# Effect of Nano-Fertilizers and DAP on the Activity of the Alkaline Phosphatase Enzyme in the Rhizosphere Soil and its Outside for the Rice Plant (*Oryza Sativa*)

### Luma Salih Jabbar AL-Taweel and Mustafa Abdul Sada Hassan Al-Karawi

Department of Soil Sciences and Water Resources, College of Agriculture, Al-Qadisiyah University, Iraq

Emails: Luma.altaweel@qu.edu.iq, Mustfaabd308@gmail.com

#### Abstract

The experiment was conducted with the aim of studying the effect of adding different sources of DAP fertilizers and integrated nano silicon and nano fertilizers on the effectiveness of the alkaline phosphatase enzyme in the Rhizosphere area and outside it, and for the rice plant of the Amber 33 cultivar, where the experiment was conducted in one of the rice fields located in Ghamas district / Al-Diwaniyah province, Iraq in summer season of 2019 in clay loam soil, The rice seeds were cultivated (Amber 33 cultivar), the total field area was 350 m2, and the experiment treatments were two levels of DAP fertilizer (Control, M-DAP, O-DAP and O-DAP High K) were (0.240) kg h<sup>-1</sup> and two levels of fertilizer. Nano silicon (0.2) ml.L<sup>-1</sup> and two levels of integrated nano fertilizer (0, 2)g.L<sup>-1</sup> in addition to the interactions between the treatments. The experiment was conducted according Randomized Complete Block Design (RCBD) with three replicates for each treatment. The averages were compared according to the L.S.D test at a probability level (0.05). Then the effectiveness of the alkaline phosphatase enzyme in and outside the rhizosphere was estimated after 35, 65 and 95 of cultivation. and the results of laboratory tests showed that O-DAP was significantly excelled on the other two types of DAP fertilizer in recording the highest averages of the effectiveness of the alkaline phosphatase enzyme in the rhizosphere soil and outside it, after 95 days. From cultivation as it reached (289.4 and 257.9) µg p-nitro phenol g<sup>-1</sup> soil 1h<sup>-1</sup> Respectively, The treatment of organic DAP fertilizer O-DAP High K, compared to the other two types of DAP fertilizer, achieved the highest significant averages of the effectiveness of the alkaline phosphatase enzyme after (35 and 65) days of cultivation in the rhizosphere and outside it, reaching (182.6 and 197.3) and (224.0 and 199.4)  $\mu$ g p-nitro phenol g<sup>-1</sup> soil 1h<sup>-1</sup> respectively. Nano silicon fertilizer NS achieved the highest average of measuring the effectiveness of alkaline phosphatase after 95 days of cultivation in soil outside the rhizosphere, it reached (241.4)  $\mu$ g p-nitro phenol g<sup>-1</sup> soil 1h<sup>-1</sup>, and after 35 days after planting it was (178.9 and 168.9)  $\mu$ g p-nitro phenol g<sup>-1</sup> soil 1h<sup>-1</sup> for both areas. Integrated nano fertilizer NC achieved the highest average in measuring the effectiveness of the alkaline phosphatase enzyme after 95 days of cultivation in soil outside the rhizosphere and it reached (241.0)  $\mu$ g p-nitro phenol g<sup>-1</sup> soil 1h<sup>-1</sup> while it was recorded after 65 days of cultivation of average. (232.7 and 205.7)  $\mu$ g p-nitro phenol g<sup>-1</sup> soil 1h<sup>-1</sup> for both area. Triple interaction between DAP, NS and NC recorded the highest mean alkaline phosphatase activity after 95 days of cultivation. Where O-DAP + Cont NS + Cont NC recorded the highest mean (355.9 and 295.5) µg p-nitro phenol .g<sup>-1</sup> soil.2h<sup>-1</sup> after 65 days of cultivation. The treatment of O-DAP High K + NS + NC in the Rhizosphere soil and M-DAP + NS + NC outside it achieved a mean of (259.4 and 248.8) and after 35 days of cultivation. The treatment of NC + NS + O-DAP in and outside the rhizosphere achieved the treatment of O-DAP High K + NS + a mean of (220.5 and 210.7) µg pnitro phenol  $g^{-1}$  soil  $1h^{-1}$  respectively.

Keywords: complete DAP, nano silicon fertilizer, enzyme urease, rice, Oryzae Sativa

# Introduction

The alkaline phosphatase enzyme is secreted by plants, microorganisms, and animals in the soil (Banerjee and Sanyal 2012, Tabatabai and Dick, 1984). The alkaline phosphatase enzyme participates in the phosphorous cycle in the soil and detects the behavior of phosphorous solvent in the soil (Nannipieri et al., 2011). Nannipieri, (2011) showed that the alkaline phosphatase enzyme is abundantly secreted in neutral or alkaline soils, and the enzyme is stabilized by organic colloids and trapped by humus molecules (Burns, 2013). There are a number of factors that affect the activity of the enzyme, including organic matter, temperature, clay content, vegetation, soil, climate factors, and ecosystem functions (Benitez et al., 2006). The temperature and humidity indirectly affect the enzyme activity through the growth of microorganisms, and the availability of the substrate and many other factors such as microorganism groups, organic carbon in the soil, phosphorous concentrations in addition to the C: N: P ratios of organic matter also have an effect. on the activity of enzymes (Hui, 2013). Mineral and organic fertilization affect the activity of phosphatase enzyme in the soil. (Guan ,1989) found an increase in the activity of the enzyme phosphatase after adding organic wastes to the soil.(Parham et al., 2002) demonstrated the higher biomass and the effectiveness of the alkaline phosphatase enzyme in the soil when treated with livestock waste compared to soil treated with chemical fertilizer. Because of the importance of this enzyme in the Mineralization of organic phosphorous in the soil and its important role in plant nutrition, it has been studied in the soil and that the importance of the enzyme in plant nutrition comes from its high efficacy in the Rhizosphere compared to the Bulk Soil area(Tarafdar and Bala, 1988).(Yosefi, 2011) showed that the possibility of distinguishing between bio mineralization and Biochemical mineralization of the alkaline phosphatase enzyme, but it is difficult to distinguish between the alkaline phosphatase enzyme secreted from plant roots and secreted by the phosphorous dissolving microorganisms. Zhang et al.,( 2020) noted a significant decrease in the activity values of the acid and alkaline phosphatase enzyme with an increase in soil depth from 0-10 cm to 10-20 cm, and they explained that decrease to a decrease in soil content of organic matter with an increase in depth and its association with the number and activity of microorganisms. (Read, 2002) stated that the plant liberates phosphorous through the secretion of organic acids in the root zone, and by alternating oxidation processes have clear effects on the transformation of the mineral phosphorus in soil, and the role of microorganisms in the soil has been reduced to the availability of phosphorus. However, the presence of phospholipid microorganisms (PSMs) and phosphatase activity in the rice rhizosphere indicate the potential role of PSMs in the growth and yield of rice (Adhya, et al.2015; Elizabeth, et al.2017). Diammonium phosphate is the most widely used phosphate fertilizer in the United States. It contains 18 N% and P% 20-23, which is equivalent to 36-53% P<sub>2</sub>O<sub>5</sub>. This fertilizer was prepared from ammonia treatment with phosphoric acid. The effect of this fertilizer is alkaline at the beginning (pH is more than 8), but its effect is acidic at the end because it contains the ammonium ion, where the ammonium ions are formed, which at the end are converted into nitrates by the nitrification process and the pH is reduced (Ashok, 2015). According to the study by (Peng. et al., 2015) they showed that the nano fertilizers added to the soil lead to a decrease in the biomass as well as a decrease in the activity of enzymes in the flooded rice soils. Zinc

