EFFECT OF VERMICOMPOST, SEAWEED EXTRACTS AND NITROGEN FERTILIZERS ON L-ASPERGENASE ENZYME ACTIVITY AND ITS KINETIC

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Abstract

To study the effect of application of vermicompost, seaweed extracts and nitrogen fertilizers on the activity of the amidohydrolases enzyme (L-Aspergenase) and their kinetic measures in the rhizosphere at the flowering and maturity stages of maize (*Zea mays* L. hybrid Furat) a field experiment was carried out during the autumn season of 2019-2020 in one of the fields of Al-Noria village - Al-Diwaniyah governorate / Iraq. The experiment was arrangement as factorial according to randomized complete blocks design (RCBD) at three replications. The experiment included three factors, the first factor included the application of vermicompost at three levels (0, 2 and 4 ton ha^{-1}) symbol as A0, A1 and A2 respectively, while the second factor included application of seaweed extract at two levels (0 and 40 Kg ha^{-1}) symbol as B0 and B1 respectively, whereas the third factor included application three levels of nitrogen fertilizer (0, 120 and 240 kg N ha^{-1}) symbol as C0, C1 and C2 respectively. The vermicompost levels were significantly different, the A1 treatment gave the highest means of L-Aspergenase enzyme activity (151.2 and 176.6 µg N-NH$_4^+$ g^{-1} soil 2h^{-1}) at the flowering and maturity stages respectively. Also, The application of seaweed extract (B1) was significantly effect on the most studied traits and gave the best results of L-Aspergenase enzyme activity (135.00 and 162.00 µg N-NH$_4^+$ g^{-1} soil 2h^{-1}) respectively at the flowering and maturity stages. The C1 treatment was significantly superior and gave the highest means of L-Aspergenase enzyme activity, K$_m$ and V$_{max}$ of L-Aspergenase (137.5 µg N-NH$_4^+$ g^{-1} soil 2h^{-1}, 51.4 mM, 753.00 µg N-NH$_4^+$ g^{-1} soil 2h^{-1}) respectively at the flowering stage. The interaction between vermicompost and seaweed extracts had significant effect on the L-Aspergenase enzyme activity, V$_{max}$ at the flowering and maturity stages and K$_m$ at the flowering stages, while the interaction between vermicompost and nitrogen fertilizer had significant effect on the L-Aspergenase enzymes activity at the flowering and maturity stages, whereas the interaction between seaweed extract and nitrogen fertilizer had significant effect on the L-Aspergenase enzymes activity at the flowering and maturity stages an V$_{max}$ at flowering stage. From other hand, The interaction between three factors had significant effect on the L-Aspergenase enzyme activity at flowering and maturity stages, Km at flowering stage and Vmax at maturity stage.

Key words: Amidohydrolases enzyme, rhizosphere, V$_{max}$, affinity constant, Maize

Soil is one of the natural ingredients in agriculture, and its quality and fertility plays an important role in the productivity of various plants, despite its importance, but Iraqi soils, especially in the central and southern regions, are still suffering from low readiness of many nutrients necessary for plant growth and development, which is due to many reasons, including it is related with soil characteristics such as the degree of interaction, texture and composition, as well as its low content of organic matter and others related with agricultural intensification and failure to follow the scientific method in soil nutrient management programs, which requires work to apply appropriate fertilizer programs for each soil that may lead to an improvement the soil's physical, chemical and biological properties, and as a result an increase Its fertility (Colombo et al 2002).

The application of nitrogen fertilizers is one of the factors that increase soil fertility, and it is one of the major elements necessary for the plant due to its physiological roles within the plant tissue, except that the excessive use of nitrogen fertilizers is a result of its loss from soils by volatilization and leaching (Bronson, 2004), as well as unexamined applications to nitrogen fertilizer leads to an imbalance in the readiness of the elements in the soil, which negatively affected in the soil fertility (Havlin et al 2005).

To reduce these negative impacts, modern trends of agriculture emphasize reducing pollutants and replacing mineral fertilization or part of it with organic fertilizers, such as the application of seaweed extracts, which is one of the most important biological fertilizers due to its content of organic matter that retains moisture and helps increase the readiness of nutrients (Spinelli et al 2009), as well as their content of many natural compounds that act as growth regulators (O′Dell 2003), and their role in improving the physical properties of the soil as they help in improving the conditions of ventilation, permeability, water holding, reducing soil bulk density and increasing its porosity (Naseem 2005). The application of mineral fertilizers can also be reduced by applying other types of organic fertilizers known as vermicompost, which is an organic fertilizer that earthworms produce by analyzing organic compounds and converting them to the simple substances that benefit the plant, in addition to, the studies indicated that the vermicompost contains a number of enzymes such as peroxidase, protease and Amylase that enhance the efficacy of soil microorganisms (Bottinellin et al 2010).

