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Study on Different Temperatures and Concentrations of Certain Salts on Growth of *Sclerotium rolfsii* Isolated from Carrot roots

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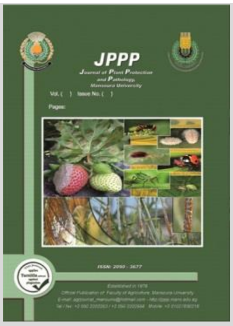
Sarah Elmahalawe[2]; S. Al Afifi[1] and M. EL-Sheshtawi[1*]

[2]Vegetables and Ornamentals Dept. Faculty of Agriculture, Mansoura University, Egypt
[1]Plant Pathology Dept. Faculty of Agriculture, Mansoura University, Egypt.



ABSTRACT

Sclerotium rolfsii is a pathogenic fungi caused rot roots of carrot In this study the best growth of *Sclerotium rolfsii* showed that the mycelial gave maximum sclerotial production growth was very fast at 22 °C , also the pathogen grow and produce sclerotia at temperatures range at minimum and maximum between 10 C° and 22° C . Among of the nutrient slots on the pathogen , there are Potassium (K), Copper (Cu) and Citric acid are proved to be essential elements which required for controlling and inhibiting the pathogen growth,. Using these salts gave promising results to inhibit *S.rolfsii* growth like potassium at all in contrast citric acid didn't prove the same effect, but copper was good at the high concentrations.

Keywords:*Sclerotium rolfsii*, potassium (K), copper (Cu),citric acid, Plant nutrients

INTRODUCTION

S.rolfsii Sacc is a soil-borne, pathogenic fungus which causes root and stem rot, sclerotial wilt and blight on many crops.(Punja, 1988) *S .rolfsii* can make great losses in yield. thus can infect over 400 species of plants including vegetables(Boland and Hall 1997).Sclerotinial rot, caused by *S. rolfsii*, is an economically important disease cause of carrot and can manifest as both a pre-harvest epidemic occurring in the field as well as a postharvest epidemic occurring during storage, with losses ranging from 1-60% in different peanut fields in southern peanut growing region of USA resulting in losses of up to US$ 10-20 million (Aycock, 1966), (Kora *et.al* 2005) (Schwartzkopf, 1972) mentioned that Potassium (K) plays the most important act in the transfer of water and nutrients in the plant. Potassium transports the nitrates, phosphates, calcium(Ca), magnesium (Mg), and amino acids which are low at phloem, the (K) job is to transport specific enzymes and hormones. (K) is necessary to efficient operation of these systems (Thomas and Thomas, 2010) found that Potassium helps to make the difference at pH ranges between 7.3 to 8 to make enzyme reactions. on the other hand it helps to control the pathogen by making difference at pH degree. (Agrios, 2005)

Copper complex developed that fungicides which has the ability to change the germination of spores and conidia of the fungus and reduce spores and germination growth. It has the skill to inhibit the spores and conidia of fungus germination.(Agrios, 2005) and (Narayanasamy, 2007)mentioned that copper helps to reach healthier yield in crop creation and also allows plants to keep themselves out from infections. Citric acid pesticides have the similar activity factor, according to the EPA toxicity tests showed that it had achieved by the formulation records(Blackwell 1998 and2001). citric acid formulations considered as pesticides at Level IV, with sign Word “carefulness”, that’s mean it is the lowest danger comparing with other pesticides at high concentration. And it also considered as a general pesticide in nature.

The aim of study is controlling this pathogen with natural and non-chemical, without using fungicides methods

MATERIALS AND METHODS

1-Effect of Heat treatments on mycelia growth of *S.rolfsii*:

Potato Dextrose Agar (PDA) media used in this study. 20 ml of melted medium was poured into each sterilized Petri dishes. 5 mm disc of *S. rolfsii* seven days old culture was taken by sterilized needle then incubated for7 days at (5,10,15 and 22)± 2 C°. One disc was placed in 1cm distance of the edge of each Petri plate. Three replications of each treatment used. The data on growth were recorded when the fungus reached the rim of the (90mm) Petri dishes, variations in the colony diameter, types of fungal growth of *S.rolfsii* were also recorded after 7 days. To realized successful heat degree treatments were done on mycelium and sclerotia in all isolations that gave success to control *S.rolfsii* in Petri dishes. All factors (media components, time and moisture) are the same except the heat factor, using 5 C° for fruits storage, 10 C° for fruits potential degrees and 15 C° for movement and transportation degrees to markets and 22±2 C° its most markets degrees. The results taken after a week to compare the isolate ability of growth in hard conditions.

2-Effect of Heat treatment on sclerotia:

S.rolfsii sclerotia treated with different degrees of heat at (20,40,70 and 100) C° for a different periods time 10min, 20min and 30min in a water bath that can control heat and time too. With 3 replications for each treatment and

*** Corresponding author.**
E-mail address: melsheshtawi@yahoo.com
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one more as a control. Then sclerotia incubated in PDA media at 20 C° for two weeks.