fertilizer (ZnO) and  $CeO_2$  nanoparticles affect the number of Azotobacter bacteria that dissolve phosphorous and potassium and inhibit the activity of soil enzymes (Chai et al., 2015).

## Materials and Methods

The experiment was conducted in one of the fields located in Ghamas district ,Al-Qadisiyah province, which reached an area of 360 m2 during the summer season of 2019 in soil, and some of its chemical, physical and biological properties are explained in Table (1), In aim to study the effect of nano fertilizers and DAP on the activity of the alkaline phosphatase enzyme in the rhizosphere soil and outside it for the rice plant, cultivar Amber 33.

Traits		Values	units	References	
pH		7.53		1965b) (Black	
EC <sub>1:1</sub>		4.96	ds.m <sup>-1</sup>	17050)•(Diack	
	Ca <sup>+2</sup>	30.0			
	$Mg^{+2}$	13.8			
Nagating and	Na <sup>+1</sup>	8.12			
Negative and	K <sup>+1</sup>	4.30	- Cmol <sub>c</sub> .L <sup>-1</sup>	(1065  (Plack))	
positive soluble ions	Cl <sup>-1</sup>	24.09	- Cmol <sub>c</sub> ,L	(1965 <sub>a</sub> ·Black)	
soluble lons	HCO <sub>3</sub>	2.7			
	$SO_4^{-2}$	19.12			
	CO3 <sup>-2</sup>	Nill			
CaCO		32.87	g.kg <sup>-1</sup> soil	1965b)،(Black	
Organic mat	ter O.M		g.kg som		
CEC		27.76	Cmol <sub>c</sub> .kg <sup>-1</sup> soil	1976). Papanicolaou)	
available ni	trogen	15.00			
available pho	sphorus	10.25	mg.kg <sup>-1</sup> soil	1982) · (Page	
available Pot	assium	251.3			
Soil	sand	308			
separators	Silt	388	g.kg <sup>-1</sup> soil		
separators	Clay	304			
Soil texture		silty loam		1965 <sub>b</sub> ). (Black	
Bulk density		1.53	Mg.m <sup>-3</sup>		
Total bac	teria	25.81x10 <sup>6</sup>	- CFU.g <sup>-1</sup> dry soil		
Total fu	Total fungi				
The effectiven	ess of the	112	μg p-nitro phenol .g <sup>-1</sup>	Tabatabai) و	
enzyme phos	phatase	112	soil.2h <sup>-1</sup>	(1972 · <b>Bremner</b>	

Table (1) Some physical, chemical and biological traits for the study soil

The experiment was conducted after distributing the 16 treatments Table (2) on the divided plots according to the Randomized Complete Block Design (RCBD) and with three replications. The

treatments and their replicates resulted in 48 experimental units, including the control treatment. Dry Rice seeds (cultivar Amber 33.) obtained from the Rice Research Station in Al-Mishkhab district / Najaf province, were sown with an amount of 120 kg.ha<sup>-1</sup> seeds by hand prose. The urea nitrogen fertilizer was added to all the experimental units to ensure a balanced growth of the plants in all the experimental units by 300 kg.ha<sup>-1</sup>.It was added in three batches, the first after a month of cultivated, (25%) of the recommendation, and the second in the tillering stage .Where (50%) was added, and the third batch was added at the beginning of the booting stage, Where the last batch of the recommendation was added. The integrated NC nanoparticles and nano silicon NS were added in two batches, the first at the elongation stage and the second in tillering stage. The mineral DAP(M-DAP) fertilizer, which contains (18%) nitrogen and (46%) phosphorous, was added in two batches, the first at the vegetative growth stage and the second in the tillering stage. As for the organic fertilizer O-DAP fortified with macro elements, which contains (16.5%) nitrogen, (48.2%) phosphorous and (2.14%) potassium. Two batches were added, the first in the vegetative growth stage and the second at the branching phase, as for O-DAP High K organic fertilizer containing (18%) nitrogen and (7%) phosphorous and (46%) potassium added in two batches, the first in the vegetative growth And the second in the tillering stage. The activity of the alkaline phosphatase enzyme was estimated in the rhizosphere soil and outside of the rice plant after (35,65 and 95) days from cultivation.