The following of the correct approach to soil service, especially the integration between the use of mineral and organic fertilizers, is the key to managing successful operations as a result of their effect on the chemical and physical properties of the soil as well as their effect on the activity of micro-organisms and the enzymatic activity in the soil, which is a biological indicator for monitoring soil quality through its participation in metabolic processes. (Yang et al 2008). One of these enzymes is L-Asparagenase, which is one of the hydrolysis enzymes that contribute to the decomposition of organic matter and has a major role in the nitrogen cycle, as well as the increasing its effectiveness in the soil leads to an increase in nutrients and the positive effect of that on increasing soil fertility (Hui et al 2013). This enzyme works on the decomposition of the aspirgin amino acid and its conversion to aspartic acid, releasing ammonia gas (Magdy et al 2008), in addition to, the activity of this enzyme reflects the ability to convert organic nitrogen into inorganic nitrogen that is easily absorbed by the plant, while the kinetic measures of the enzyme reflect the maximum velocity (V_{max}) of the maximum activity of the enzyme, and the Michaelis constant (K_m) reflects the affinity between the enzyme and the subject matter. Therefore, the present study aims to study the effect of vermicompost, seaweed extract and nitrogen mineral fertilizer as well as the interaction between them on the activity of the L-Asparagenase enzyme and its kinetic measures in the rhizosphere of maize at the flowering and maturity periods.

Material And Methods

A field experiment was carried out during the autumn season of 2019-2020 in one of the fields of Al-Noria village - Al-Diwaniyah governorate / Iraq in in a soil as shows their physical and chemical properties in Table 1, to study the effect of vermicompost, seaweed extract and nitrogen mineral fertilizer as well as the interaction between them on the activity of the L-Asparagenase enzyme and its kinetic measures in the rhizosphere of maize at the flowering and maturity periods. The experiment was arrangement as factorial experiment according to Randomized Complete Blocks Design (RCBD) at three replications. The experiment included three factors, the first factor

included the application of vermicompost at three levels (0, 2 and 4 ton ha^{-1}), the physical and chemical properties of vermicompost shows in Table 2, while the second factor included application of seaweed extract at to levels (0 and 40 Kg ha^{-1}), whereas the third factor included application three levels of nitrogen fertilizer (0, 120 and 240 kg N ha^{-1}).

Soil management especially plowing were carried out as required, the net area of sub sub plot was (2 m long x 2 m width) 4 m^2 which contained 4 rows, 0.50 m apart and 0.25 m within the plants. Recommended phosphorus fertilizer (100 Kg P ha^{-1}) as super triphosphate (48% P$_2$O$_5$) and potash fertilizer (120 Kg K ha^{-1}) as a potassium sulfate (41.5% K) were applied at the time of planting, while the vermicompost and nitrogen fertilizers (as a urea 46% N) were applied according to treatments in two equal doses (1/2 at the time of planting and 1/2 at flowering stage), whereas the seaweed extracts was applied in four equal doses according to growth season peroid. The seeds of maize hybrid (Furat) were sown on 27 July 2019 by placing 3 seeds in the hill, and then thinning to a one plant after emergence. Crop management were carried out as needed, and the plants were harvested after the appearance of maturity signs.

Table (1): Physical and chemical properties of soil

Trait	Value	Unit
Sand	217	g Kg^{-1} soil
Loam	368	
Clay	415	
Bulk density	1.33	mg m^{-3}
pH 1:1	7.69	-----
Ec 1:1	2.53	ds m^{-1}
CEC	22.13	Cmol$_c$ Kg^{-1} soil
Ca^{2+}	15.24	Cmol$_c$ L^{-1}
Mg^{2+}	10.43	
Na$^+$	22.20	
K$^+$	1.58	
Cl$^-$	23.00	
SO$_4^{2-}$	15.10	
HCO$_3^-$	19.00	
CO$_3^{2-}$	Nill	
CaCO$_3$	255.00	g Kg^{-1} soil
O.M	4.65	%
Total nitrogen	0.40	
Available N	26.11	mg Kg^{-1} Soil
Available P	13.10	
Available K	137.00	
L-Aspergenase activity	36.20	µg N-NH$_4^+$ g^{-1} soil 2 h^{-1}

