ACTIVITY AND THERMODYNAMIC PARAMETERS OF UREASE IN SOILS AMENDED WITH ORGANIC RESIDUES

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ABSTRACT

An experiment was conducted to study the thermodynamic parameters (Q_{10}, Ea, ΔHa, ΔSa and ΔGa) of urease in loamy sand and silty clay soils treated with different sources of organic residues (cow residue, alfalfa leaves, wheat straw and poultry residue). Residues were mixed with soil at rate of 2%. The amended soils were incubated at 30°C for 30 days to undergo biochemical, chemical and physico-chemical reaction. Amended soils were incubated at 10, 20, 30, 40, 50, 60, 70 and 80°C. Urease activity was assayed and thermodynamic parameters were calculated. Results indicated that urease activity of amended soil was higher than that of control soils at various temperatures of incubation. Urease activity increased by increasing temperature of incubation from 10 to 40°C. Q_{10}, Ea, ΔHa, and ΔSa values of silty clay soil were in the order of : poultry manure > cow manure > alfalfa residue > wheat residue > control soil. However, in loamy sand soil the order was cow manure > alfalfa residue > wheat residue > control > poultry manure. Order of ΔGa was control > wheat residue > alfafa residue > cow manure > poultry manure at both soils. Except at poultry manure, all thermodynamic parameters values were higher in loamy sand soil as compared with their values in silty clay soil.

Key words:enzyme, soil, Ea, Q_{10}, ΔH, manures.

مجلة العلوم الزراعية العراقية -2019 :50(3):783-792 الانصاري وآخرون

فعالية انزيم اليوريز وثوابته الثرموودينامكية في الترب المعاملة بالمخلفات العضوية

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المستخلص :

نفذت هذه التجربة لدراسة الثوابت الثرموداينامكية (Q_{10}, Ea, Ha, ΔSa, ΔGa) لانزيم اليوريز في تربتين رملية مزيجة وطينية غرينية عوملت بمصادر عضوية مختلفة (مخلفات الابقار واوراق الجت وقش الحنطة ومخلفات الدواجن). خلطت المصادر العضوية مع التربة بمستوى 2% ثم حضنت عند درجة حرارة 30 م لمدة 30 يوماً لتحقيق التوازن الكيميوحيوي والكيميائي والكيموفيزيائي . حضنت الترب المعاملة عند درجات حرارة 10 و20 و 30 و 40 و 50 و 60 و 70 و 80 م ، قدر بعدها فعالية انزيم اليوريز ومنه حسبت الثوابت الثرموديناميكية. اشارت النتائج الى ان فعالية انزيم اليوريز للترب المعاملة بالمخلفات العضوية كان اعلى مما هو للترب غير المعاملة وعند كافة درجات حرارة الحضن. ازداد فعالية الانزيم بزيادة درجة حرارة الحضن من 10 الى 40 مْ. تأثرت الثوابت الثرموداينامكية بنوع المخلفات العضوية فقد ازدادت قيم Q_{10} و Ea وΔHa وΔSa في التربة الطينية الغرينية وفق التسلسل التالي: مخلفات الدواجن > مخلفات الابقار > اوراق الجت > قش الحنطة > المقارنة، بينما ازدادت القيم في التربة الرملية المزيجة وفق التسلسل التالي: مخلفات الابقار > اوراق الجت > قش الحنطة > المقارنة > مخلفات الدواجن. اما بالنسبة لـ Ga Δ فقد ازدادت وفق التسلسل التالي وللتربيتين : المقارنة > قش الحنطة > اوراق الجت > مخلفات الابقار > مخلفات الدواجن . كذلك اشارت النتائج ان قيم الثوابت الثرموديناميكية في التربة الرملية المزيجة اعلى مما هي للتربة الطينية الغرينية عند جميع انواع المخلفات العضوية فيما عدا محلفات الدواجن.

الكلمات المفتاحية: انزيم ، تربة، Ea ، Q_{10} ، ΔH، سماد عضوي.

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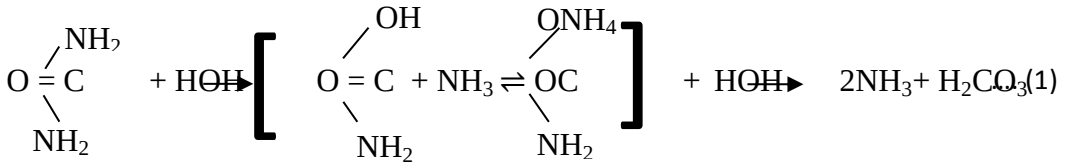
INTRODUCTION

Soil enzymes mediate biological reaction involve in nutrient cycling in soil, and can be used as potential indicators of soil quality(11,19,24). Enzymes activity in soil significantly correlated with organic residue (39) and soil microbial biomass (36 , 38). Organic residues influence microbial population since, they provide the soil with carbon and other nutrients essential for microbial growth. However, activity of soil enzymes are not affected only by organic matter content but also its type (9,33). Component of aparticular organic residue determine its effect on soil properties and biological activity (6, 7). Urease (urea amidohydrolase, EC 3.5.1.5) is one of the hydrolases enzymes play major roles in recycling plant nutrients in soil. Urease enzyme involves in nitrogen cycle, and its presence in soil is very important in hydrolysis of urea added to soil as fertilizer and animal waste (36). The general equation of the reaction catalyzed by urease as reported by Tabatabai (36) is