No.	Treatments	treatments symbol with its fertilizer recommendation		
1	Control	Without adding		
2	Nano Silicon	NS (2 ml.L <sup>-1</sup> )		
3	Nano Complete	NC $(2 \text{ g.L}^{-1})$		
4	Mineral –DAP	M-DAP (240 kg.ha <sup>-1</sup> )		
5	Organic-DAP	O-DAP (240 kg.ha <sup>-1</sup> )		
6	Organic DAP High K	O-DAP high-K (240 kg.ha <sup>-1</sup> )		
7	Nano Silicon +Nano complete	NS $(2 \text{ ml.L}^{-1}) + \text{NC} (2 \text{ g.L}^{-1})$		
8	Mineral –DAP+ Nano Silicon	M-DAP (240 kg.ha-1) +NS (2 ml.L <sup>-1</sup> )		
9	Mineral –DAP+ Nano complete	M-DAP (240 kg.ha <sup>-1</sup> ) + NC (2 g.L <sup>-1</sup> )		
10	Organic-DAP + Nano Silicon	O-DAP (240 kg.ha-1) + NS 2 ml.L <sup>-1</sup> )		
11	Organic-DAP + Nano complete	O-DAP (240 kg.ha <sup>-1</sup> ) + NS 2 (ml.L <sup>-1</sup> ) + NC (2g.L <sup>-1</sup> )		
12	Organic DAP High K+ Nano Silicon	(240kg.Ha <sup>-1</sup> ) O-DAP high-K NS + 2ml.L <sup>-1</sup> )		

 Table (2) Distribution of experiment treatments with symbols and their fertilizer recommendation

13	Organic DAP High K+ Nano	O-DAP high-K (240 kg.ha <sup>-1</sup> )
15	complete	$NS + 2 ml.L^{-1}$ )
14	Mineral –DAP+ Nano Silicon	M-DAP (240 kg.ha-1) + NS
14	+ Nano complete	$(2ml.L^{-1}) + NC (2g.L^{-1})$
15	<b>Organic-DAP + Nano + Silicon</b>	<b>O-DAP (240 kg.ha<sup>-1</sup>) + NS</b>
15	Nano complete	$(2ml.L^{-1}) + (NC (2g.L^{-1}))$
16	Organic DAP High K+Nano	<b>O-DAP high-K (240 kg.ha<sup>-1</sup>) +</b>
10	Silicon + Nano complete	NS $(2 \text{ ml.L}^{-1}) + \text{NC} (2g.L^{-1})$

The activity of the alkaline phosphatase enzyme was estimated in the rhizosphere soil and outside of the rice plant after (35,65 and 95) days from cultivation. The activity of the alkaline phosphatase enzyme was determined according to the method of Eivazi (Tabatabai 1977) by placing 1 g of soil in a 50 ml volumetric flask containing 0.2 ml of coloring and 4 ml of the Modified Universal Buffer (MUB) (boric acid, citric acid, Malic acid, sodium hydroxide and THAM, With pH = 11, 1 ml of p-nitro phenyl phosphate was added to it as a Substrate to the enzyme, the soil is then incubated at a temperature of 37 ° C for a period of one hour, After incubation, add 1 ml of a 0.5 M potassium chloride solution and 4 ml of 0.5 M NaOH. The soil suspension was then filtered and the enzyme activity was estimated with the amount of released P-nitro phenol, measured by a spectrophotometer at a wavelength of 420 nm.

### **Results and discussion**

The effect of DAP fertilizer and Integrated nano silicon and nano fertilizer on the effectiveness of the  $\mu$ g p-nitro phenol g<sup>-1</sup> soil2h<sup>-1</sup> enzyme in the rhizosphere and outside the rhizosphere of the rice plant after (35, 65 and 95) days after cultivation. The results in tables (3 and 4) indicate the presence of significant differences in the levels of DAP fertilizer (Control, M-DAP, O-DAP, O-DAP High K) after 35 days of cultivation in the activity of the alkaline phosphatase enzyme in two regions. Where the O-DAP High K treatment recorded the highest average (182.66 and 197.36) µg p-nitro phenol g<sup>-1</sup> soil 1h<sup>-1</sup> compared to the control treatment C which gave the lowest average (161.97 and 131.92) µg p-nitro phenol g<sup>-1</sup> soil 1h<sup>-1</sup>. This is due to the enzyme phosphatase secreted by microorganisms or endemic soil animals, as well as plant roots, where it operates a dual function, which is to break down organic matter into simpler forms and then acquire sources for the producing enzymes (Sinsabaugh and Moorhead, 1994). The results in Tables (3 and 4) show that there is a significant difference in the treatment of NS fertilizer in the average activity of the alkaline phosphatase enzyme in and outside the rhizosphere. The NS treatment gave the highest average of (178.97 and 168.93)  $\mu$ g p-nitro phenol g<sup>-1</sup> soil 1h<sup>-1</sup> By compared with the control treatment Cont NS, which gave the lowest mean (158.02 and 145.55) µg pnitro phenol g<sup>-1</sup> soil 12h<sup>-1</sup>, The reason for the significantly excelled of NS treatment is that the addition of silicates to the soil leads to an increase in the availability of phosphorous in the soil, where silicates replace the reciprocal phosphates and lead to the release of phosphorous and from the absorption surfaces and thus increase its availability in the soil (Giraldo et al., 2014). The results of the two tables showed that there was no significant difference in the treatment of NC integrated nano fertilizer in the average activity of the alkaline phosphatase enzyme in the rhizosphere and outside it. The results of the two tables show that there are significant differences in the