Table (2): Physical and chemical properties of vermicompost

Trait	Value	Unit
Moisture	26.40	%
Ec 1:5	1.68	ds m^{-1}
pH	6.20	-----
O.M	43.24	
O.C	25.14	%
N	1.50	
C:N ratio	16.76	-----
Fe	0.11	
Mn	0.68	
Zn	0.05	
B	0.29	mg Kg^{-1} Soil
Cu	0.33	
Co	0.04	
Ni	0.04	

L-Aspergenase enzyme activity (µg N-NH$_4^+$ g^{-1} soil 2h^{-1}) was estimated in the rhizosphere of maize during flowering and maturity stages according to Frankenberger and Tabatabai (1999), while its kinetic (K$_m$ and V$_{max}$) were estimated at the flowering and maturity stages by using six concentrations of L-aspargein acid (0.10, 0.25, 0.50, 0.75, 1.00 and 1.25) mmoler according to Senow and Tabatabai (1996) which reported in Hofstee (1952), and then K$_m$ and V$_{max}$ values were estimated according to modified Hanes-Woolf equation from Michaelis-Menten equation as follows:

$$S/V = {K_m}/{V_{max}} + 1/{V_{max}} \ [S]$$

As:

V = Reaction velocity

V_{max} = Maximum reaction velocity

K_m = Michaelis constant (moler L^{-1})

[S] = Substrate concentration (mmoler)

The recorded data were statistically analyzed according to the analysis of variance by using the Gnestat software. The least significant difference (LSD) was used to compare calculated average of studied traits (Suitover et al 2003).

Results and Methods

L-Aspergenase enzyme activity (µg N-NH$_4^+$ g^{-1} soil 2h^{-1})

The results at the tables 3 and 4 show that the application of vermicompost at level had 2 ton ha^{-1} (A1) gave the best results of L-Aspergenase enzyme activity (151.2 and 176.6 µg N-NH$_4^+$ g^{-1} soil 2h^{-1}) compared without application of vermicompost which gave the lowest means (79.9 and 85.9 µg N-NH$_4^+$ g^{-1} soil 2h^{-1}) at flowering and maturity stages respectively. The reason of increasing may be due to the physical and chemical properties of vermicompost (Table 2), or may be attributed to the increase soil's content of organic matter, which is an important source for energizing soil microorganisms, which increases its activity in secreting enzymes. These results are in line with the findings of Al-Taweel (2015) which indicated that the increasing of organic matter in the soil led to an increase in the enzymatic activity due to the increase in the activity of microorganisms at the rhizosphere, as the organic matter is a source of energy and food necessary for the growth of

microorganisms and increase their biomass and thus increase the amount of the secreted extracellular enzymes and their activity values,

L-Aspergenase enzyme activity was significantly affected by application of seaweed extracts (B1) and recorded the highest means (135.0 and 162.0 µg N-NH$_4^+$ g^{-1} soil 2h^{-1}) compared with control (B0) which recorded the lowest (116.0 and 126.7 µg N-NH$_4^+$ g^{-1} soil 2h^{-1}) at flowering and maturity stages respectively. The reason of the an increasing may be due to the role of organic matter in an increasing the numbers of microorganisms through its supply of nutrients and providing the appropriate medium for their growth and reproduction and then increasing their enzymatic secretions (Okabe et al 2012).

The application of 120 Kg N ha^{-1} (C1) was significantly superior and gave the highest means of L-Aspergenase enzyme activity (137.5 µg N-NH$_4^+$ g^{-1} soil 2h^{-1}) while the control (C0) gave the lowest (107.2 µg N-NH$_4^+$ g^{-1} soil 2h^{-1}) at flowering stage. The reason of the an increase may be attributed to the role of the nitrogen fertilizer in changing the physical, chemical and biological properties of soil and increasing the activity of the microorganisms responsible for the decomposition of organic matter and the positive reflection of that in an increasing the secreted enzymes such as amidohydrolases enzymes (Makinde et al 2010), while the nitrogen levels were non-significant difference at maturity stage.

The interaction between vermicompost and seaweed extract had significant effect on the L-Aspergenase enzyme activity, the A1B1 had the highest values (166.5 and 195.0 µg N-NH$_4^+$ g^{-1} soil 2h^{-1}) compared with A0B0 (60.3 and 43.4 µg N-NH$_4^+$ g^{-1} soil 2h^{-1}) at flowering and maturity stages respectively.