Application of organic residue as animal wastes or plant residues with or without urea fertilizer is a common practice by farmers to restore soil fertility. Yuan and Yue (42) reported that organic matter applied to soil provides enough substrate to increase microbial biomass, hence higher enzyme production. The activity of enzymes are directly and indirectly affected by soil temperature (21, 23). Their determination at different temperature can not only provide the optimum range of temperature or thermal stability for certain enzyme, but also allow to estimate thermodynamic parameters. Transition-state theory uses thermodynamic concepts to describe chemical-and enzyme-catalyzed reaction (20). Perucci and Scarponi (30) indicated that general conclusion of enzyme reactions in soil can be reached from the thermodynamic parameters of the enzyme-soil system. Such parameters include: temperature coefficient (Q_{10}), activation energy (Ea), change in enthalpy (ΔHa) and in entropy (ΔSa), and free energy of activation (ΔGa). Little informations are available about the effect of type of organic residues on urease activity and its kinetic and thermodynamic parameters. The purpose of this study is to present effect of temperature on activity and thermodynamic parameters of urease enzymes in soil amended with different sources of organic residues.

MATERIALS AND METHODS

A surface layer samples (0 – 15 cm) of soil from Al – Zubair area (coayse , mixed, calcareous, hyper thermic, Typic Torripssaments) and Abul – Khasib area (fine, mixed, calcareous, hyper thermic , Typic Torriflovents) were collected. Soils texture were loamy sand and silty clay, respectively. Soils were collected to obtain wide range in O.M, total N, $CaCO_3$, electrical conductivity, and texture. Four different types of organic residues were used: alfalfa leaves (*Madicago sativa* L.), wheat straw (*Triticum aestivum* L.), cow residue, and poultry residue. Organic residues were oven-dry (70℃), grounded then sieved to pass 2mm mesh. Sieved residues were mixed with soil at rate of 2% (organic residue:soil). The amended soils were incubated at 30℃ for 30 days to undergo biochemical, chemical and physico-chemical reaction as reported by Perucci and Scarponi (30). Moisture content was maintained at field capacity by adding water throughout incubation period by periodic weighing of samples. Some chemical,physical and biological properties of soils and organic residues were determined following standard procedures describe by Page *et al*. (29) (Table 1).

Table 1. Some properties of soils and organic materials used.

Soil or organic material	E.C. dS m^{-1}	pH *	Organic C	Total N	$CaCO_3$	C/N	Moisture content at field capacity(%)	Urease activity $\mu gNH^{+}_4–Ng^{-1}$ soil. $2hrs^{-1}$
			g kg^{-1}					
Loamy sand soil**	2.93	8.00	0.33	0.03	82.31	11.00	20.00	16.16
Silty clay soil**	5.99	7.77	3.94	0.36	316.88	10.94	31.31	40.73
Cow residue	11.10	7.20	258.60	18.40	---	14.00	---	---
Alfalfa leaves	10.10	5.80	416.00	27.70	---	15.00	---	---
Wheat straw	11.70	6.00	392.60	4.20	---	92.70	----	---
Poultry residue	11.02	6.50	265.4	27.30	---	9.72	---	---

*** 1:1 for soil and 1:5 for organic residues**

**** particle size distribution of Loamy sand soil (84.50% sand,7.00 % silt and 8.50 % clay) and of Silty clay soil (14.62% sand ,44.91% silt and 40.47 % clay).**

To study the temperature effect on urease activity, soil samples amended with organic residues and incubated at different temperatures(10, 20, 30, 40 ,50 , 60, 70 and 80℃) for 2 hours then brought to room temperature. Urease activity was assayed by following procedure described by Tabatabai and Bremner(36). Five grams of amended soils were incubated with 9 ml of 0.05 *M* pH 9.0 tris (hydroxymethyl)- aminomethane (THAM) buffer, 0.2 ml of toluene, and 1 ml of 0.2 *M* substrate(urea) solution at 37˚C for 2 hours. After incubation, urea was inhibited by addition of KCl – Ag_2SO_4 solution, then NH_4^+-N released was determined by distillation procedure. The activation energy (Ea) of urease in amended soils was calculated from enzyme activity obtained at 0.25*M* urea but temperatures of incubation varid from 10 to 40℃. The activation energy were calculated using Arrhenius equation plot of Log K against 1/T (40):

$K=A.\exp(-Ea/RT)$ -----(2)

where A is the preexponential factor, Ea is the energy of activation, R is the gas constant and T is the temperature in degree Kelvin. The temperature coefficient (Q_{10}) was calculated by the equation described by Frankenberger (11):

$$Q_{10} = \exp \frac{1000\ Ea}{8.314\ T\ (T+10)} \quad \text{---------- (3)}$$

Change in enthalpy (ΔHa) and entropy (ΔSa) of activation values have been calculated from equation described by Perucci and Scarponi (30):=

$\Delta Ha = Ea - RT$ ---------- (4)

$\text{Log } K/T = 10.319 + \Delta Sa/4.574 - \Delta Ha/5.574\ T$ ---- (5)

The free energy of activation (ΔGa) values were calculated according the equation:

$\Delta Ga = \Delta Ha - T\ \Delta Sa$ ---------- (6)

All results reported are average of three replicates expressed on air-dry basis. All values are expressed as the mean of three replicates and analyzed by two–way analysis of variance (ANOVA) using procedure of SPSS 20 . Differences among treatment means were statistically measured using Revised LSD(4).