treatment of the bi-interaction between DAP fertilizer and NS in the average effectiveness of alkaline phosphatase, as the O-DAP treatment gave High K + Cont NS in the rhizosphere, while the treatment of O-DAP High K + NS in soil outside the rhizosphere was the highest average. (201.82 and 207.31) µg p-nitro phenol g<sup>-1</sup> soil 1h<sup>-1</sup> compared with DAP Cont and Cont NS treatment, which gave the lowest mean (123.93 and 105.94)  $\mu$ g p-nitro phenol g<sup>-1</sup> soil 1h<sup>-1</sup> respectively. The results in tables (3 and 4) also showed that there were significant differences in the treatment of the bi- interaction between DAP fertilizer and NC fertilizer in the average effectiveness of alkaline phosphatase in the rhizosphere area and outside it for the rice plant. The treatment of O-DAP High K + NC and O-DAP High K + Cont NC gave the highest mean of (190.00 and 202.33)  $\mu$ g p-nitro phenol g<sup>-1</sup> soil 1h<sup>-1</sup> respectively compared to the treatment of DAP Cont and Cont NC that gave The lowest average was (148.16 and 134.52) µg pnitro phenol g<sup>-1</sup> soil 1h<sup>-1</sup>. The results in the two tables show that there are significant differences in the treatment of the bi-interaction between the NS fertilizer treatment and the NC fertilizer treatment in the average activity of the alkaline phosphatase enzyme in the rhizosphere area and outside it for the rice plant., where the NS and NC treatment gave the highest average of (186.29 and 177.77) µg p-nitro phenol g<sup>-1</sup> soil 1h<sup>-1</sup> respectively compared to the Cont NS and Cont NC treatment which gave the lowest mean (161.68 and 150.05) µg p-nitro phenol g<sup>-1</sup> soil 1h<sup>-1</sup> respectively. The results in the two tables showed significant differences in the triple interaction treatment between DAP fertilizer, NS fertilizer treatment and NC fertilizer in the average activity of the alkaline phosphatase enzyme in the rhizosphere and outside it, for the rice plant. The addition of NC + NS + O-DAP and + NS + O-DAP High K NC gave the highest mean (220.54 and 210.72) µg p-nitro phenol g<sup>-1</sup> soil 1h<sup>-1</sup> compared with DAP Cont and Cont NS. Cont NC which gave the lowest mean (116.68 and 97.64) µg p-nitro phenol g <sup>1</sup> soil 1h<sup>-1</sup> respectively.

Table (3) The effect of DAP fertilizer and integrated silicon and nanostructures on the activity of g alkaline phosphatase enzyme p-nitro phenol g<sup>-1</sup> soil 1h<sup>-1</sup> in the rhizosphere area for Rice plant after 35 days of cultivation

DAP adding levels	DAP Cont	M-DAP	O-DAP	O-DAP High K	
	161.97	162.82	166.54	182.66	
L.S.D		10	.12	·	
NS adding levels	Co	ntNS		NS	
	15	8.02	1	78.97	
L.S.D		7.16			
NC adding levels	Cor	nt NC	NC		
	16	6.74	170.24		
L.S.D		Ν	. <b>C</b>		
·	Bi-intera	action between DAP	and NS		
	Cont DAP	M-DAP	O-DAP	O-DAP High K	
NS Cont	123.93	160.12	146.02	201.82	
NS	199.83	165.61	186.94	163.51	
L.S.D	14.32				

		Bi-interac	ction between DAP	and NC	
		Cont DAP	M-DAP	O-DAP	O-DAP High K
NC	Cont	148.16	166.88	146.34	175.27
	NC	175.79	158.96	183.77	190.00
L	.S.D		14	.32	
		Bi-intera	ction between NS a	nd NC	
		NS C	Cont		NS
NC	Cont	161.84		1	54.17
	NC	171.66		186.29	
L	.S.D		10	.12	
		Triple interact	tion between DAP,	NS, and NC	
		Cont DAP	M-DAP	O-DAP	O-DAP High K
Cont	Cont NC	116.68	167.65	145.24	217.96
NS	NC	131.33	152.56	146.92	185.75
NC	Cont NC	234.82	165.95	153.45	132.55
NS	NC	164.84	165.25	220.54	194.46
L	.S.D		20	.25	•

Table (4) The effect of DAP fertilizer and integrated silicon and nanostructures on the activity of g alkaline phosphatase enzyme  $\mu$ g p-nitro phenol g<sup>-1</sup> soil 1h<sup>-1</sup> in the soil of an area outside the rhizosphere for rice plants after 65 days of cultivation

DAP adding levels	DAP Cont	M-DAP	O-DAP	O-DAP High K		
	131.92	145.64	153.98	197.36		
L.S.D	9.59					
NS adding levels	Cor	ntNS		NS		
	14:	5.55	1	68.93		
L.S.D		6.	78			
NC adding levels	Con	it NC		NC		
	155.15 159.32			59.32		
L.S.D		Ν	. <b>C</b>			
	Bi-intera	ction between DAP	and NS			
	Cont DAP	M-DAP	O-DAP	O-DAP High K		
NS Cont	105.94	140.39	148.22	187.44		
NS	157.82	150.93	159.74	207.31		
L.S.D	13.57					
	Bi-interaction between DAP and NC					
	Cont DAP	M-DAP	O-DAP	O-DAP High K		
NC Cont	134.52	132.72	150.96	202.33		

]	NC	129.22	158.61	157.04	192.48		
L	.S.D	13.57					
		Bi-intera	ction between NS a	and NC			
	NS Cont NS						
NC	Cont	150	.05	1	60.24		
]	NC	140	.93	1	77.72		
L	.S.D		10	10.12			
		Triple interact	tion between DAP,	NS, and NC			
		Cont DAP	M-DAP	O-DAP	O-DAP High K		
Cont	Cont NC	97.64	142.23	159.44	200.75		
NS	NC	114.38	138.58	136.92	174.12		
NC	Cont NC	171.59	123.16	142.39	203.83		
NS	NC	144.17	178.73	177.14	210.72		
L	.S.D		19	.18	•		

