cropping soybeans and maize reduced soil urease activity (Dai et al. 2013). Up to date, many factors that affect soil enzyme activity in different agroecosystems had been revealed, however, the difference of soil enzyme activity in the same area under different crop species has not been reported.

Anguo city locates in Taihang piedmont plain, belonging to Hebei province, China. Anguo city is famous for Chinese medicinal plant cultivation lasting more than 400 years. Nowadays, cropland and herbal land coexists together, and the percent of medicinal plant cultivation area exceeds 35% of the total arable land. In Anguo city, the distribution of herbal fields and crop fields is concentrated, cross and adjacent over many years. Currently, about 300 herbal plant species were cultivated on ten thousand hectares (Chu et al. 2010). So, it is meaningful to investigate soil fertility evolution for guiding agronomy management to realize the sustainable development of medicinal plants cultivation in Anguo city. However, previous reports were focused on status quo of soil nutrients (Li et al. 2011; Du et al. 2013), heavy metals (Chu et al. 2010) and mycorrhizal fungi (Zhao et al. 2010) issues of herb soil. No study has been carried out to investigate soil enzymatic activity of herb fields, meanwhile making a comparison with cropland field. Our earlier work found that soil nitrate and available P of medicinal crop fields were higher than neighboring crop fields, while soil pH of cropland were higher than medicinal crop fields in Anguo city (Du et al. 2013). Whereas, there is no information about soil enzyme activities and their differences of the fields growing medicinal crops and food crops in Anguo city. Within this context, the objectives of the study are a) to evaluate the spatial distribution characteristics of the five enzyme activities along three vertical soil layers in herbal fields and crop fields, and b) to compare the differences of five soil enzyme activities between herbal fields and crop fields.

2. Materials and Methods

2.1 Field Selection and Soil Sampling

The investigation was carried out during 19-21 May, 2012 in Anguo city. In order to evaluate enzyme (urease, protease, catalase(CAT), polyphenol oxidase(PPO) and alkaline phosphatase(ALP)) activity variability along 0-60 cm soil depth of herbal fields and their neighboring crop fields in Anguo city, forty herbal fields and twenty neighboring crop fields was selected in ten villages of four townships for soil sampling. Each sampling area is lesser and each field is about 0.01 ha, so the sampling number was chosen three. The three representative locations were selected in each field where soil sampling was conducted at three soil layers, i.e., 0-20, 20-40 and 40-60 cm soil depths. Soil samples from each location at same soil layer in one field were thoroughly mixed to obtain the representative samples. The sampling fields were growing twenty-one species of medicinal plants and two food crops including wheat and peanuts. Detail information about sampling fields and the crop species planted in the sampling fields are listed in Du et al. (2013).

2.2 Determination Methods

Soil samples were air-dried and passed through 2 mm sieve to test five enzyme activities including urease, protease, CAT, PPO and ALP. The enzyme activities were determined according to the methods described by Tabatabai and Bremner (1969) and Guan (1986). Soil urease activity was determined by phenol-sodium hypochlorite colorimetric method and was expressed as mg ammonia generated by per gram dry soil. Soil protease activity was measured using ninhydrin-colorimetric and was expressed as mg glycine equivalents per gram soil. ALP activity was determined using ammonium chloride-ammonium hydroxide (pH 9.8) as the buffer solution and disodium phenyl phosphate (0.5%, $w\cdot v^{-1}$) as the substrate and was expressed as mg of phenol equivalents per gram dry soil. Soil CAT activity was measured by titration method and was expressed as the amount of potassium permanganate solution (0.1 $mol\cdot L^{-1}$) that was consumed per gram dry soil. Soil PPO activity was measured by iodometric titration and was expressed by the amount of standard iodine (0.005 $mol\cdot L^{-1}$) that was used to titrate equivalent 1g soil for filtrate.

2.3 Data Analysis

Data processing and analysis were made with Excel 2007 and SAS 8.2; *t*-test was used to examine the difference between soil enzyme activities of various depths, and herbal fields and crop fields.