RESULTS AND DISCUSSION

Figs. 1 and 2 show that at various temperatures of incubation urease activity of amended soils was higher than that of control soils and were higher in loamy sand soil than in silty clay soil. These results in according with those of Abdulkareem *et al*.(2); Kizilkaya and Bayrackli (19); Meloro *et al*.(25) and Okur *et al*.(27). Similar results reported by Dora *et al*.(12) for phosphatase activity in soil. Wolinska and Stepniewska (41) stated that there is high correlation coefficient between enzymatic activity and TOC (Total Organic Carbon). Initial soil properties have influenced the stimulatory effect of organic residue on enzyme production and activity in soil(31). Lai and Tabatabai(20) showed that adsorption of enzyme on clay in soil reduced enzyme activity. Furthermore, Boyed and Mortland(3) reported that urease adsorbed on montmorillonite-organic complaex reduced its activity and Km values. Urease activity in soils under study increased by increasing incubation temperature from 10 to 40ºC. Dash *et al*.(10)

reported maximum urease activity in some Indian soil obtained at 47ºC. Incubation at 37ºC has been used to assay urease activity in most temperate soils (37). At all treatments increasing temperature of incubation beyond 40ºC significantly decreased urease activity. Maximum urease activity in our study was reach at temperature lower than those suggested by Tabatabai (36), who reported that enzyme activities in soil inactivated at temperature between 60-70ºC.

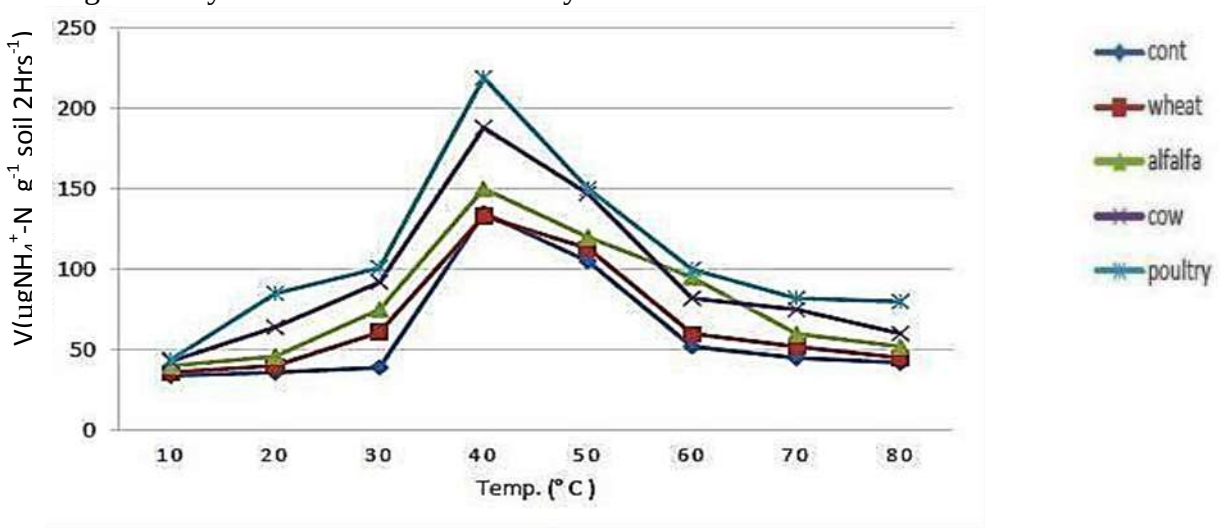


Fig.1: Effect of incubation temperature on urease activity in Silty Clay soil amended with organic residues