The results of the statistical analysis in Table (5 and 6)indicate that the addition of DAP fertilizer (Control, M-DAP, O-DAP, O-DAP High K) led to significant differences after 65 days of cultivation in average of activity values of the enzyme alkaline phosphatase in the rhizosphere and outside it for the rice plant, where the treatment of O-DAP High K in the rhizosphere and M-DAP outside it recorded the highest average of (224.02 and 207.82 ) $\mu$ g p-nitro phenol g<sup>-1</sup> soil1 h<sup>-1</sup> Compared to the control treatment Cont DAP, which gave the lowest average of the efficacy values (188.53 and 190.54) µg pnitro phenol g<sup>-1</sup> soil 1h<sup>-1</sup>. The reason for the treatment of O-DAP High K and M-DAP significantly excelled on the rest of the treatments may be that the DAP mineral fertilizer will provide the living with two important elements of the macro elements, namely nitrogen and phosphorous, As these organisms need these two elements to sustain their vital activities, including their secretion of enzymes (Banerjee and Sanyal, 2012), Organic acids have a role in improving the soil's chemical and physical properties, significantly increasing biomass and providing nutrients to the plant (Muter et al., 2015). The results in the two tables showed that there was no significant difference in the treatment of NS fertilizer in the mean values of the activity of the alkaline phosphatase enzyme in the rhizosphere and outside it for the rice plant. The results in the two tables show that there is a significant difference in the treatment of NC fertilizer in the average activity of the alkaline phosphatase enzyme in the rhizosphere and outside it for the rice plant, where the NC recorded the highest average of (232.79 and 205.73) µg p-nitro phenol g<sup>-1</sup> soil 1h<sup>-1</sup>, respectively. comparison with Cont NC treatment, which gave an average of (191.54 and 188.52)  $\mu$ g p-nitro phenol g<sup>-1</sup> soil 1h<sup>-1</sup>. The reason for the significant of the NC treatment is that the nanoparticles, with their traits such as the high surface area, lead to an increase in the adsorbed quantities of enzymes on their surfaces and thus increase their effectiveness (Prasad et al., 2016). The integrated nano fertilizer contains the Macro and Micro elements in appropriate quantities that increased the enzyme's effectiveness, where boron is one of the small elements that activate the work of enzymes and form proteins, and manganese works to bind the enzyme to the substrate (the alkaline

material) (AZIZ et al., 2018b). The results in Table (6) showed that there were significant differences in the treatment of the bi-interaction between DAP fertilizer and NS in the average activity of the alkaline phosphatase enzyme in an area outside the rhizosphere, where the M-DAP + NS treatment recorded the highest average of (220.92)  $\mu$ g p-nitro phenol g<sup>-1</sup> soil 1h<sup>-1</sup> compared to the treatment of DAP Cont and Cont NS, which gave the lowest average of the efficacy values of (180.62) µg p-nitro phenol g<sup>-1</sup> soil 1h<sup>-1</sup>, while the mentioned treatment did not record a significant difference in the rhizosphere, Table (5). The results in the two tables show that there are significant differences in the treatment of the bi-interaction between the DAP fertilizer and the NC in the values of the activity of the alkaline phosphatase enzyme in the rhizosphere. The treatment of O-DAP High K + NC was recorded and outside it, the treatment of Cont DAP + NC recorded a mean of (244.26 and 222.11) µg p-nitro phenol g<sup>-1</sup> soil 1h<sup>-1</sup>respectively as compared to the control treatment for DAP Cont and Cont NC which gave an average (148.84 and 158.91)  $\mu$ g p-nitro phenol g<sup>-1</sup> soil 1h<sup>-1</sup> respectively. The results in the table show that there were significant differences in the triple interaction treatment between DAP, NS and NC fertilizers in the values of the alkaline phosphatase enzyme activity in the rhizosphere. The treatment was recorded as O-DAP High K + NS + NC and outside it, the treatment of M-DAP + NS + NC recorded the highest average. (259.46 and 240.83)  $\mu$ g p-nitro phenol g<sup>-1</sup> soil 1h<sup>-1</sup>respectively by compared to the control treatment DAP Cont, Cont NS, and Cont NC gave the lowest mean (154.14 and 142.31)  $\mu$ g p-nitro phenol g<sup>-1</sup> soil 1h<sup>-1</sup> respectively.

Table (5) The effect of DAP fertilizer and integrated silicon and nanostructures on the activity of g alkaline phosphatase enzyme  $\mu$ g p-nitro phenol g<sup>-1</sup> soil 1h<sup>-1</sup> in the outside the rhizosphere soil area for Rice plant after 65 days of cultivating

DAP adding levels	DAP Cont	M-DAP	O-DAP	O-DAP High K		
	188.53	219.82	215.94	224.02		
L.S.D	8.58					
NS adding levels	Con	itNS		NS		
	211	.53	2	12.62		
L.S.D		Ν	. <b>C</b>			
NC adding levels	Cont	t NC		NC		
	191.54 232.79			32.79		
L.S.D		6.	07			
	Bi-intera	ction between DAP	and NS			
	Cont DAP	M-DAP	O-DAP	O-DAP High K		
NS Cont	192.20	217.34	210.55	226.11		
NS	184.89	222.42	221.33	221.96		
L.S.D		Ν	.C			
	Bi-interaction between DAP and NC					
	Cont DAP	M-DAP	O-DAP	O-DAP High K		
NC Cont	148.84	206.94	206.23	203.96		
NC	228.12	232.72	225.66	244.26		

L	.S.D	12.14				
	·	Bi-intera	ction between NS a	nd NC		
		NS C	Cont		NS	
NC	Cont	199	.92	1	83.18	
	NC	223	.23	2	42.16	
L	.S.D	8.58				
	·	Triple interact	ion between DAP,	NS, and NC		
		Cont DAP	M-DAP	O-DAP	O-DAP High K	
Cont	Cont NC	154.14	202.73	219.42	223.37	
NS	NC	230.32	231.93	201.71	228.92	
NS	Cont NC	143.61	211.25	193.17	184.48	
110	NC	226.01	233.63	249.54	259.46	
L	.S.D		17	.17		

Table (5) The effect of DAP fertilizer and integrated silicon and nanostructures on the activity of g alkaline phosphatase enzyme  $\mu$ g p-nitro phenol g<sup>-1</sup> soil 1h<sup>-1</sup>in the soil of an area outside the rhizosphere for rice plants after 65 days of cultivating

DAP adding levels	DAP Cont	M-DAP	O-DAP	O-DAP High K	
	190.54	207.82	190.73	199.42	
L.S.D	11.83				
NS adding levels	Con	ntNS		NS	
	196	5.32	19	97.92	
L.S.D		Ν	.C		
NC adding levels	Con	t NC		NC	
	188	3.52	20	05.73	
L.S.D	8.36				
	<b>Bi-interaction between DAP and NS</b>				
	Cont DAP	M-DAP	O-DAP	O-DAP High K	
NS Cont	180.62	194.72	197.55	212.29	
NS	200.31	220.92	183.94	186.91	
L.S.D		16	.72		
	Bi-intera	ction between DAP	and NC		
	Cont DAP	M-DAP	O-DAP	O-DAP High K	
NC Cont	158.91	217.72	177.67	199.90	
NC	222.11	197.83	203.99	198.91	
L.S.D		16	.72		
	Bi-intera	action between NS a	and NC		
	NS	Cont		NS	