3. Results

3.1 Soil Enzyme Activities in Herbal and Crop Fields

As shown in Table 1, there were significant differences between soil urease and ALP activities in herbal fields between three soil layers, and their activities decreased with increment in soil depth remarkably. Urease and ALP activities in 0-20, 20-40 and 40-60 cm soil layers were 0.78-6.60, 0.48-5.22, and 0.26-2.66 $mg\cdot g^{-1}$, and 0.10-0.26, 0.06-0.18, and 0.05-0.13 $mg\cdot g^{-1}$, respectively. Urease and ALP activities of upper soil layers (0-20 cm or 40-60cm) were significantly higher than those of lower soil layers. Soil protease activities along with the increase of soil depth gradually reduce and the CAT and PPO activities increased with increasing soil depth, but there was no statistically significant difference of soil protease, CAT, PPO activities between three soil layers of herbal fields.

The same as herbal fields, there were significant differences between soil urease and ALP activities in crop fields between three soil layers, and they decreased with increment in soil depth remarkably (Table 1). Urease and ALP activities in 0-20, 20-40 and 40-60cm soil layers were 1.36-4.93, 0.77-3.50, and 0.39-2.68 $mg \cdot g^{-1}$, and 0.11-0.24, 0.07-0.13, and 0.05-0.12 $mg \cdot g^{-1}$, respectively. Urease and ALP activities of upper soil layers (0-20 cm or 40-60cm) were significantly higher than those of lower soil layers except ALP activities between soil layers of 20-40cm versus 40-60cm. Protease, CAT and PPO activities had the similar change trend, namely the enzyme activities of the surface layer (0-20cm) were highest, the bottom layer(20-40cm) were center ,and the middle layer (20-40cm)were slightly low, but there was no statistically significant difference of soil protease, CAT, PPO activities between three soil layers of crop fields.

Table 1. Soil urease, protease, CAT, PPO, and ALP activities at the three soil layers of herbal fields and crop fields

Soil enzymatic indices	Soil layers (cm)	Herbal fields mean ±SD	Crop fields mean±SD
	0-20	2.94±1.37**	3.01±3.01**
Urease（$mg \cdot g^{-1}$）	20-40	1.82±0.90**	1.65±0.86**
	40-60	1.07±0.59**	1.00±0.62**
	0-20	14.29±9.21	15.96±8.74
Protease（$mg \cdot g^{-1}$）	20-40	11.35±6.79	11.33±6.36
	40-60	10.76±5.90	11.45±9.31
	0-20	1.85±0.50	1.85±0.51
CAT （$ml \cdot g^{-1}$）	20-40	1.89±0.60	1.74±0.47
	40-60	1.92±0.67	1.78±0.52
	0-20	2.23±0.54	2.39±0.56
PPO（$ml \cdot g^{-1}$）	20-40	2.29±0.48	2.37±0.63
	40-60	2.38±0.62	2.42±0.85
	0-20	0.15±0.03**	0.15±0.03**
ALP（$mg \cdot g^{-1}$）	20-40	0.10±0.02**	0.10±0.02**
	40-60	0.09±0.02**	0.09±0.02

Notes: * and ** indicate significant difference for the enzymatic activity among three soil layers at P<0.05 and P<0.01, respectively.

3.2 Comparison of Soils Enzyme Activities of Herbal and Crop Fields

As shown in Table 2, soil urease activities of 0-20cm soil layers in herbal fields were slightly below the crop fields, and the enzyme activities of 20-40cm and 40-60cm were opposite. Protease activities in 0-20cm and 40-60cm soil layers of herbal fields were slightly below the crop fields in the same layers, and on the contrary of 20-40cm soil layers. CAT activities in 0-20cm soil layers of herbal and crop fields were same, and the activities of 20-40cm and 40-60cm soil layers in herbal fields showed larger values than crop fields. PPO activities of three soil layers in herbal fields were slightly lower than crop fields of corresponding soil layers. Soil ALP activities of three soil layers in herbal and crop fields were similar. But the difference between herbal and crop fields at the three soil layers was not significant due to large variability.