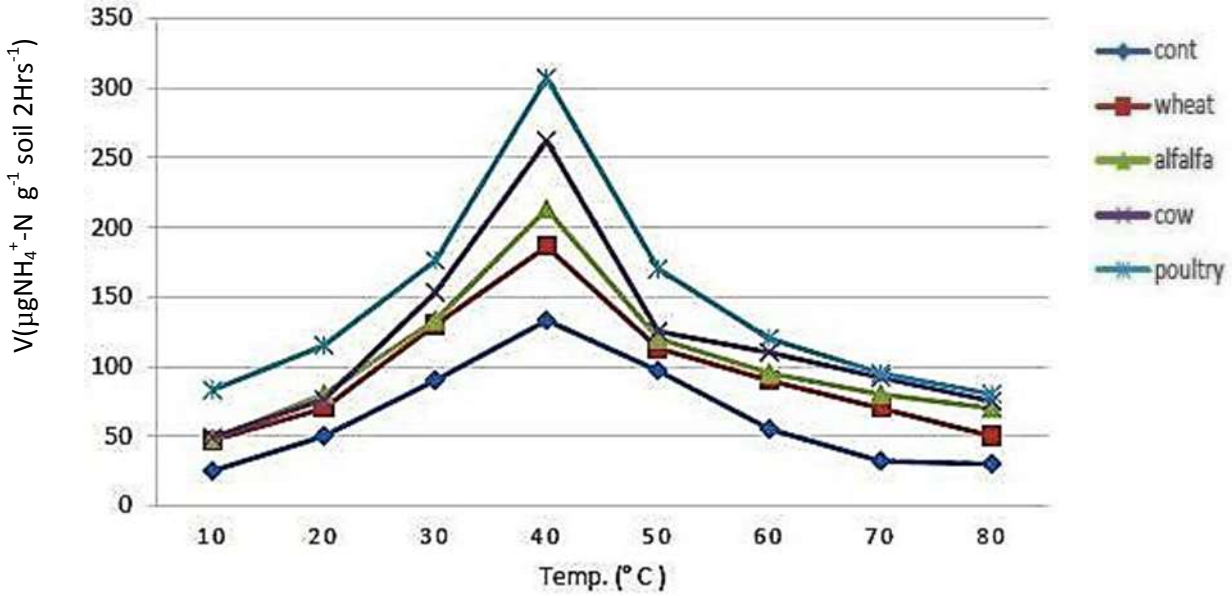


Fig 2 : Effect of incubation temperature on urease activity in Loamy Sand soil amended with organic residues

The rate of enzyme catalysis generally increase with increase in temperature until it reached the optimum temperature then decline as a result of enzyme denaturation, hence its activity reduced (41). Moyo *et al*.(26) reported that effect of temperature on urea hydrolysis affected by pH, urea concentration, water content, and other factors. The stability of enzymes increased as temperature decreased below optimum for activity. Khazeiv (18) reported that, free and weakly bound soil enzymes are most active, but with increasing temperature to optimum, the immobilized enzymes become more active. He also reported that the changes in temperature in thermodynamic characteristics indicate heterogeneity in the composition and state of soil enzyme. O'Toole and Morgan (28) suggested existence of a number of types of organic material-urease complexes in soils,

each possessing destinate characteristics including thermal stability. The temperature dependence of the rate constant of an enzyme reaction can be described by Arrhenius equation which could express as:

Log K= - (Ea/2.303 RT) + Log A. --(7)

The constant A is a factor related to the frequency of collisions of the reactants (e.g. substrate) with the enzyme. The collisions frequency is influenced by the concentration of the reactant molecules and their kinetic energy (14). The Arrhenius plot for urease enzyme activity of different treatments under study were linear between 10 and 40ºC (figs. 3 and 4). The activation energy values for reaction catalyzed by urease expressed in kJ mol^{-1} were significantly affected by organic residues source(table 2)and ranged between 47.10 in loamy sand soil treated with cow manure and 35.61 in soil treated with poultry manure, while those of silty clay soil the values were 41.74 in soil trwith eated poultry manure and 29.67 for control soil (table 3). The values obtained in this study were higher than the reported by Juan *et al*.(17) (21.06-23.99 kJ mol^{-1}), but lower than that reported by Su (35) (59.6-77.8 kJ mol^{-1}). Lai and Tabatabai (20) reported values in range 13.5-39.1 kJ mol^{-1} for immobilized enzyme. Gutfreund (15) reported that a typical activation energy for urease enzyme is about 42 kJ mol^{-1}, while Zhang *et al*.(44) reported Ea values for urease range from 30.10 to 173.89 kJ mol^{-1}.Theoretically, the activation energy for a single enzyme catalyzed reaction could not vary. However, Moyo *et al*.(26) attributed the variation in Ea values for soil urease reported in literatures to factors such as pH, urea concentration, or water content other than temperature and to various assay techniques used which could induce conditions that influence differently the energy requirements for the formation of enzyme-substrate complex and, hence, the energy of a ctivation. Lai and Tabatabai (20) indicated activation energies should be compared only under similar conditions such as pH , choice of buffer , substrate concentration and temperature of incubation. Su (35) reported that urease enzyme reaction was more likely to occur, and less energy barrier needed to overcome in soil amended with organic fertilizer than urea. However, Zhange *et al*.(44) stated that protection of urease by the association with the soil organic and inorganic minerals complexes, the higher the barrier to be overcome during formation of enzyme-substrate complex.

Table 2. RLSD values of thermodynamic parameters of soil urease

S.o.v	RLSD(0.05)				
	E a	**Q_{10}**	**ΔH**	**ΔS**	**ΔG**
Treatment(t)	**2.66**	**0.08**	**2.20**	**18.18**	**0.34**
Temperature(T)	**___**	**0.05**	**Ns**	**12.12**	**0.18**
Soil(s)	**1.83**	**0.03**	**1.64**	**8.14**	**Ns**
t x T	**____**	**0.13**	**4.00**	**23.32**	**0.55**
t x s	**4.14**	**0.10**	**3.43**	**20.93**	**Ns**
T x s	**____**	**0.10**	**Ns**	**20.00**	**0.40**
t x T x s	**____**	**Ns**	**Ns**	**30.86**	**0.68**

