

Effect of copper sulfate on some soil fungi isolated from AL- Qadisiya District fields

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Abstract

Four fungi isolated from the rhizosphere of **Vicia faba** in the fields of AL-Qadisiya district, were selected for their variable resistance to toxic copper sulfate at the range of 25 , 50, 100 ppm concentration . These fungi were **Rhizopus stolonifer** (high tolerance) , **Aspergillus niger** , **Trichoderma harzianum** (Moderate tolerance) and **Fusarium solani** (low tolerance). In regard to the effect of copper sulfate , it was found that the growth of **R.stolonifer** no affected as compared with the comparison control in solid media (P.D.A) with the added copper sulfate, but the growth of **A.niger** decreased with 50 ppm concentration copper sulfate and the growth of **T.harzianum** decreased with 25 ppm concentration copper sulfate , but the growth of **F.solani** inhibited with three concentration of copper sulfate . The result have shown that the copper sulfate activated the dry weight of mycellium of **R.stolonifer** ,**A.niger** with three concentration of copper sulfate in the liquid media, but the dry weight of mycellium of **T.harzianum** activated with 25, 100 ppm of copper sulfate and decreased with 50 ppm concentration. In the same time the result have shown that the dry weight of mycellium of **F.solani** was activated with 25 ppm concentration , but inhibited with 50, 100 ppm concentration .

Introduction

Most heavy metals can affect microorganisms at concentration found in the environment (1,2,3). However, the toxicity of a metal on the physiochemical characteristics of the environment were it is deposited (4). Abiotic factors such as PH, temperature, pressure, and ionic strength

affect the ability of metals to complex with various ligands(5,6). Copper presence in soils and surface waters and sediments leads to its accumulation in microorganisms, higher plants and animals (7,8,9,10). Several studies have considered the role of heavy metals as inhibitors of spore germination . (11) indicate under the experimental conditions , number of deaths in the sun flower seedlings and in reducing the frequency with which **Phytophthora drechsleri** could be isolated from the roots of the surviving plants and in the same time copper, at concentrations above 10 M , totally inhibited zoosporangial production in **P. drechsleri** and severely reduced production in **P. cinnamomi** .(12) found that **Penicillium lilacinum** was strongly resistance to zinc, copper, tin and cadmium whereas **P. purpurogenum** show resistance to cadmium and copper only .They observed that ED50 of the heavy metals .(13) found that the growth of **A.niger** , **R .stolinifer**, **T.harzianum** started to decrease in solid media (P.D.A) with the added cadmium chloride as compared with comparison control except **F.solani** which did not show any growth . (14) reported that cupric ions were most toxic, followed by chromate and chromic to sludge microorganisms .(15) found that **Cunninghamella echinulata** being more tolerate to the large doses than **Fusarium oxysporium f. sp. Lycopersici** with chromium ions .

Material and Methods

Four organisms viz . **Rhizopus stolinifer**, **Asprgillus niger**, **Trichoderma harzianum**, **Fusarium solani** were selected from the rizospher of **Vicia faba** . Copper sulfate was added to the agar media (P.D.A) While still hot then aseptically poured in sterile 9- cm diameter Petri dishes. The final copper sulfate concentration would be 25,50,100 ppm. 4 mm diameter disc from the periphery of 7- day old cultures of each of the four fungi were centrally inoculated in the agar media and dailyt measurements of the colony diameter were carried out incubation at 25c in the dark . This was performed on the 3 replicate dishes for each copper sulfate concentration , as well as control Similar experiments were carried out using 50ml aliquots of liquid media (Trace salts solution) containing the same concentration of the copper sulfate in order to obtain the dry weight of the produced mycellial felts at 7 day . Flask were incubated at 25c and after 7day the mycellial felts were washed with distilled water and dried in a continous hot air oven at 120c for 30 min. and then at 80c until constant weight. Three replicate samples were used in each treatment. The liquid medium has the

following constitution $MgSO_4 \cdot 7H_2O, 0.1g$, $ZnSO_4 \cdot 7H_2O, 0.1g$, $MnCl_2 \cdot 4H_2O, 0.1g$, $FeSO_4 \cdot 7H_2O, 0.1g$, all these components dissolved in one liter distilled water .