Table 2. Comparison of soil enzyme activities between herbal fields and crop fields

Soil enzymatic indices	Field types	Soil depth (cm)		
		0-20	20-40	40-60
Urease（$mg \cdot g^{-1}$）	Herbal field	2.94	1.82	1.07
	Crop field	3.01	1.65	1.00
Protease（$mg \cdot g^{-1}$）	Herbal field	14.29	11.35	10.76

	Crop field	15.96	11.33	11.45
CAT（ml·g^{-1}）	Herbal field	1.85	1.89	1.92
	Crop field	1.85	1.74	1.78
PPO（ml·g^{-1}）	Herbal field	2.23	2.29	2.38
	Crop field	2.39	2.37	2.42
ALP（mg·g^{-1}）	Herbal field	0.15	0.10	0.09
	Crop field	0.15	0.10	0.09

3.3 Correlation of Soil Enzyme Activities with Soil Physicochemical Properties in Herbal and Crop Fields

According to the preliminary analysis of soil physical and chemical properties, carried out on soil enzyme activities and soil physical & chemical properties in herbal and crop fields. The correlation coefficients were listed in Table 3. The table3 showed that soil urease activities of herbal fields were positively correlated with soil organic matter (SOM) and pH value, and the correlation of soil urease activities and alkali-hydrolysable nitrogen (A-H-N), available phosphorus (avail.-P), nitrate, water content were negative. Protease activities were positively correlated with A-H-N, avail.-P and water content, and negatively correlated with pH and Ec. ALP activity and avail.-P, nitrate, water content were positively correlated, negatively related to pH.CAT activities were positively correlated with A-H-N, nitrate, water content, and the correlation between CAT activities and SOM was power exponent, the formula was y1=0.3653x10.2153.The correlation of CAT activities and avail.-P was quadratic polynomial, the formula was y2=-4.763x22+14.34x2+12.065.There were no correlation between PPO activities and all soil physical properties. Soil urease activities in crop fields were negatively correlated with A-H-N and water content. Soil protease activities were positively related to avail.-P and nitrate, and were negatively related to A-H-N. ALP activities and avail.-P were positively related, and the correlation of ALP activities and water content was quadratic polynomial, the formula was y3 = 4.2382 x32- 0.9677x3 + 0.1921.CAT activities were positively correlated with A-H-N and water content. PPO activities were positively correlated with avail.-P and nitrate content, and the correlation of soil pH, EC , water content and SOM were quadratic polynomial, the formulas were y4=0.3334x42-1.5323x4+8.543, y5=33.987x52-148.76x5+278.72, and y6=0.0242x62-0.1148x6+0.2676, y7=-0.099x72+0.5571x7-0.1974, respectively. In the above seven formulas, y1,y2,y3,y4,y5,y6 and y7 represented SOM and avail.-P in herbal fields, avail.-P, pH, EC, water content and SOM, respectively. The x1and x2 represented CAT activities in herbal fields, x3 represented ALP activities in crop fields, x4,x5,x6,x7 represented CAT activities in crop fields.

Table 3. Correlative coefficients between soil enzyme activities and soil physicochemical properties of herbal fields and crop fields

Soil physicochemical properties	Soil enzyme activities of herbal fields (n=120)					Soil enzyme activities of crop fields (n=60)				
	urease	protease	ALP	CAT	PPO	urease	protease	ALP	CAT	PPO
O.M. content	0.29**	0.04	0.07	0.18*	0.03	0.02	0.18	0.20	0.01	0.32**
AHN content	0.32**	0.43**	0.14	0.27**	0.12	0.33**	0.30*	0.18	0.32**	0.07
Avail.-P content	0.33**	0.27**	0.57**	0.18*	0.09	0.001	0.43**	0.43**	0.10	0.28*
Nitrate content	0.24**	0.03	0.19**	0.19*	0.02	0.07	0.45**	0.09	0.14	0.27*
pH	0.18**	0.41**	0.18**	0.11	0.22	0.04	0.15	0.11	0.002	0.37**
EC	0.07	0.17*	0.02	0.0007	0.03	0.08	0.10	0.004	0.10	0.51**
Water content	0.34**	0.12	0.23**	0.31**	0.02	0.41**	0.08	0.25**	0.56**	0.44**

Note: * and ** indicate significant difference of the correlation between soil enzyme activities and soil physicochemical properties of herbal fields and crop fields at $P<0.05$ and $P<0.01$, respectively.