Also, The interaction between vermicompost and nitrogen fertilizer levels had significant effect on the L-Aspergenase enzyme activity, the A1C1 recorded the highest values (172.1 and 182.6 µg N-NH$_4^+$ g^{-1} soil 2h^{-1}) while the A0C0 recorded the lowest (55.5 and 74.6 µg N-NH$_4^+$ g^{-1} soil 2h^{-1}) at flowering and maturity stages respectively.

The interaction between application of seaweed extracts and nitrogen fertilizer levels had significant effect on the L-Aspergenase enzyme activity, the B1C1 and B1C0 gave the highest values (146.6 and 167.8 µg N-NH$_4^+$ g^{-1} soil 2h^{-1}) compared with B0C0 which gave the lowest (93.6 and 114.1 µg N-NH$_4^+$ g^{-1} soil 2h^{-1}) at flowering and maturity stages respectively.

The results at the tables 3 and 4 reveal that the interaction between three factors had significant effect on the L-Aspergenase enzyme activity, the A1B1C1 and A1B1C0 had the highest values (187.2 and 212.5 µg N-NH$_4^+$ g^{-1} soil 2h^{-1}) whereas while the A0B0C0 had the lowest (36.8 and 32.5 µg N-NH$_4^+$ g^{-1} soil 2h^{-1}) at flowering and maturity stages respectively. The results show that the L-Aspergenase enzyme activity values at the maturity stage were higher than of flowering stage in all tri-combination except the combination A0B0C0 which may be due to the depletion of nitrogen fertilizer by the plant and its entry into other pathways within the nitrogen cycle (Jansson and Person 1982), while organic fertilizers are slow to dissolve and the residual effect of organic fertilizer remains for a longer period than mineral fertilizers (Havlin et al 2005).

Table 3. Effect of vermicompost, seaweed extracts and nitrogen fertilizer on L-Aspergenase enzyme activity (µg $N-NH_4^+$ g^{-1} soil $2h^{-1}$) at floweing stage

Vermicompost levels (ton ha^{-1})	A0	A1	A2
	79.9	151.2	145.3
LSD .05	6.2		

Seaweed extracts (Kg ha^{-1})	B0		B1	
	116.0		135.0	
LSD .05	5.1			

Nitrogen levels (Kg N ha^{-1})	C0	C1	C2
	107.2	137.5	131.7
LSD .05	6.2		

Interaction A x B

	B0	B1
A0	60.3	99.5
A1	136.0	166.5
A2	151.8	138.9
LSD 0.05	8.8	

Interaction A x C

	C0	C1	C2
A0	55.5	78.1	106.1
A1	133.8	172.1	147.8
A2	132.4	162.4	141.2
LSD 0.05	10.8		

Interaction B x C

	C0	C1	C2
B0	93.6	128.4	126.0
B1	120.9	146.6	137.4
LSD 0.05	8.8		

Interaction A x B x C

		C0	C1	C2
A0	B0	36.8	52.4	91.7
	B1	74.1	103.9	120.6
A1	B0	108.8	156.9	142.1
	B1	158.8	187.2	153.4
A2	B0	135.3	176.0	144.1
	B1	129.6	148.7	138.3
LSD 0.05		15.3		

Table 4. Effect of vermicompost, seaweed extracts and nitrogen fertilizer on L-Aspergenase enzyme activity (μg N-NH$_4^+$ g^{-1} soil 2h^{-1}) at maturity stage

Vermicompost levels (ton ha^{-1})	A0	A1	A2
	85.9	176.6	170.6
LSD .05	7.4		

Seaweed extracts (Kg ha^{-1})	B0	B1
	126.7	162.0
LSD .05	6.0	

Nitrogen levels (Kg N ha^{-1})	C0	C1	C2
	140.9	148.2	144.0
LSD .05	N.S		

Interaction A x B	B0	B1
A0	43.4	128.5
A1	158.2	195.0
A2	178.6	162.6
LSD 0.05	10.5	

Interaction A x C	C0	C1	C2
A0	74.6	82.0	101.2
A1	174.9	182.6	172.3
A2	173.3	179.9	158.4
LSD 0.05	12.8		

Interaction B x C	C0	C1	C2
B0	114.1	136.7	129.4
B1	167.8	159.7	158.5
LSD 0.05	10.5		

Interaction A x B x C		C0	C1	C2
A0	B0	32.5	37.7	60.1
	B1	116.7	126.3	142.4
A1	B0	137.3	172.3	165.2
	B1	212.5	193.0	179.5
A2	B0	172.5	200.1	163.1
	B1	174.2	159.8	153.7
LSD 0.05		18.1		

