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EFFECT OF AGRICULTURAL SULPHUR AND *THIOBACILLUS* BACTERIA ON MICRONUTRIENTS AVAILABILITY IN SALINE SOIL

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Abstract

The experiment was carried out in the laboratory to investigate the effect of agricultural sulfur and inoculation with sulfur oxidized bacteria (Guns: *Thiobacillus*) on micro nutrient availability of salined soil. Soils were collected from Alfaw region, south of Al Basrah Governorate, The soil was treated with four levels of sulfur (0, 500, 1000 and 2000 kgSha^{-1}), four levels of salinity (3, 6, 12 and 24dsm^{-1}) and two inoculated with *Thiobacillus* bacteria (inoculation and un inoculation), The containers wereincubated at 32c for 30 days. The results showed that application of agricultural sulfur had significant effect on reducing the availability of Zn^{+2} and Cu^{+2} and increasing Fe^{+2} and Mn^{+2} in soil after theperiod incubation. Inoculated with sulfur oxidizing bacteria were increasing the availability of Fe^{+2}, Zn^{+2} and Mn^{+2} but had no effect on Cu^{+2}. The best treatment ($In._1$ * S_{2000} * M_{24}) was lead increasing the availability of Fe^{+2} and Mn^{+2}. So that ($In._1$ * S_{500} * M_{12}), ($In._1$ * S_{500} * M_{24}) treatment were lead to increasing the availability of Zn^{+2} and Cu^{+2} respectively.

Key word: Micronutrients -sulphur-salinity-*Thiobacillus* bacteria.

Introduction

Salinity is one of the major problems of arid and semi-arid regions and a limiting factor in crop growth. In saline and sodic soils, the solubility of micronutrients (e.g Fe, Cu, Mn, Mo and Zn) is particularly low and plants grown on such soils were suffered from deficiencies in these elements (Page *et al*., 1990, AL-Taey and Saadoon, 2014).

The low availability of nutrients rather than low nutrient content is one of the major factors for the widespread occurrence of plant nutrient deficiency in calcareous soil among these factors, high soil pH and $CaCO_3$ contents are responsible for low availability of plants nutrients (Kaya *et al*., 2009, AL-Taey *et al*., 2010).

The accumulation of salts in the soil lead to increased osmotic pressure and food imbalance and when using this water requires the creation of ways and means for the purpose of successful use without the adverse impact of land productivity (AL-Taey and AL-Musawi, 2019).

Agricultural land of Iraq is calcareous soil that contains relatively high amounts of $CaCO_3$ and extremely poor organic matter resulting in high pH.

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Nutrient availability of soils can be increased by application of sulfur (Hydarnezhad *et al*., 2012). The acidifiying function of Sulfur originated from its microbial oxidation to sulphuric acid over time (Vidyalakshmi*et al*., 2009). The biochemical oxidation of sulfur produces H_2SO_4 which decreases soil pH and solubilizes $CaCO_3$ in; Calcareous soils to make soil conditions more favorable for plants growth including the availability of plants nutrients (Abdou, 2006).

Kaplan and Omran (1998) and Erdal *et al*., (2004) reported that soil pH decreased by 0.11-0.37 unit with the application of S^0, resulting in an increase in nutrient concentration, plant nutrient uptake, chlorophyll concentration, root nodules and dry matter production.

The research aimed to determine the effect of sulfur and sulfur oxidized bacteria on availability of Fe^{+2}, Zn^{+2}, Mn^{+2} and Cu^{+2} elements in the salined soil.

Material and Methods

The experiments were carried out in the laboratory of college of Agriculture, Al Basrah University. Soil was collected from Albhar in, Alfaw region, south of Basrah Governorate, at a depth of (0-30)cm. Soil was air- dried,

Table 1: Some chemical and physical properties of the studied soil.