ns: not significant

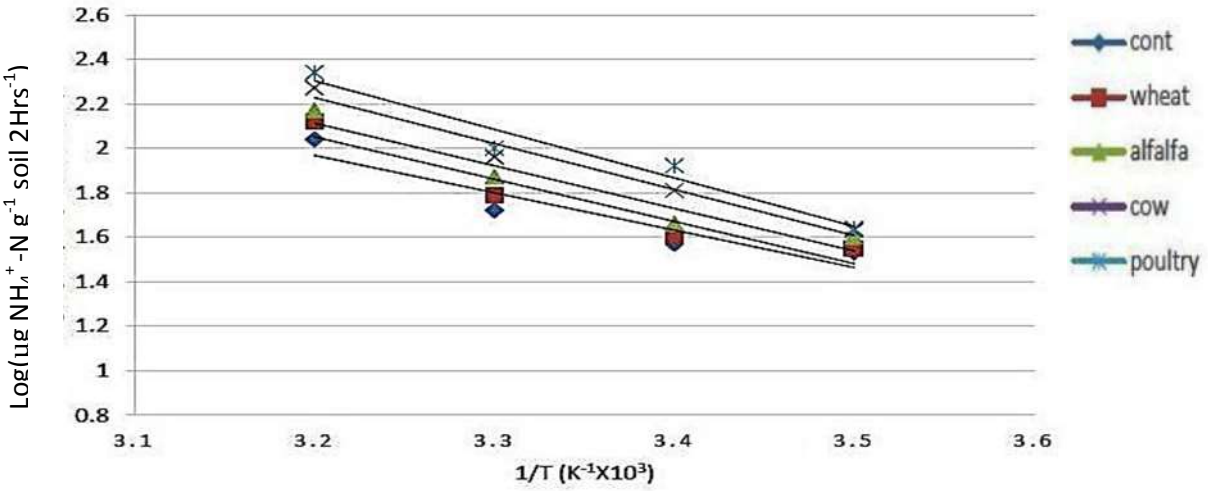


Fig.3: Arrhenius equation plot of urease in Silty Clay soil amended with organic residues

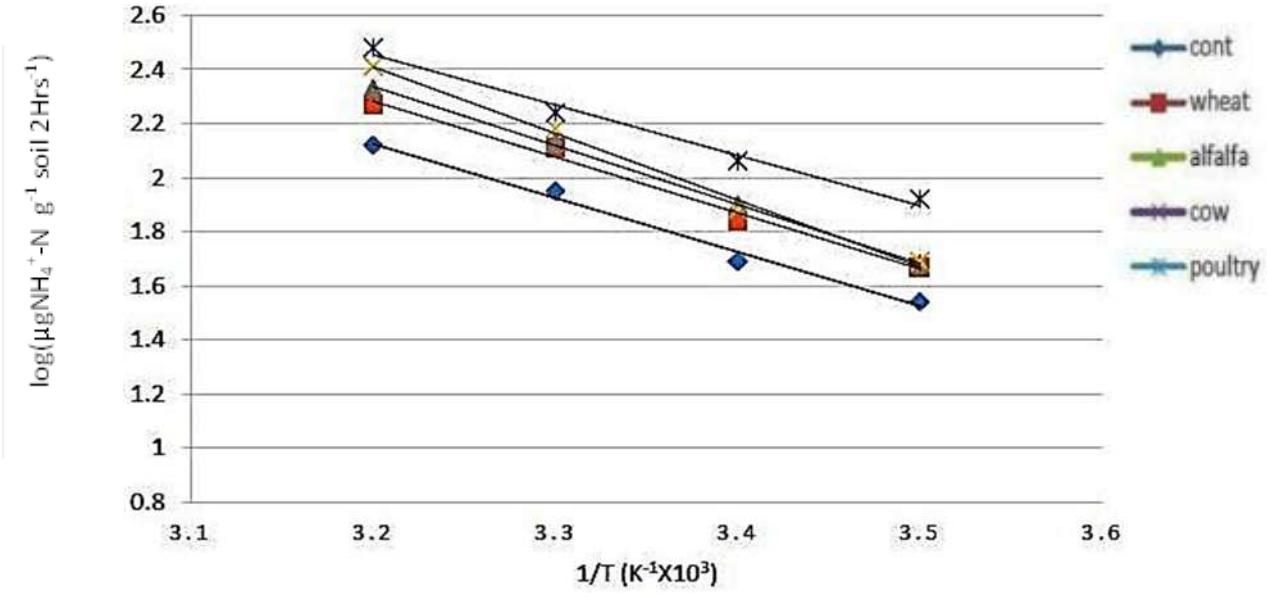


Fig.4: Arrhenius equation plot of urease in Loamy Sand soil amended with organic residues

Table 3. Activation energy (Ea) and temperature coefficient (Q_{10}) of urease in soils amended with organic residues

Treatment	Loamy sand soil						Silty clay soil					
	Ea kJ mol^{-1}	Q_{10}					Ea kJ mol^{-1}	Q_{10}				
		10°C	20°C	30°C	40°C	Mean of Q_{10}		10°C	20°C	30°C	40°C	Mean of Q_{10}
Poultry manure	35.61	1.67	1.61	1.56	1.52	1.59	41.74	1.83	1.75	1.69	1.64	1.72
Wheat residue	39.63	1.77	1.70	1.65	1.60	1.68	36.37	1.69	1.63	1.58	1.54	1.61
Cow manure	47.10	1.97	1.89	1.81	1.75	1.85	39.42	1.77	1.70	1.64	1.59	1.67
Alfalfa residue	41.74	1.83	1.75	1.69	1.64	1.72	36.76	1.70	1.64	1.59	1.54	1.61
Control	38.29	1.74	1.67	1.62	1.57	1.65	29.67	1.53	1.49	1.45	1.42	1.47
Mean	40.47	1.79	1.72	1.66	1.61	1.69	36.79	1.70	1.64	1.59	1.54	1.62