K_m (moler L^{-1}) and V_{max} (μg N-NH$_4^+$ g^{-1} soil 2h^{-1})

The results at the tables 5, 6, 7 and 8 indicate that the application of vermicompost levels and seaweed extracts were non-significant difference in K_m and V_{max} values of L-Aspergenase enzyme at the flowering and maturity stages, also, the levels of nitrogen fertilizer were non-significant difference in K_m and V_{max} values of L-Aspergenase enzyme at the maturity stage only. However, the application of 120 Kg N ha^{-1} (C1) was significantly superior and gave the highest means of K_m (51.0 mmoler) and V_{max} (753 μg N-NH$_4^+$ g^{-1} soil 2h^{-1}) compared with control (C0) which gave the lowest (30.4 mmoler and 409 μg N-NH$_4^+$ g^{-1} soil 2h^{-1}) at flowering stage. The reason of the an increase may be attributed to superior same treatment in L-Aspergenase enzyme activity (Table 3), as the ease of the decomposition of urea and the release of the ammonium ion resulting from its

decomposition leads to an increase in the activity of biomass, which leads to an increase the enzymes activity and there kinetic measurement.

The interaction between vermicompost and seaweed extract had significant effect on the K_m values at flowering stage only (Table 5 and 7) and V_{max} at two stages (Table 6 and 8). The A2B1 had the highest values of K_m (65.2 mmoler) compared with A0B0 which had lowest (7.5 mmoler). Regarding the V_{max}, the A0B1 and A2B0 gave the highest values (753 and 136 µg N-NH_4^+ g^{-1} soil $2h^{-1}$) compared with A0B0 and A1B0 which gave the lowest (123 and 70 µg N-NH_4^+ g^{-1} soil $2h^{-1}$) at flowering and maturity stages respectively.

While the interaction between vermicompost and nitrogen fertilizer levels had non-significant effect on the Km and Vmax values at flowering and maturity stages respectively.

The interaction between application of seaweed extracts and nitrogen fertilizer levels had non-significant effect on the K_m values at flowering and maturity stages, whereas the interaction between two factors had significant effect on the Vmax at flowering stage only, the B1C1 recorded the highest values (970 µg N-NH_4^+ g^{-1} soil $2h^{-1}$) compared with B1C2 and B0C0 which recorded the lowest (260.0 and 325 µg N-NH_4^+ g^{-1} soil $2h^{-1}$) respectively.

The results at the tables 5, 6, 7 and 7 indicate that the interaction between three factors had significant effect on the K_m values at flowering stage only and V_{max} values at maturity stage only. Regarding K_m values, the A1B1C1 had the highest values (105.5 mmoler) whereas while the A0B0C1 had the lowest (14.6 mmoler). While V_{max} values, the A2B0C0 gave the highest value (199 µg N-NH_4^+ g^{-1} soil $2h^{-1}$) compared with A0B0C0 which gave the lowest (27 µg N-NH_4^+ g^{-1} soil $2h^{-1}$).

The relationship between the concentrations of the substrate (Asparagine) and the velocity of its degradation for the study treatments is a positive relationship, as Figures 1 and 2 show that an increase in the concentration of the substrate led to an increase the velocity of its degradation (L-Aspergenase enzyme activity) at the flowering and maturity stages respectively to a certain extent and after that any increase in the concentration of the substrate was not accompanied by an increase in the activity of the enzymes, as the reaction follows the reaction of the first order to the point of the concentration at which the maximum activity (V_{max}), followed by the reaction after the order of zero (Frankber and Tabatabai 1991). This may be due to the fact that the substrate molecules may combine with the enzyme to the extent that these sites are saturated and then the activity of the enzyme remains constant, such that any increase in the concentration of the substrate does not lead to an increase in the activity (Al-Muzaffar 1983). It is also noted from Figures 1 and 2 that the rate of degradation of the substrate differed between the study treatments because vermicompost has a role in increasing the readiness of nutrients and then increasing the absorption of NPK, which is positively reflected on plant growth and then the increase in the activity and growth of the root system and the increase in biomass activity (Al-Shammari and Daoud 2011). Regarding the seaweed extracts, its contain some vitamins, hormones and amino acids which plays the role of the plant growth and then increase the effectiveness of the roots and the activity of microorganisms (Jasim et al 2006). In the same direction, Burns (1986) explained that the higher K_m value, the affinity constant between the enzyme and the substrate, the weaker the affinity between the enzyme and substrate and this is due to the strong blocking of the enzyme by the soil components.