4. Discussion

The current results showed that soil urease and ALP activities decreased with soil depth both in herbal and crop fields. Taylor (2002) found that in sandy soils enzymatic activities decreased with depth, and positive correlations were found between enzyme activities and SOM content. Franken and Dick (1983) showed that ALP activities were significantly related to microbial respiration, while urease activities were not significantly correlated to microbial respiration, ALP activities were highly correlated with microbial biomass. Lan (2011) found that urease activities were positively related to soil organic carbon, nitrogen and avail-P. This is consistent with previous studies. In the preliminary study, Du et al. (2013) showed that soil nutrients and electrical conductivity (EC) of fields under medicinal plants decreased with soil depth. Agricultural activities are concentrated in the surface layer(0-20cm), and the oxygen content and fertilizers were concentrated in 0-20cm soil layers, so the number of rhizosphere microbial was large and more frequent activity, and increased with soil enzyme activities accordingly. With increasing soil depth, soil microbial quantity and SOM content decreased gradually, ultimately led to soil enzyme activities decreasing.

There was no significant difference in five soil enzyme activities between herbal fields and crop fields in various soil layers. The previous results showed that soil nutrients (nitrate, avail.-P contents), and soil chemical properties (EC and pH) differed largely between herbal fields and crop fields. However, no difference was found with respect to SOM and soil water content (Du et al. 2013). We suggest that the similar of SOM and microbial species due to long-time distribution caused no difference between enzyme activities under two kinds of utilization ways.

Both in herbal and crop fields, ALP activities were increased with the content of avail.-P. Soil ureas activities of herbal fields were increased with SOM content, however, in crop fields soil urease activities were not significant relative with SOM content. Fan (2002) showed that soil urease activities increased with soil organic carbon, and ALP activities closely related to the content of avail.-P. Their findings can be supported by our results. Annual organic additions generally stimulated the size of microbial biomass carbon and enzyme activity and may be more efficient in maintaining long-term soil productivity with repeated applications or in combination with inorganic fertilizers (Lalande 1998).

5. Conclusions

This study presents some novel findings on soil enzyme activity differences between herbal fields and crop fields under the long-term cultivation practices in Anguo city, China. First, soil enzyme activities were similar in the two type lands cultivated herbal plants and food crops, respectively. Secondly, the urease and ALP activities were sensitive to soil depth both for herbal and crop fields, though the variability of other three enzyme activities in soil depth was not significant. Moreover, soil enzymatic activity variation may subject to many soil physicochemical traits developed during cultivation of medicinal plants and crops during the long-term production.

Acknowledgements

I want to thank Jiaojiao Zhao, a master student, for her soil sampling and soil pretreatment.

References

Bandick, A. K., & Dick, R. P. (1999). Field management effects on soil enzyme activities. *Soil Biology & Biochemistry*, *31*(11), 1470-1479. https://doi.org/10.1016/S0038-0717(99)00051-6

Chu, Z. D., Liu, W. J., Xiao, Y. B., Zhu, Y. G., Zheng, W. J., & Duan, Y. H. (2010). Survey and assessment of heavy metals in soils and herbal medicines from chinese herbal medicine cultivated regions. *Environmental Science*, *31*(6),1600-1607.

Dai, J. J., Song, P. H., Yan, M. C., & Xu, T. T. (2013). Effect of different cropping patterns of soybean and maize seedlings on rhizosphere soil enzyme activities and microbial biomass carbon and nitrogen. *Journal of Northeast Agricultural University*, *44*(21), 17-22.