Temperature coefficient (Q_{10}) most often is used to study the effect of temperature on enzyme catalyzed reaction, however, enzyme catalyzed reaction are less sensitive to temperature change than their uncatalyzed counter parts, whereas the uncatalyzed rate may double with every 10℃ increase in temperature, the enzyme catalyzed reaction rate will increase by a factor of < 2 (43). The lower Q_{10} values of soil hydrolases, the lower thermodynamic effect on enzyme reaction. Data in Table 3 shows that Q_{10} values of urease enzymes in loamy sand soil (1.69) were greater than in silty clay soil (1.61)(Table 2). In most cases Q_{10} values (1.42-1.89) were within range of Q_{10} values reported in literatures (1.10-1.80) (5). Juan *et al*.(17) reported Q_{10} values for urease in soil ranged from 1.32 to 1.42, while Jayan (16) reported values between 0.5 and 2.4 .Q_{10} values of both soils under study were in order of: cow manure > alfalfa residue > poultry manure> wheat residue> control soil with significant differences(Table 2). The Q_{10} values of soil enzymes is an indicator of kinetics energy required for the reaction catalyzed by enzyme in soil. The lower the Q_{10} value, the lower the kinetics required for a reaction. Q_{10} of 2 is equivalent to an Ea of about 12600 cal mol^{-1} (53 kJ mol^{-1}) at 25℃ (32). Thermodynamic parameters (ΔHa, ΔSa and ΔGa) were calculated and presented in table (4). The ΔHa values for urease enzymes in loamy sand soil with values range from 49.70 to 37.96 kJ mol^{-1} were significantly higher (Table 2) than those of their counter parts of silty clay soil with values between 44.34-32.04 kJ mol^{-1}. Lai and Tabatabai (20) reported values for ΔHa values for urease in range of 36 kJ mol^{-1}. Juan *et al*.(17) reported values between 19.08-21.64 kJ mol^{-1}. In loamy sand soil effect of organic residue of ΔHa values was in the following order: cow manure > alfalfa residue > wheat residue > control > poultry manure. However, in silty clay soil, the order was poultry manure > cow manure > alfalfa residue > wheat residue > control soils. Increasing temperature of incubation from 10ºC to 40ºC did not significantly affect the obtained ΔHa values for both control and amended soils. Similar results were reported by Juan *et al*.(17). Lai and Tabatabai (20) and Cornish-Bowden (8) reported that a large enthalpy of activation (ΔHa) indicates that a large amount of stretching, squeezing or even breaking of chemical bonds is necessary for the formation of transition state. Perucci and Scarponi (30) reported that amendment soil with crop residue lowers the steric effect and stabilized the formation of enzyme-substrate complex. At all incubation temperatures and at both soils amended with different organic residues, except poultry manure, ΔS values for loamy sand soil were significantly higher (Table 2) than those of silty clay soil and range between 124.04 and 81.74 J $degree^{-1}mol^{-1}$ for loamy sand soil and 96.65 and 61.77 J $degree^{-1}$ mol^{-1} for silty clay soil, while those of poultry manure treatment were in range of 84.51 and 74.84 J $degree^{-1}$ mol^{-1} for loamy sand soil and 104.92 and 93.67 J $degree^{-1}$ mol^{-1} in silty clay soil (Table 4). Negative values for ΔS

Table 4. ΔHa (kJ mol^{-1}) , ΔSa (J degree^{-1} mol^{-1}) and ΔGa (kJ mol^{-1}) of urease in soils amended with organic residues

Treatment	Parameter	Loamy sand soil					Silty clay soil				
		10ºC	20ºC	30ºC	40ºC	Mean	10ºC	20ºC	30ºC	40ºC	Mean
Poultry manure	ΔHa	37.96	38.04	38.12	38.21	38.08	44.09	44.17	44.25	44.34	44.21
	ΔSa	84.51	80.80	77.55	74.84	79.42	104.92	101.11	96.35	93.76	99.03
	ΔGa	14.04	14.36	14.62	14.78	14.45	14.39	14.54	15.06	14.99	14.74
Wheat Residue	ΔHa	41.98	42.06	42.14	42.23	42.10	38.72	38.80	38.88	38.97	38.84
	ΔSa	97.59	93.54	90.23	86.69	92.01	85.53	81.28	77.98	75.61	80.10
	ΔGa	14.36	14.65	14.80	15.09	14.72	14.51	14.98	15.25	15.30	15.01
Cow Manure	ΔHa	49.45	49.53	49.61	49.70	49.57	41.77	41.85	41.93	42.02	41.89
	ΔSa	124.04	119.19	115.19	11.25	117.41	96.65	92.64	88.82	86.03	91.03
	ΔGa	14.34	14.60	14. 71	14.87	14.63	14.41	14.71	15.02	15.09	14.80
Alfalfa residue	ΔHa	44.09	44.17	44.25	44.34	44.21	39.11	39.19	39.27	39.36	39.23
	ΔSa	105.07	100.99	97.29	93.74	99.27	87.11	82.89	79.66	77.10	81.69
	ΔGa	14.35	14.58	14.77	14.99	14.67	14.45	14.90	15.13	15.22	14.92
Control	ΔHa	40.64	40.72	40.80	40.89	40.76	32.02	32.10	32.18	32.27	32.14
	ΔSa	92.28	88.29	85.06	81.75	86.84	61.77	58.31	54.96	53.83 57.21	
	ΔGa	14.52	14.85	15.03	15.30	14.92	14.53	15.01	15.53	15.42	15.12

for urease in soil were reported by Rao (31), Srinivas (34) and Jayan (16).