Table 5. Effect of vermicompost, seaweed extracts and nitrogen fertilizer on K_m of L-Aspergenase enzyme activity (mmoler) at flowering stage

Vermicompost levels (ton ha^{-1})		A0	A1	A2
		31.4	32.2	47.5
LSD .05			N.S	
Seaweed extracts (Kg ha^{-1})		B0		B1
		30.4		44.9
LSD .05			N.S	
Nitrogen levels (Kg N ha^{-1})		C0	C1	C2
		30.4	51.4	31.1
LSD .05			21.4	
Interaction A x B				
		B0		B1
A0		17.9		44.8
A1		43.6		24.7
A2		29.7		65.2
LSD 0.05			35.3	
Interaction A x C				
		C0	C1	C2
A0		23.4	45.2	25.4
A1		22.4	43.9	36.3
A2		45.5	65.1	31.7
LSD 0.05			N.S	
Interaction B x C				
		C0	C1	C2
B0		21.5	35.2	34.6
B1		39.4	67.7	27.7
LSD 0.05			N.S	
Interaction A x B x C				
		C0	C1	C2
A0	**B0**	24.4	14.6	14.6
	B1	22.4	75.8	36.2
A1	**B0**	15.3	66.1	49.5
	B1	29.5	21.7	23.0
A2	**B0**	24.8	24.8	39.6
	B1	66.2	105.5	23.8
LSD 0.05			43.8	

Table 6. Effect of vermicompost, seaweed extracts and nitrogen fertilizer on V_{max} of L-Aspergenase enzyme (µg $N-NH_4^+$ g^{-1} soil $2h^{-1}$) at flowering stage

Vermicompost levels (ton ha^{-1})	A0		A1		A2
	438		495		668
LSD .05	N.S				
Seaweed extracts (Kg ha^{-1})	B0			B1	
	491			574	
LSD .05	N.S				
Nitrogen levels (Kg N ha^{-1})	C0		C1		C2
	409		753		437
LSD .05	264				

Interaction A x B			
	B0		B1
A0	123		753
A1	743		246
A2	609		724
LSD 0.05	373		

Interaction A x C			
	C0	C1	C2
A0	311	732	271
A1	223	585	675
A2	692	941	365
LSD 0.05	N.S		

Interaction B x C			
	C0	C1	C2
B0	325	535	614
B1	492	970	260
LSD 0.05	373		

Interaction A x B x C				
		C0	C1	C2
A0	B0	88	143	137
	B1	533	1322	404
A1	B0	243	819	1167
	B1	203	351	184
A2	B0	644	644	539
	B1	741	1238	192
LSD 0.05		N.S		

Table 7. Effect of vermicompost, seaweed extracts and nitrogen fertilizer on K_m of L-Aspergenase enzyme activity (mmoler) at maturity stage

Vermicompost levels (ton ha^{-1})		A0		A1		A2
		14.6		14.0		12.0
LSD .05		N.S				
Seaweed extracts (Kg ha^{-1})		B0			B1	
		13.9			13.2	
LSD .05		N.S				
Nitrogen levels (Kg N ha^{-1})		C0		C1		C2
		14.6		12.5		13.5
LSD .05		N.S				
Interaction A x B						
		B0			B1	
A0		14.8			14.5	
A1		13.3			14.7	
A2		13.4			10.5	
LSD 0.05		N.S				
Interaction A x C						
		C0		C1		C2
A0		11.7		15.2		17.0
A1		17.2		13.4		11.4
A2		14.9		9.0		12.1
LSD 0.05		N.S				
Interaction B x C						
		C0		C1		C2
B0		13.4		13.1		15.1
B1		15.9		12.0		11.8
LSD 0.05		N.S				
Interaction A x B x C						
		C0		C1		C2
A0	B0	8.6		16.2		19.6
	B1	14.9		14.1		14.4
A1	B0	16.1		11.1		12.9
	B1	18.4		15.8		9.9
A2	B0	15.5		11.9		12.9
	B1	14.4		6.0		11.2
LSD 0.05		N.S				