Dick, R. P. (1994). Soil enzyme activities as indicators of soil quality. In Doran, J. W., Coleman, D. C., Bezdicek, D. F., & Stewart, B. A. (Eds.), Defining soil quality for a sustainable environment. Soil Science Society of America, Madison, 107-124. https://doi.org/10.2136/sssaspecpub35.c7

Dick, R. P., Breakwill, D., & Turco, R. (1996). Soil enzyme activities and bio-diversity measurements as integrating biological indicators. In Doran, J. W., & Jones, A. J. (Eds.), *Handbook of Methods for Assessment of Soil.*

Doran, J. W., & Parkin, T. B. (1994). Defining and assessing soil quality. In Doran, J. W., Coleman, D. C., Bezdicek, D. F., & Stewart, B. A. (Eds.), *Defining soil quality for a sustainable environment. Soil Science Society of America, Madison* (pp. 3-21). https://doi.org/10.2136/sssaspecpub35.c1

Du, L. F., Zhao, J. J., Farhat, A., & Liu, W. K. (2013). Higher nitrates, P and lower pH in soils under medicinal plants versus crop plants. *Environmental Chemistry Letters*, *11*(4), 385-390. https://doi.org/10.1007/s10311-013-0419-1

Fan, J., & Hao, M. D. (2002). Kinetic characteristics of urease and alkaline phosphatase in dry land farming. *Agricultural Research in the Arid Atea,* 20(1), 35-37.

Frankenberger, W. T., & Dick, W. A. (1983). Relationships between enzyme activities and microbial growth and activity indices in soil. *Soil Science Society of America Journal*, *47*(5), 945-951. https://doi.org/10.2136/sssaj1983.03615995004700050021x

Guan, S. Y. (1986). Soil enzyme and its methods of research. Beijing: Agricultural Press, 303-337.

Jiao, X. G., Wei, D., & Sui, Y. Y. (2011). Effects of long-term fertilization on the soil enzyme activities and soil nutrients of the black and dark brown soils. *Chinese Journal of Soil Science*, *42*(3), 698-703.

Lalande, R., Gagnon, B., & Simard, R. R. (1998). Microbial biomass C and alkaline phosphatase activity in two compost amended soils. *Canadian J. Soil Science*, *78,* 581-587. https://doi.org/10.4141/S98-004

Lan, Y., Han, X. R., & Yang, J. F. (2011). Temporal and spatial dynamics of enzyme activities under long-term fertilization in a maize growing brown soil. *Plant Nutrition and Fertilizer Science*, *17*(5), 1197-1204.

Li, S. Q., Wang, H., Zhang R. F., & Zhao B. (2011). Analysis on distribution characteristics of soil nutrients in cultivated land of Anguo City. *Journal of Anhui Agricultural Science*, *39*(36), 22401- 22402,22915

Masciandaro, G., Ceccanti, B., Benedicto, S., Lee, H. C., & Cook, H. F. (2004). Enzyme activity and C and N pools in soil following application of mulches. *Canadian. J. Soil Science, 84*, 19-30. https://doi.org/10.4141/S03-045

Tabatabai, M. A., & Bremner, J. M. (1969). Use of p-nitrophenyl phosphate for assay of soil phosphatase activity. *Soil Biology and Biochemistry*, *1*(4), 301-307. https://doi.org/10.1016/0038-0717(69)90012-1

Taylor, J. P., Wilson, B., & Mills, M. S. (2002). Comparison of microbial numbers and enzymatic activities in surface soils and subsoils using various techniques. *Soil Biology & Biochemistry*, *34,* 387-401. https://doi.org/10.1016/S0038-0717(01)00199-7

Wang, J. H., Yin, R., Zhang, H. Y., Lin, X. G., Chen, R. R., & Qin, S. W. (2007). Changes in soil enzyme activities, microbial biomass,and soil nutrition status in response to fertilization regimes in a long-term field experiment. *Ecology and Environment, 16*(1), 191-196.

Zeng, L. L., Zhang, X., M., & Hong, Y. (2008). Effect of long- term fertilization and tillage system on soil enzyme activities. *China soil and fertilizer*, (2), 27-30.