Clay	Silt	sand	PH	E.C.	Cations mmol/L				Anions mmol/L		
431.30	514.10	54.60	(1:1)	(dsm^{-1})	Ca^{++}	Mg^{++}	Na^{+}	K^{+}	HCO_3^-	Cl^{-1}	SO_4^{--}
Texture Silty clay			8.04	3.40	4.30	4.10	21.12	0.78	6.20	25	1.73

OM (gm kg^{-1})	$CaCO_3$ (gm kg^{-1})	DTPA Micronutrients available (mg kg^{-1})				Total Micronutrients (mg kg^{-1})			
		Fe^{+2}	Zn^{+2}	Mn^{+2}	Cu^{+2}	Fe	Zn	Mn	Cu
7.53	290	3.968	1.722	14.241	1.722	12977	108.500	421.400	26.736

grinded and sieved by 2mm to estimate the physical and chemical properties of soil table 1 the soil was salined from 3ds m^{-1} to reach the saline levels of 6, 12 and 24 dS m^{-1} by washing soil until equilibrium between the added saline solution and saline of drained water. Experiment was included four levels of sulfur (0, 500, 1000, 2000) Kg S ha^{-1}, two levels of inoculation (inoculation and uninoculation) and four levels of salinity (3, 6, 12 and 24 dS m^{-1}) (M_1, M_2, M_3 and M_4 respectivity. The experiment was carried out by taking 250g of air- dried of salinzed soil. It was placed in a 500g plastic container. The agricultural sulfur levels and 1 g of sterile organic material (cows' residues) were added, mixed with soil and moisted to the limits of field capacity. Half of the treatments were treated with 2 ml of inoculation *Thiobacillus* bacteria and the other half was left without inoculationand all container were incubatedfor 30 days at 32 Cand keeping the moistureat field capacity. After each incubation periodtheavailable Fe, Zn, Mn and cu were determined. All the properties of soil and water were carried out by the routine analysis (Lindsay and Norvell, (1979), Page *et al*., (1982), APAP, (2005), APAP, (1999)).

Results and Discussion

Concentration of Fe

The iron concentration influenced with the application of sulfur along with levels salinity and *Thiobacillus* bacteria. Iron concentration was remarkably higher in tested soil compared to initial status table 2, 3. Available iron concentration increased with increasing of sulfur levels, This may be attributed to oxidizing the sulfur with *thiobacillus* bacteria and formation sulfuric acid which increased iron solubility (Besharati and Hatami, 2007).

Inoculated with bacteria *Thiobacillus* alone or in combined with sulfur were increased the available Fe^{+2} in soil (Vidyalakshmi *et al*., 2009). Studies of Crowley *et al.,* (1991) have shown that the production of chelating compounds by microorganisms increases iron solubility in the rhizosphere and hence increase plant iron uptake.

Table 2: Some chemical of the studied water.

E.C. ds m^{-1}	Cations mg L^{-1}				Anions mg L^{-1}			
	Ca^{++}	Mg^{++}	Na^{+}	K^{+}	HCO_3^-	CO_3^-	Cl^{-1}	SO_4^{--}
147.00	584.31	422.80	1700.11	201.00	85.01	00	885.00	2017.19

PH	Total Micronutrients (ppm)			
	Fe	Zn	Mn	Cu
8.40	3.197	0.276	0.221	0.022

Table 3: Effect of Sulfur Levels ,inoculated with sulfur oxidizing bacteria and Soil Salinity Levels in Iron availability in Soils (mg Fe^{+2} kg^{-1} Soil).

Inoculation	Salinity levels (M)	(S)Sulfur levels 0	500	1000	2000	In. x M
In. $_0$	M_1	5.751	5.282	3.195	10.587	6.204
	M_2	6.055	5.574	4.184	6.314	5.532
	M_3	6.181	6.144	5.019	4.893	5.559
	M_4	5.516	3.737	6.928	7.151	5.833
In. $_1$	M_1	6.041	15.138	6.139	11.274	9.648
	M_2	6.005	5.702	12.932	7.051	7.923
	M_3	5.933	5.492	6.482	13.120	7.757
	M_4	5.576	4.677	6.125	18.054	8.608
R.L.S.D.$_{0.05}$	0.367					0.098

Iron availability was much higher by treatment (Inoculation + 2000 kg S ha^{-1} + 24 dS m^{-1}) 18.054 mg Fe^{+2} kg^{-1} soil compared with treatment (uninoculation + 1000 kg S ha^{-1} + 3dS m^{-1}) which record 3.195mg Fe^{+2} kg^{-1} soil.