Increasing in temperature of incubation from 10ºC to 40ºC significantly lower ΔS values at all treatments. Amended both soils with organic residue resalted in significant effect (table 2) on ΔS and following the order : cow manure > alfalfa residue > Poultry manure > wheat residue > control soil. The ΔGa values reported in Table 4 showed that the free energy of urease enzyme in treatments of the study were similar inspite of differences in ΔHa and ΔSa values obtained for different treatments. Similar trends for inorganic phosphatase (1) and arylsulphatase (30) were reported. Liang *et al*.(22) stated that improving soil condition will decrease Q_{10} , Ea, and ΔH values. The relatively low Q_{10}, Ea, and ΔH values increase enzymatic reaction in soil. It could be concluded that soil temperature has profound effect on urease activity and thermodynamic properties in soil amended with organic residues. Hence, controlling soil temperature in a feasible way regulating biochemical transformation of nutrient catalyzed by soil urease.

REFERENCES

1- Abdulkareem, M. A. and A. M. S. Al-Ansari .2008. Thermodynamic parameters of inorganic pyrophosphatase in marsh land south of Iraq. Marsh Bull. 3:95-103

2- Abdulkareem, M.A., A. M. Al-Anasri and S. J. Kadhum. 2012. Urease activity and kinetic parameters in soil amended with organic residues.J.of King Saud Univ., Agric. Sci. 25:17-26

3-Boyd,S.A. and M.M. Mortland . 1986. Selective effects of smectite-organic complexes on the activities of immobilized enzymes . J.Molecular Cat. 34:1-8

4-Al-Rawi,K.M. and A.M. Kalafali.1980. Design and Analysis of Agricultural Experiments.1st ed. Musul Univ.Press .pp:488. (In Arabic).

5- Browman, M. G. and M. A. Tabatabai. 1978. Phosphodiesterase activity of soils. Soil Sci. Soc. Amer. J., 42: 284-290

6- Charves, B. , S. DeNeve, G. Hofinan , P. Boeckx and O. Van Cleemput, 2004. Nitrogen mineralization of vegetable root residues and green manures as related to their (bio) chemical composition. Eur. J. Agron., 21: 161-170.

7- Clement, A., J. K. Ladha and F. P. Chalifour. 1998. Nitrogen dynamics of various green manure species and the relationship to low land rice production. Agron. J. 90: 149-154

8- Cornish-Bowden, A. 2004. Fundamentals of Enzyme Kinetics. 3rd ed. Portland Press, London. pp:244

9- Dalal, R. C. 1982. Effect of plant growth and addition of plant residues on the phosphatase activity in soil. Plant and Soil, 66: 265-269

10- Dash, M. C., P. C. Mishra, R. K. Mohanty and N. Bhatt. 1981. Effect of specific conductance and temperature on urease activity in some Indian soils. Soil Biol. Biochem. 13:73-74

11- Dick, R. P. 1998. Soil Enzyme Activities as Integrative Indicators of Soil Health. In: CABI Pub. Biological Indicators of Soil Health. pp: 156-212

12- Dora, A. S., C. Domuta, M. Sandor and V. Adrian. 2010. The estimation of phosphatase activity in soil. Res. J. Agric. Sci. 42: 311-314.

13- Frankenberger, W. T. J.R. 1983. Kinetic properties of L-Histidine ammonia-Lyase activity in soils. Soil Sci. Soc. Amer. J. 47: 71-74.

14- Gray, C. L. 1971. Enzyme-Catalyzed Reactions. Van Nostrand Rein-Hold Co. Landon .pp:86

15- Gutfreund, H. 1972. Enzymes: Physical Principle. Wiley-Interscience, London

16- Jayan, V. L. 2012. Urease and Phosphomonoesterase Activities in Soil-Their Distribution, Kinetics and Influence of Management Practices on Their Activities. Ph.D. Dissertation. Coll. Agric. Acharya. N. G. Ranga Agricultural Univ. Hyderabad.pp:219.

17- Juan, Y. H., Z. H. Chen, L. J. Chen, Z. J. Wu, R. Wang, W. T. Sun and Y. L. Zhang. 2010. Kinetic thermodynamic behaviors of soil urease as affected by urease inhibitors. R.C. SueloNutr. Veg. 10:1-11

18- Khazeiv, F. 1975. Thermodynamic characteristics of enzymatic reactions in soil. Biol-Nauki 10: 121-127

19- Kizilkaya, R. and R. Bayrokli. 2005. Effects of N-enriched sewage sludge on soil enzyme activities. Appl. Soil. Ecol., 30: 192-202

20- Lai, C. M. and M. A. Tabatabai. 1992. Kinetic parameters of immobilized urease. Soil Boil. Biochem., 24: 225-228.