Results and Discussion

1- Daily Linear Growth Rate:

Fig (1) shows that the growth of **Rhizopus stolonifer** no affected as compared with the comparison control in solid media (P.D.A) with the added copper sulfate but the growth of **Aspergillus niger** inhibited 10.0% with 50 ppm concentration and the growth activated with 100 ppm concentration. The growth of **Trichoderma harzianum** inhibited 10.0% with 25 ppm concentration and activated with 50,100 ppm copper sulfate , but the growth of **Fusarium solani** inhibited 20.3%, 30.9% , 60% with 25,50,100 ppm concentration of copper sulfate. The statistical methods showed highly significant differences between the concentrations of copper sulfate and very high significant differences between these fungi and copper sulfate concentrations and during the period. This differences might be attributed to better utilization of copper sulfate . These results were similar to results reported by (16) when found that inorganic tin in solid medium can be more toxic than inorganic tin in liquid medium .(10,17) suggested that the behaviour of metals in field depending environment parameters(e.g.salinity, PH,organic matter, particle size) which need to be characterized both by experimentation (e.g. in the laboratory) and by field observation , if possible. (18) suggested that the toxicity of a heavy metal pollutant depend, in part, on the anionic composition of the environment into which the pollutant is deposited .In assessing the toxicity of pollutants to the biota , attention must therefore be focused on the specific physiochemical abiotic factors of the recipient environment, which may mediate or potentiate the toxicity of the contamination(19) .

2-Mycelial Dry Weight:

Fig (2) shows that the copper sulfate activated the dry weight of mycellium of **R.stolonifer,A.niger**, with three concentration of copper sulfate in the liquid media ,but the dry weight of mycellium of **T.harzianum** activated with 25, 100 ppm of copper sulfate and decreased

with 50 ppm concentration and the dry weight of mycellium of **F.solani** activated with 25 ppm concentration and inhibited 10.8% with 50, 100 ppm concentration of copper sulfate. This differences might be attributed to better utilization of copper sulfate. The observed tolerance or sensitivity of these fungi does not appear to inherent to the organisms but may also be due to other factors prevailing in the medium; the possibility that interactions occur between CU and the other cations in the mineral salts solution i.e. Ca , Mg , K , and Fe (11) . (20, 21) found that fungi can adapt to heavy metals and in recent years resistance to heavy Metals has been demonstrated under field conditions .

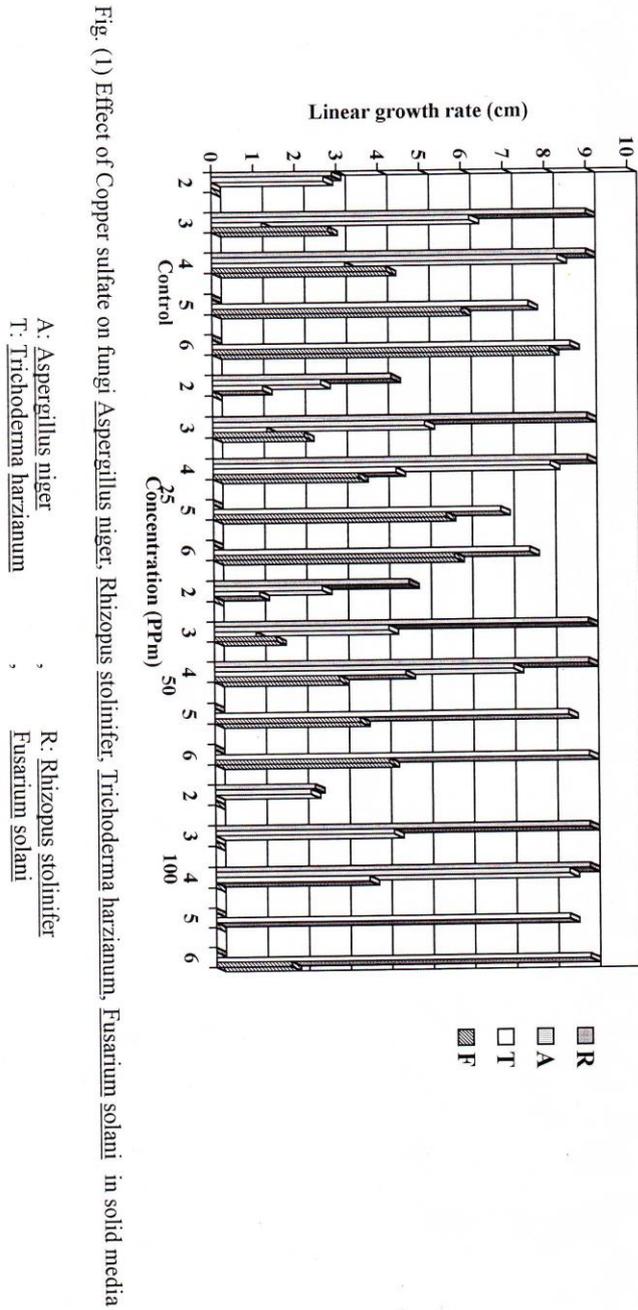
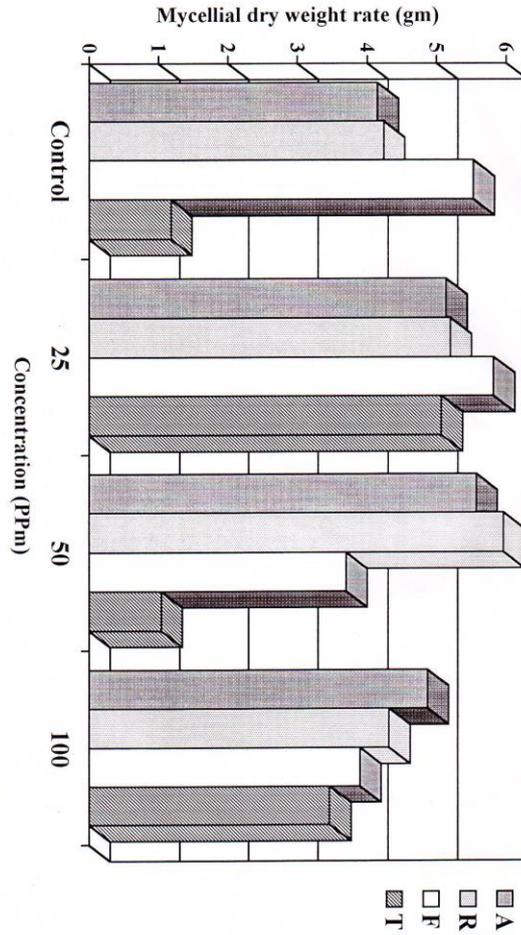