Concentration of Zn

Zinc availability was much higher with the application of sulfur at the rate of 500 kg ha^{-1} alone or in combined with inoculated with *Thiobacillus* bacteria at 30 day and thereafter declined with increasing sulfur. This may attributed to the role of some minerals rich in calcium, phosphorus and sulfur in well ventilated soils that help to increase zinc fixation and decrease it availability. Kayser *et al*., (2001) demonstrated that the application of element sulfur increased zinc solubility in the soil. While Skwierawska *et al*., (2012) reported that the application of sulfate and elemental sulfur decreased the zinc content of 0-40 and 40-80 cm.

The best treatment (Inoculation + 1000 kg S ha^{-1} + 6 dS m^{-1}) was lead to increasing availability of zincby 171 % compared with control. This treatment was not different significantly with (Inoculation + 500 kg

S ha^{-1} + 12 dS m^{-1}) 17.052 mg Zn kg^{-1} soil.

Increasing the salinity to 24 ds m^{-1} has lead to decreasing the availability of zinc ions in soil. this may attributed to the increase in the ionic strength of the soil solution by increasing the saline level, which encourages the adsorption of zinc on the surface of colloids, Also the increase in exchangeable sodium in soil increase the soil ability to absorb zinc compared to other positive ions (Awad, 1987).

Concentration of Mn

The results showed that the application of sulfur at 2000 kg ha^{-1}level were increased Mn^{+2} available in soil by 21.43% compared with out sulfur application table 5. Skwieawska *et al.*, (2008a), concluded that the application of sulfur into soilswhich increased the content of available manganese. Inoculation with *Thiobacillus* was increased Mn^{+2} availability by 67.84% compared with uninoculation, increasing the Mn^{+2} availability could be interpretedto be the results of inoculation and sulfur application in calcareous soils enhanced the biological sulfur oxidation consistently and increased Mn^{+2} availability in the field (Cifuentes and Lindemann, 1993).

Increasing of soil salinity from 3 to 24ds M^{-1} where increasing the availability Mn^{+2}inthe soil by 316.23% measured by the control (13.747) mg kg^{-1} soil.

The superior treatment of triple interaction between sulfur, saline and *Thiobacillus* bacteria was (In * 2000 kg S ha^{-1} * 24 dS m^{-1}) 88.606 mg Mn^{+2}kg^{-1} soil which increased the availability of Mg^{+2} by 601.550 % compared with control (12.630) mg Mn^{+2}kg^{-1} soil.

Variations in Mn behavior, against Fe and Zn, might be related to the soild phase dominance in the tested soils where $CaCO_3$ is considered as the major active soild phase compared to the other reactive phase. The sorption tendency of $CaCO_3$ toward Mn is higher than its affinity toward Fe and Zn, While soil organic matter and Fe hydroxides prefer the sorption and initiate complex formation with Fe and Zn rather than with Mn and Zn. Which can also be fixed or initiate complexes with clay minerals (Ford and Sparks, 2000).

Concentration of Cu

The results showed that the increasing sulfur rate application were reduced the available copper in the soil. The maximum amount was 1.911 ma Cu^{+2} kg^{-1} soil at 500 kg S ha^{-1} soillevel table 6. This may attributed to the formation of copper sulfide complexes that were unavailable. (Awad, 1987). Also Some elements including zinc and copper precipitate as sulfides and sulfates to produce forms that are relatively immobile in the soil profile (Kabata -endias and Pendias, 1992). Also Mahmoud *et al.*, (2013) reported that the application of sulfur (100, 200 and 300 kg ha^{-1}) soil extractable copper concentration remained at stable level or little change

Table 4: Effect of Sulfur Levels , inoculated with sulfur oxidizing bacteria and Soil Salinity Levels in Zinc availability in Soils (mg Zn kg^{-1} Soil).