21- Leiros, M. C., C. Trasar-Cepeda, F. Garcia-Fernandez and F. Gil-Sotres. 1999. Defining the validity of a biochemical index of soil quality. Boil. Fert. Soils. 30: 140-146.

22- Liang, L. I., X. Q. Liang, H. Li, Y. J. Ji, J. Liu, Y. S. Ye, G. M. Tian, Y. X. Chen and Y. M. LUO. 2015. Phosphomonoesterase activities, kinetics and thermodynamics in paddy soil after receiving swine manure for six years. Pedosphere. 25: 294-306

23- McClaugherty, C. A. and A. E. L. Linkins. 1990. Temperature responses of enzymes in two forest soil. Soil Biol. Biochem. 22: 29-33

24- McLatchey, G. P. and K. R. Reddy. 1998. Regulation of organic matter decomposition and nutrient release in a wetland soil. J. Environ. Qual., 27: 1268-1274

25- Melero, S., J. C. R. Porass, J. F. Herencia and S. Madejon. 2006. Chemical and biochemical properties in a silty loam soil under conventional and organic management. Soil Till. Res. 90: 162-170

26- Moyo, C. C., D. E. Kissel and M. L. Cabrer. 1989. Temperature effects on soil urease activity. Soil Biol. Biochem.21: 935-938

27- Okur, N., A. Altindisli and M. Cengel. 2009. Microbial biomass and enzyme activity in vineyard soils under organic and conventional farming systems. Turk. J. Agric. For. 33: 413-423

28- O'Toole, P. and M. A. Morgan. 1984. Thermal stabilities of urease enzymes in some Irish soils. Soil Biol. Biochem. 16: 471-474

29- Page, A. L., R. H. Miller and D. R. Keeny. 1982. Methods of Soil Analysis. 2nd ed., Madison, Wisconson, USA

30- Perucci, P. and L. Scorponi. 1984. Arylsulphatase activity in soils amended with crop residues. Kinetic and thermodynamic parameters. Soil. Boil. Biochem. 16: 605-608

31- Rao, S. V. 1989. Distribution, Kinetic and Some Interaction of Urease and Phosphatases in Soils. Ph.D. Dissertation. Acharya. N. G. Ranga Agricultural Univ. Hyderabad.pp:249

32- Segel, L. H. 1975. Enzyme Kinetics Behavior and Analysis of Rapid Equilibrium and Steady-State Enzyme Systems. John Wiley and Sons. Inc. New York.pp:957

33- Speir, T. W. and D. J. Ross. 1978. Soil Phosphatase and Sulphatase. In: R. G. Burns (ed.). Soil Enzymes Acad. Press, London. pp: 197-250

34-Srinivas,D.1993. Phosphomonoesterase Activity in Some Soils of Andhra Pradesh. Distribution, Kinetics and Variation Due to Plant Cover, Pesticide Application and Submergence. Ph.D. Dissertation. Acharya. N. G. Ranga Agricultural Univ. Hyderabad.pp:283

35- Su, M. M. 2013. Effect of Fertilization on Kinetic and Thermodynamic Parameters of Soil Urease in Paddy Fields. M.Sc. Thesis. China Glob Thesis.pp:125

36- Tabatabai, M. A. 1994. Soil Enzymes. In: R. W.Weaver , S. Augle, P. J. Bottomly, D. Bezdicek, S. Smith, M. A. Tabatabai and A. Wollum. (eds.) 2010. Methods of soil analysis. Part 2. Soil. Sci. Soc. Amer. Madison. pp: 775-833

37- Tabatabai, M. A. and J. M. Bremner. 1972. Assay of urease activity in soils. Soil. Biol. Biochem. 4: 479-487

38- Tejada, M., M. T. Hernandez and C. Garcia. 2009. Soil restoration using composted plant residues : Effects on soil properties. Soil Tell. Res. 102: 109-117.

39- Tejada, W., C. Garcia, J. L. Gonzalez and M. T. Hernandez. 2006. Organic amendment based on fresh and composted beet vinasse: influence on physical, chemical and biological properties and wheat yield. Soil Sci. Soc. Amer. J. 70: 900-908

40-Trasar-Cepeda,C. ,F.Gil-Stores and M.C. Learos.2007 .Thermodynamic parameters of enzymes in grassland soils from Galicia NW Sprin.Soil Biol.Biochem.30:311-319

41- Wolinska, A. and Z. Stepniewska. 2012. Dehydrogenase Activity in The Soil Environment. In: Rosa Anela (ed.). Biochemistry, Genetics and Molecular Biology. CC By license. pp: 183-210.

42- Yuan, B. and D. Yue. 2012. Soil microbial and enzymatic activities across a chronosequence of Chinese pine China. Plantation development on the Loess planteau of Pedosphere. 22: 1-12.

43- Zeffren, E. and P. L. Hall. 1973. The Study of Enzyme Mechanisms. John Wiley and Sons Inc., New York.pp:284

44- Zhang, Y. L., L. J. Chen, C. X. Sun, D. P. Li, Z. J. Wu and Z. H. Duan. 2010. Kinetic and thermodynamic properties of hydrolases in northeastern China soils as affected by temperature. Agrochemica. 54: 231-242.