Fig. (1) Effect of Copper sulfate on fungi *Aspergillus niger*, *Rhizopus stolonifer*, *Trichoderma harzianum*, *Fusarium solani* in solid media

Fig. (2) Effect of Copper sulfate on mycellial dry weight to fungi *Aspergillus niger*, *Rhizopus stolonifer*, *Trichoderma harzianum*, *Fusarium solani* in the liquid in 7th day after treatment



A: *Aspergillus niger*
 T: *Trichoderma harzianum* ; R: *Rhizopus stolonifer*
 ; *Fusarium solani*

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تأثير كبريتات النحاس على بعض فطريات التربة المعزولة من محافظة القادسية .

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الخلاصة

اختبرت اربع فطريات معزولة من حول جذور نباتات الباقلاء في حقول محافظة القادسية لتمثل المقاومة المتباينة لسمية كبريتات النحاس بتركيزات 25 و 50 و 100 جزء بالمليون ، وهذه الفطريات هي **Rhizopus stolinifer** (عالي المقاومة) و **Aspergillus niger** و **Trichoderma harzianum** (متوسط المقاومة) و **Fusarium solani** (واطيء المقاومة) . ومن حيث تأثير كبريتات النحاس وجد ان نمو الفطر **R.stolinifer** لم يتأثر مقارنة بمعاملة السيطرة في الوسط الصلب (P.D.A) المضاف اليه كبريتات النحاس ، بينما انخفض نمو الفطر **A.niger** مع تركيز 50 جزء بالمليون ، في حين انخفض نمو الفطر **T.harzianum** مع التركيز 25 جزء بالمليون من كبريتات النحاس ، بينما تثبط نمو الفطر **F.solani** مع التراكيز الثلاثة من كبريتات .

كما اشارت النتائج الى ان كبريتات النحاس حفزت الوزن الجاف للغزل الفطري للفطر **R.stolinifer** و **A.niger** ومع التراكيز الثلاثة من كبريتات النحاس في الوسط السائل . بينما تحفز الوزن الجاف للغزل الفطري للفطر **T.harzianum** مع التركيز 25 و 100 جزء بالمليون من كبريتات النحاس ، وانخفض الوزن الجاف له مع التركيز 50 جزء بالمليون ، وفي نفس الوقت اشارت النتائج الى ان الوزن الجاف للغزل الفطري للفطر **F.solani** تحفز مع التركيز 25 جزء بالمليون ، بينما تثبط مع التركيز 50 و 100 جزء بالمليون .