Inoculation	Salinity levels (M)	(S) Sulfur levels 0	500	1000	2000	In.x M
$In._0$	M_1	6.092	5.820	6.600	5.500	6.003
	M_2	12.612	11.922	14.192	14.024	13.188
	M_3	4.380	5.070	6.740	5.810	5.500
	M_4	5.210	5.472	5.380	6.056	5.530
$In._1$	M_1	7.640	7.324	7.118	5.556	6.910
	M_2	14.888	13.564	17.342	13.332	14.782
	M_3	4.236	17.052	3.230	4.748	7.317
	M_4	6.368	4.244	6.438	6.718	5.942
$R.L.S.D._{0.05}$		0.398				0.106

Table 5: Effect of Sulfur Levels , inoculated with sulfur oxidizing bacteria and Soil Salinity Levels in manganese availability in Soils (mg Mn kg^{-1} Soil).

Inoculation	Salinity levels (M)	(S) Sulfur levels 0	500	1000	2000	In.x M
$In._0$	M_1	12.630	14.546	14.728	11.870	13.444
	M_2	9.126	12.086	15.908	12.620	12.436
	M_3	18.118	14.456	11.644	23.946	17.041
	M_4	60.498	39.818	35.522	56.904	48.186
$In._1$	M_1	14.972	14.206	15.216	11.802	14.049
	M_2	11.916	16.066	37.166	25.164	22.578
	M_3	54.116	37.120	45.454	63.436	50.032
	M_4	61.044	47.414	67.948	88.606	66.253
$R.L.S.D._{0.05}$		1.504				0.301

Table 6: Effect of Sulfur Levels , inoculated with sulfur oxidizing bacteria and Soil Salinity Levels in copper availability in Soils (mg Cu kg^{-1} Soil).

Inoculation	Salinity levels (M)	(S) Sulfur levels 0	500	1000	2000	In.x M
$In._0$	M_1	1.695	1.597	1.542	1.417	1.563
	M_2	1.769	1.711	2.069	2.030	1.895
	M_3	1.484	1.678	1.692	1.552	1.602
	M_4	1.673	2.155	2.172	1.802	1.951
$In._1$	M_1	1.656	1.639	1.585	1.537	1.604
	M_2	1.984	1.947	1.793	1.656	1.845
	M_3	2.140	1.774	2.112	2.076	2.026
	M_4	2.430	2.788	1.913	2.218	2.337
$R.L.S.D._{0.05}$		0.178				0.036

with all treatment of sulfur.

The available copper was much higher with level of salinity (24ds M^{-1}) alone or in combined with Inoculated with *Thiobacillus* bacteriawhich record 2.144 and 1.953 mg Cu^{+2} kg^{-1} soil respectively. The copper availability was much higher at treatment (In.* 500 kg S ha^{-1}* 24 dS m^{-1}) 2.788 mg Cu^{+2} kg^{-1}soil compared with control 1.695mg Cu^{+2} kg^{-1} soil. This reason may be attributed to the prevalence of chloride ion in the saline solution compared to sulfates. Add to that the chloride which has an atomic size smaller than sulfates and had one negative charge. There for, copper and iron bonding with chlorine is more than that with sulfates and itcreatesdissolved complexes compared as bound with sulfates which creates un dissolved complexes.

Conclusion

We concluded that

1. The adding of agriculture sulfur lead to increasing Fe^{+2} and Mn^{+2} and reducing the Zn^{+2} and Cu^{+2} availability

The treatment ($In._1$ * S_{2000} * M_{24}) was lead to increasing availability of Fe^{+2} and Mn^{+2}, so that treatments ($In._1$ * S_{500} * M_{12}) and ($In._1$ * S_{500} * M_{24}) increased availability of Zn^{+2} and Cu^{+2}respectivly.

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