Table 8. Effect of vermicompost, seaweed extracts and nitrogen fertilizer on V_{max} of L-Aspergenase enzyme (µg N-NH$_4^+$ g^{-1} soil 2h^{-1}) at maturity stage

| Vermicompost levels (ton ha^{-1})		A0	A1	A2
		93	99	118
LSD .05		N.S		
Seaweed extracts (Kg ha^{-1})		B0		B1
		95		111
LSD .05		N.S		
Nitrogen levels (Kg N ha^{-1})		C0	C1	C2
		124	91	95
LSD .05		N.S		
Interaction A x B				
		B0		B1
A0		80		105
A1		70		128
A2		136		99
LSD 0.05		52		
Interaction A x C				
		C0	C1	C2
A0		96	104	78
A1		118	79	100
A2		157	89	106
LSD 0.05		N.S		
Interaction B x C				
		C0	C1	C2
B0		98	93	95
B1		150	88	95
LSD 0.05		N.S		
Interaction A x B x C				
		C0	C1	C2
A0	B0	27	134	78
	B1	166	73	77
A1	B0	69	38	103
	B1	168	120	98
A2	B0	199	107	103
	B1	1166	72	110
LSD 0.05		90		
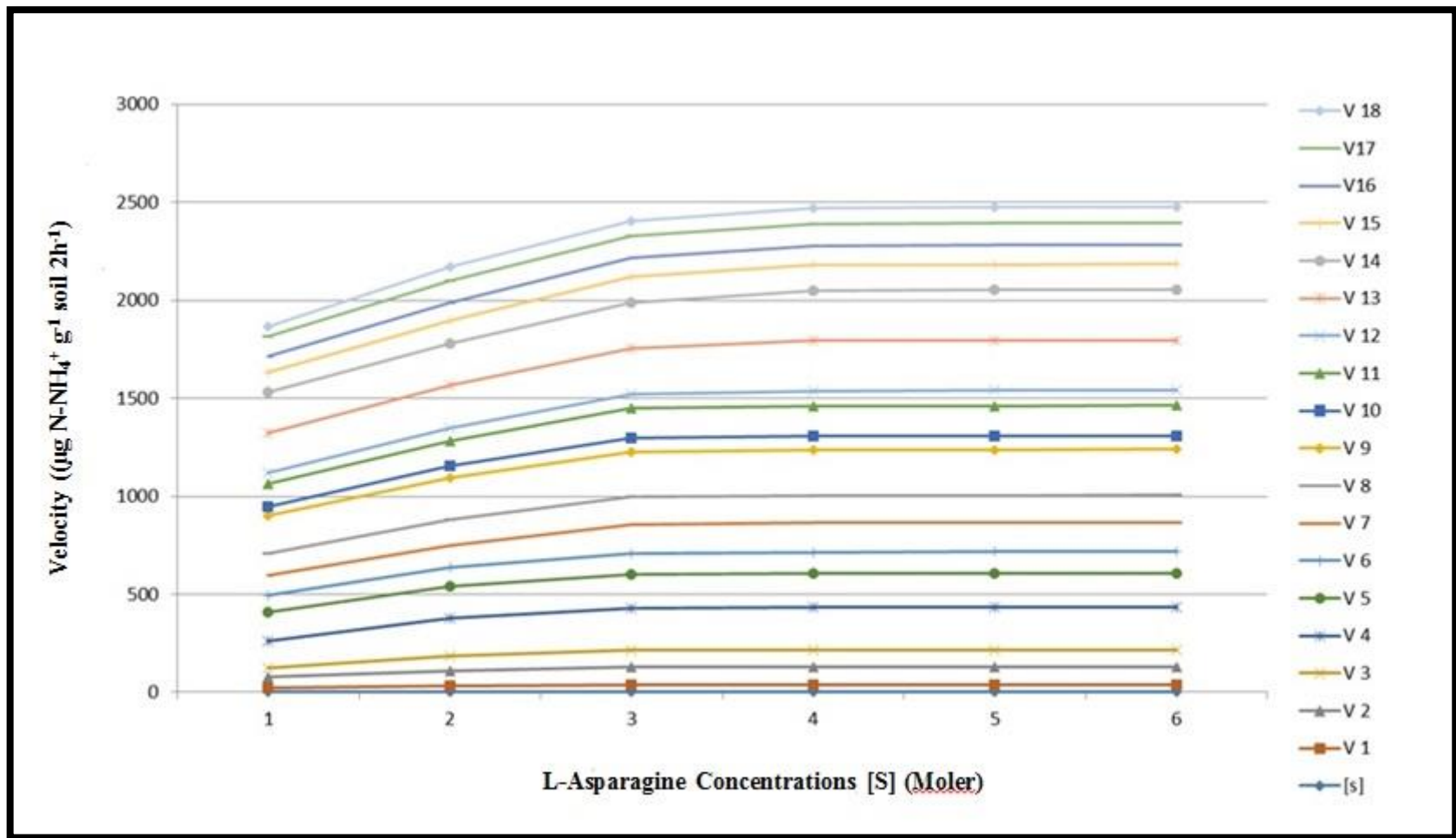
Fig (1): Relationship between L-Asparagine Concentrations [S] and its degradation velocity (V) at flowering stage
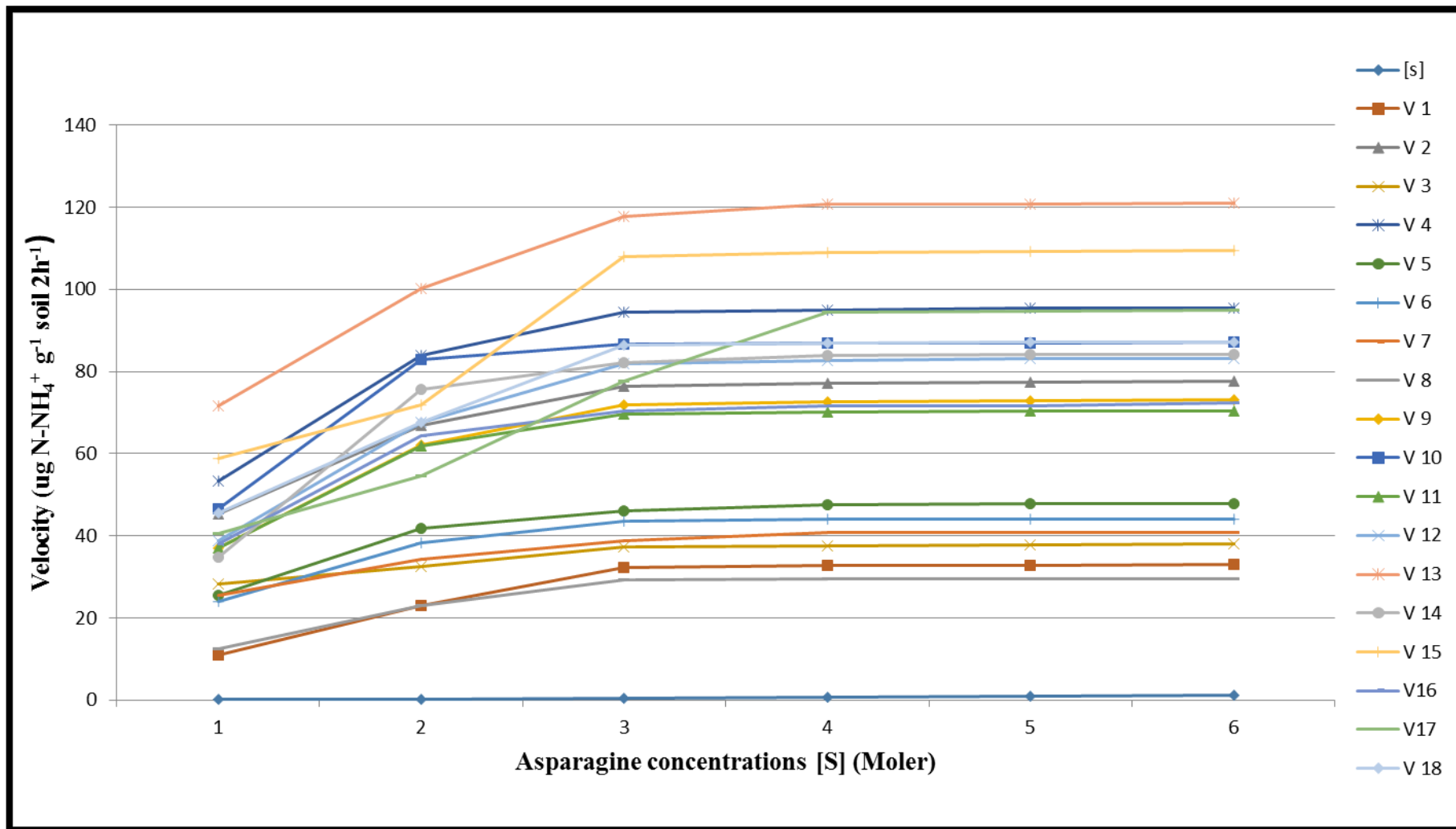
Fig (2): Relationship between L-Asparagine Concentrations [S] and its degradation velocity (V) at maturity stage

References