NC	Cont	188.62		20	04.00
]	NC	188	.53	20	07.41
L	.S.D		Ν	.C	
	·	Triple interact	tion between DAP,	NS, and NC	
		Cont DAPM-DAPO-DAPO-DAP High k			O-DAP High K
Cont	Cont NC	142.31	194.72	186.13	231.32
NS	NC	219.05	194.82	209.04	193.11
NC	Cont NC	175.52	240.83	169.22	168.64
NS	NC	225.12 200.93		198.79	204.74
L	.S.D	23.65			·

The results of the statistical analysis indicate in Tables (7 and 8) that adding (Control, M-DAP, O-DAP, O-DAP High K) fertilizer led to significant differences after (95) days of cultivation in the average of activity of the alkaline phosphatase enzyme in the rhizosphere and outside it, Where the O-DAP treatment gave the highest mean of (289.42 and 257.92) µg p-nitro phenol g<sup>-1</sup> soil1h<sup>-1</sup> compared to the Cont DAP treatment, which gave the lowest mean activity values of (233.72 and 208.82) µg pnitro phenol g<sup>-1</sup> soil 1h<sup>-1</sup>, The increase in efficacy when treating O-DAP is due to the fertilizer containing humic acids, including humic acid, which plays a role in improving the biological properties of soil, including a significant increase in biomass and an increase in enzymatic activity (Muter et al., 2015). The results in Table (8) showed that there is a significant difference in the treatment of adding NS fertilizer in the average activity of the alkaline phosphatase enzyme in an area outside the rhizosphere. The NS treatment gave the highest average of (241.43)  $\mu$ g p-nitro phenol g<sup>-1</sup> soil1h<sup>-1</sup> <sup>1</sup>compared to the treatment. Cont NS, which gave the lowest mean activity values (231.32) µg p-nitro phenol g<sup>-1</sup> soil 1h<sup>-1</sup>, While we did not record a significant difference in the same treatment in the rhizosphere, Table (7), the increase in efficacy in the NS treatment may have resulted in the addition of silica to the soil, which increased the phosphorous availability ,As silicates replace the exchanged phosphates, which leads to the release of phosphorous from the adsorption surfaces and to increase the availability of phosphorous in the soil (Benzon et al., 2015), The slow degradation of nanofertilizers increases the diversity and number of organisms, thus increasing the enzymatic activity and reducing the loss of nutrients from the soil (Rajput et al., Teng; 2018 et al., 2018). The results in Table (8) showed that there was a significant difference in the treatment of integrated nano fertilizer NC in the average activity of the alkaline phosphatase enzyme in an area outside the rhizosphere, where the NC treatment gave the highest average of (241.00)  $\mu$ g p-nitro phenol g<sup>-1</sup> soil 1h<sup>-1</sup> compared to the control treatment Cont NC, which gave a mean of (231.89) µg p-nitro phenol g<sup>-1</sup> soil 1h<sup>-1</sup>, While the same treatment did not record a significant difference in the rhizosphere, Table (7) that the increase in the enzymatic activity by adding integrated nano fertilizer may be due to the nature of the nano fertilizers with unique physical and chemical properties, including the small size of its minutes and its high surface permeability that allows the adsorption of quantities of the enzyme and thus effectiveness increases (Giraldo et al., 2014), The macro elements available within the integrated nanofertilizer

formulation constitute the active reward in the prophet of the Cofactors or Catalyst of the enzymes, and at the same time, these elements stimulate the enzymes (Rizwan et al., 2019). The results in the two tables show that there are significant differences in the treatment of the bi-interaction between DAP fertilizer and NS in the average activity of the alkaline phosphatase enzyme in the rhizosphere and outside it where the O-DAO and NS treatment gave the highest average of (313 .65 and 269.93) compared to the control treatment DAP Cont and Cont NS which gave a mean (221.00 and 192.62), µg p-nitro phenol  $g^{-1}$  soil  $1h^{-1}$ . The results of the two tables show that there are significant differences in the treatment of the bi-interaction between DAP fertilizer and NC in the values of the activity of the alkaline phosphatase enzyme  $\mu$ g p-nitro phenol. (306.92 and 269.93)  $\mu$ g p-nitro phenol g<sup>-1</sup> soil 1h<sup>-1</sup> respectively as compared to the DAP Cont and Cont NC treatment which gave a mean (227.13 and 199.04)  $\mu$ g p-nitro phenol g<sup>-1</sup> soil 1h<sup>-1</sup>Respectively. The results in tables (7 and 8) showed that there were no significant differences in the treatment of the bi-interaction between the NS and NC fertilizers in the values of the activity of the alkaline phosphatase enzyme in the rhizosphere and outside it. The results of the two tables show that there are significant differences in the triple interaction between DAP, NS and NC fertilizers in the values of the activity of the alkaline phosphatase enzyme in the rhizosphere and outside it . Where O-DAP + Cont NS + Cont NC recorded the highest mean (355.94 and 295.50) µg p-nitro phenol g<sup>-1</sup> soil 1h<sup>-1</sup> compared to the control treatment DAP Cont, Cont NS and Cont NC, which gave a mean (202.84 and 170.81).  $\mu$ g p-nitro phenol g<sup>-1</sup> soil 1h<sup>-1</sup>. The reason for the significantly excelled in the aforementioned triple interaction treatment is that the nanoparticles combined with the DAP fertilizers provided the plant with available nutrients in the form of nanoparticles, which. This increased the biomass that the root secretions provided. As well as the reaction materials represented by high potassium organic DAP fertilizer, as this combination provided a balanced growth and activity of micro-organisms in and outside the rhizosphere, which increased the enzyme activity in the two regions (Bais et al., 2003).

area for Rice plant at	the set ango of curr				
DAP adding levels	DAP Cont	M-DAP	O-DAP	O-DAP High K	
	233.72	281.63	289.42	254.48	
L.S.D	12.81				
NS adding levels	Con	tNS		NS	
	263.65		265.99		
L.S.D	N.C				
NC adding levels	Cont	: NC		NC	
	263	.57	2	66.00	
L.S.D		N	. <b>C</b>		
	Bi-interac	ction between DAP	and NS		
	Cont DAP	M-DAP	O-DAP	O-DAP High K	
NS Cont	221.00	264.82	313.65	255.20	

Table (7) The effect of DAP fertilizer and integrated silicon and nanostructures on the activity of g alkaline phosphatase enzyme µg p-nitro phenol g<sup>-1</sup> soil1h<sup>-1</sup> in the outside the rhizosphere soil area for Rice plant after 95 days of cultivating

NS	246.42	298.53	265.11	253.61	
S.D	18.12				
·	Bi-interac	tion between DAP a	and NC		
	Cont DAP	M-DAP	O-DAP	O-DAP High K	
Cont	227.13	270.82	306.92	249.33	
NC	240.32	292.43	271.92	259.69	
S.D	18.12				
•	Bi-intera	ction between NS a	nd NC		
	NS (	Cont	NS		
Cont	261.29		265.82		
NC	266.14		265.93		
S.D		12.81			
·	Triple interact	ion between DAP,	NS, and NC		
	Cont DAP	M-DAP	O-DAP	O-DAP High K	
Cont NC	202.84	257.07	355.94	228.92	
NC	239.16	272.53	271.33	281.51	
Cont NC	251.38	284.65	257.89	269.66	
NC	241.51	312.31	272.48	237.74	
S.D	25.62				
	S.D S.D Cont Cont Cont Cont Cont Cont Cont Cont	S.D       Bi-interact         Cont DAP       Cont DAP         Cont       227.13         NC       240.32         S.D       Bi-intera         S.D       Bi-intera         Cont       261         NS C       266         S.D       Triple interact         Cont       202.84         NC       239.16         Cont NC       251.38         NC       241.51	S.D       18.         Bi-interaction between DAP a         Cont DAP       M-DAP         Cont       227.13       270.82         NC       240.32       292.43         S.D       240.32       292.43         S.D       18.         Bi-interaction between NS and NS Cont       18.         Cont       261.29         NC       266.14         S.D       12.         Triple interaction between DAP, Interaction betw	S.D         18.12           Bi-interaction between DAP and NC           Cont DAP         M-DAP         O-DAP           Cont         227.13         270.82         306.92           NC         240.32         292.43         271.92           S.D         18.12         18.12           Bi-interaction between NS and NC           NS Cont         18.12           Cont         261.29           NC         266.14         22           S.D         12.81         2           NC         266.14         2           S.D         12.81         1           Cont DAP           MC         202.84         257.07           S.D         12.81         1           Triple interaction between DAP, NS, and NC           Cont NC         202.84         257.07           NC         239.16         272.53         271.33           Cont NC         239.16         272.53         271.33           NC         251.38         284.65         257.89           NC         241.51         312.31         272.48	

Table (8) The effect of DAP fertilizer and integrated silicon and nanostructures on the activity of g alkaline phosphatase enzyme  $\mu$ g p-nitro phenol g<sup>-1</sup> soil 1h<sup>-1</sup>in the soil of an area outside the rhizosphere for rice plants after 95 days of cultivating

•					
DAP Cont	M-DAP	O-DAP	O-DAP High K		
208.82	245.77	257.92	233.10		
12.76					
ContNS		NS			
231.32		241.43			
9.02					
Cont NC		NC			
23	1.89	241.00			
9.02					
Bi-intera	action between DAP	and NS			
Cont DAP	M-DAP	O-DAP	O-DAP High K		
192.62	228.63	267.87	236.39		
225.04	262.98	2468.11	229.86		
18.04					
	208.82 Con 23 Cor 23 Bi-intera Cont DAP 192.62	208.82     245.77       12       ContNS       231.32       Cont NC       231.89       9.       Bi-interaction between DAP       Cont DAP       M-DAP       192.62     228.63       225.04     262.98	208.82       245.77       257.92         12.76         ContNS         231.32       2         9.02         Cont NC         231.89       2         9.02         Bi-interaction between DAP and NS         Cont DAP         192.62       228.63       267.87         225.04       262.98       2468.11		

		Bi-interac	tion between DAP	and NC		
		Cont DAP	M-DAP	O-DAP	O-DAP High K	
NC Cont		199.04	228.02	269.93	230.22	
NC		218.66	263.51	246.00	235.91	
L	.S.D	18.04				
		Bi-intera	ction between NS a	nd NC		
		NS Cont		NS		
NC	Cont	224.52		239.04		
NC		238.24		243.83		
L.S.D		12.76				
		Triple interact	ion between DAP,	NS, and NC		
		Cont DAP	M-DAP	O-DAP	O-DAP High K	
Cont	Cont NC	170.81	206.17	295.50	225.54	
NS	NC	214.36	251.20	240.27	247.11	
NS	Cont NC	227.14	249.99	244.33	234.99	
	NC	222.89	275.91	251.95	224.73	
L.S.D		25.52				

# References

- 1. **Banerjee, A ; S . Sanyal and S. Sen . 2012.** .Soil phosphatase activity of agricultural land: A possible index of soil fertility. Agric Sci Res J 2(7): 412-419
- 2. Dick, W. A. and M. A. Tabatabai (1984). Kinetic parameters of phosphatases in soils and organic waste materials. Soil. Sci., 137: 7-15.
- 3. Nannipieri , P ; L. Giagnoni ; Landi and G. Renella .2011. Role of phosphatase enzymes in soil. In: Phosphorus in action, Springer-Verlag, Berlin Heidelberg.
- 4. Nannipieri, P., Giagnoni, L., Landi, L., & Renella, G. (2011). Role of phosphatase enzymes in soil. In *Phosphorus in action* (pp. 215-243). Springer, Berlin, Heidelberg.
- 5. Burns, R.G; J.L. Deforest ;J. Marxsen ;R.L. Sinsabaugh ; M.E. Stromberger 2013. Soil enzymes in a changing environment: Current knowledge and future directions. Soil Biol Biochem 58: 216-234.
- 6. Benitez, E; R. Nogales; M. Campos and F. Ruano. 2006. Biochemical variability of oliveorchard soils under different management systems. Applied Soil Ecology, vol. 32, no. 2, pp. 221–231.
- 7. Hui , D ; A.M. Mayes and G. Wang .2013. Kinitic parameters of phosphatase : Aquantitative synthesis . Soil Biology& Biochemistry 65,105-113
- Guan, S.Y. 1989. Studies on the factors influencing soil enzyme activities: I. Effects of organic manures on soil enzyme activities and N and P transformations. Acta Pedologica Sinica 26:1: 72 –78
- 9. Parham, J.A ; S.P. Deng ; W.R. Raun and G.V. Johnson .2002. Long-term cattle manure

application in soil. I. Effect on soil phosphorus levels, microbial biomass C, and dehydrogenase and phosphatase activities. Biology and Fertility of Soils 35:5: 328–337.

- 10. **Tarafdar**, **J.C**; **A**.**V. rao and K. Bala**. **1988**. Production of phosphatase by fungi isolated from desert soils. Folia microbiologia 33, 453-457.
- 11. Yosefi, K., Galavi, M., Ramrodi, M., & Mousavi, S. R. (2011). Effect of bio-phosphate and chemical phosphorus fertilizer accompanied with micronutrient foliar application on growth, yield and yield components of maize (Single Cross 704). *Australian journal of crop science*, *5*(2), 175-180.
- 12. Hoppe, H. G. (2003). Phosphatase activity in the sea. *Hydrobiologia*, 493.
- Zhang, X., Ren, C., Hu, F., Gao, Y., Wang, Z., Li, H., ... & Yang, C. (2020). Detection of bacterial alkaline phosphatase activity by enzymatic in situ self-assembly of the AIEgen-peptide conjugate. *Analytical chemistry*, 92(7), 5185-5190
- George, T. S., Gregory, P. J., Wood, M., Read, D., & Buresh, R. J. (2002). Phosphatase activity and organic acids in the rhizosphere of potential agroforestry species and maize. *Soil Biology and Biochemistry*, 34(10), 1487-1494.
- 15. Chai, C., Duan, D., Xu, C., Chen, Y., Sun, L., Zhang, H., ... & Shi, J. (2015). Translocation and biotransformation of CuO nanoparticles in rice (Oryza sativa L.) plants. *Environmental Pollution*, 197, 99-107.
- 16. Alori, E. T., Glick, B. R., & Babalola, O. O. (2017). Microbial phosphorus solubilization and its potential for use in sustainable agriculture. *Frontiers in microbiology*, *8*, 971.
- 17. Ashok, V., Kumar, S. and Singh, R.P. 2015. Enhanced growth and yield of rice (*Oryza sativa* L.) and soil enrichment are mediated by enhanced availability of N and P in soil and plant leaves on application of organic matrix entrapped urea and DAP. International Journal of Plant and environment, 1(1), 57-68.
- 18. Eivazi, F and M.A. Tabatabai. 1977. Phosphatases in soils. Soil Biol. Biochem 9: 167-172
- 19. Sinsabaugh , R.L and D.L. Moorhead .1994. Resource allocation to extracellular enzyme production: a model for nitrogen and phosphorus control of litter decomposition. Soil BiolBiochem 26:1305–1311
- 20. Giraldo JP; MP, Landry; SM,Faltermeier; Mc,Nicholas TP; Iverson NM.2014. Plant nanobionics approach to augment photosynthesis and biochemical sensing. Nature materials; 13: 400-408.
- 21. Adhya TK, Kumar N, Reddy G, Podile RA, Bee H, Samantaray B (2015) . Microbial mobilization of soil phosphorus and sustainable P management in agricultural soils. Special section: sustainable phosphorus management. Curr Sci 108(7):1280–1287.
- 22. Banerjee, A ; S . Sanyal and S. Sen . 2012. .Soil phosphatase activity of agricultural land: A possible index of soil fertility. Agric Sci Res J 2(7): 412-419
- 23. Muter O. ; B. Limane ; S. Strikauska and M. Klavins .2015. Effect of humic-rich peat extract on plant growth and microbial activity in contaminated soil. Material Science and Applied Chemistry. doi: 10.1515/msac-2015-0012 2015/32.
- 24. **Prasad R; R. Pandey; I ,Barman .2016.** Engineering tailored nanoparticles with microbes: quo vadis. Wires Nano-med Nanobiotechnol 8:316–330.

- 25. Abdel-Aziz, H.M.M., Hasaneen, M.N.A. and Omer, A.M. 2018 b. Effect of foliar application of nano chitosan NPK fertilizer on the chemical composition of wheat grains. Egyptian Journal of Botany, 58(1), 87-95.
- 26. Benzon, H.R.L., Rubenecia, M.R.U., Ultra Jr, V.U. and Lee, S.C. 2015. Nano-fertilizer affects the growth, development, and chemical properties of rice. International Journal of Agronomy and Agricultural Research, 7(1), 105-117
- 27. Rizwan, M., Ali, S., ur Rehman, M.Z., Malik, S., Adrees, M., Qayyum, M.F. and Ahmad, P. 2019. Effect of foliar applications of silicon and titanium dioxide nanoparticles on growth, oxidative stress, and cadmium accumulation by rice (*Oryza sativa*). Acta Physiologiae Plantarum, 41(3), 1-12.
- 28. Bais, H. P.; R. Vepachedu; S. Gilroy; R.M. Callaway and J. M. Vivanco .2003. Allelopathy and exotic plant invasion: from molecules and genes to species interactions. Science. 301:1377–80.
- 29. Chen, F., Liu, Z., Guan, Z., Liu, Z., Li, X., Deng, Z., ... & Tang, A. (2018). Chloridepassivated Mg-doped ZnO nanoparticles for improving performance of cadmium-free, quantum-dot light-emitting diodes. *ACS Photonics*, 5(9), 3704-3711.
- 30. Rajput, V. D., Minkina, T. M., Behal, A., Sushkova, S. N., Mandzhieva, S., Singh, R., ... & Movsesyan, H. S. (2018). Effects of zinc-oxide nanoparticles on soil, plants, animals and soil organisms: a review. *Environmental Nanotechnology, Monitoring & Management*, 9, 76-84.