3-Salts experiment on *S.rolfsii* by Using 2.4,Hexadienoic acid potassium, Copper II sulfate and Citric acid:

Prepared PDA media with different concentrations of salts like (500-1000-5000) ppm of each salt and three replications and untreated control. Salts solved in media before poured 90mm in sterilized Petri dishes. After the media solidified, 5mm discs of the pathogen with 7 days old were put in 1cm of the edge of the dish, only on discs on Petri dish, after that incubated for 2 weeks on 15C°.

RG%=A-B/A

RG=parentage of redial growth
A=the pathogen redial growth on control
B=the pathogen redial growth on treatments

The obtained data were subjected and analysis of variance (ANOVA)(Steel and Terrie 1960) Duncan's multiple range test (DART) were applied for comparing means under study (Duncan, 1955)

RESULTS AND DISCUSSION

1- Effect of heat treatments on the mycelial growth of *S. rolfsii*:

Table 1 show that at 10 ºC and above the radial growth of the pathogen was very high from 8.333 to 8.500 and INH was very low from 2% to 0%, on other side at 5 Cº the redial growth was so tinny 2.033 and INH was76.12% , and that mean that optimal degree on the growth is from 10 ºC to .22 ºC. These results are in agreement with other studies reporting that optimum temperature for mycelia growth and dry weight production was 25°C. (Pane, *et.al*, 2007) and (Zape, *et.al*, 2013) other authors also found that 25°C to 30°C were more effective for the vegetative growth of *S. rolfsii* (Mahen, *et.al*, 1995) (Lin *et.al*, 2010) agree with that growth of *S. rolfsii* was maximum at 25°C but decreased considerably below 20°C and above 35°C.

Table 1. Effect of heat treatments on the mycelial growth of *S. rolfsii*:

Heat degree ºC	RG	INH.%
5Cº	2.033 b	76.12%
10Cº	8.333 a	2%
15 Cº	8.500 a	0%
22 Cº as a (control)	8.500 a	0%

Values within a column followed by the same letter are not significantly different according to Duncun's multiple range test (P≤0.05).
***R.G=Radial growth (cm.) INH =in-habitation**

2- Effect of heat treatments effect on sclerotial growth of *S.rolfsii*:

Table 2 shows that 20 ºC was the optimum degree which the redial growth was the best at it was 8.50 and INH was 0% on 10 min, and on 20 min redial growth was 8.46 and INH was 0.5%, while on 30min redial growth was 8.23 and INH was 3.2%, after that 40 ºC was considered the second suitable one, while 100 ºC had a good effect to make the sclerotia in a low growth level because the redial growth was 4.96 ºC and INH was 41.6% on 10 min and on 20 min redial growth was 41.6% and INH was 41.6% . That means that 100 ºC is a perfect degree to controlling *S. rolfsii*, but it's so harmful for plants cellular tissue so we cannot use it. While 20c and 40 ºC are not enough to inhabitant the growth. These results agree with study indicate that sclerotia mortify quickly at 40:70 °C temperatures **(Vannacci, *et.al* 1988 and Lin, *et.al* 2010).**

Table 2. Effect of Heat treatments effect on sclerotial growth of *S.rolfsii*:

Time	10mis		20mis		30mis	
Heat degree	RG	InH.%	RG	Inh.%	RG	Inh.%
20C	8.50 a	0%	8.46 a	0.5%	8.23 a	3.2%
40C	8.36 a	1.6%	8.03 b	5.5%	7.36 b	13.4%
70C	7.66 b	9.18%	7.13 c	16.11%	6.30 c	25.88%
100C	4.96 c	41.6%	4.70 d41.6%	44.7%41.6%	2.56 d	69.8%
CONTROL			8.5 a		0.00%	

Values within a column followed by the same letter are not significantly different according to Duncun's multiple range test (P ≤0.05).
***R.G=Radial growth (cm.) INH =inhabitation**

3- Effect of Salts concentrations on the pathogen redial growth:

Table 3 Show that potassium is considered the best salt which can inhabits the pathogen growth at all concentrations 0.05,0.1, and 0.5, while copper achieved the best result only at high concentration 0.5, but at low concentration 0.05 results were as similar as the control.

Table3. Effect of Salts concentrations on the growth of *S. rolfsii*:

Concentration	0.05/150ml			0.1/150ml			0.5/150ml		
Treatment	RG	INH.%	NO.S	R.G	INH.%	NO.S	R.G	INH.%	NO.S
Potassium	0.00 b	100%	0.00 c	0.00 b	100%	0.00 a	0.00 b	100%	0.00 b
Citric acid	8.50 a	0%	36.00 a	8.50 a	0%	36.6 a6	8.50 a	0%	34.33 a
Cooper	8.50 a	0%	33.33b	8.50 a	0%	34.66 a	0.00 b	100%	0.00 b
Control	8.5 a	0%	43 a	8.5 a	0%	41 a	8.5 a	0%	48 a

Values within a column followed by the same letter are not significantly different according to Duncun's multiple range test (P≤0.05).
***R.G=Radial growth (cm.) INH =inhabitation No.S=number of sclerotia**

At the other side citric acid has no effect on pathogen growth at all concentrations used, the citric acid results as the same as the control. This result agree with Mandira and Khan (2003) and Pekarskas and Bartaseviciene (2007) who found that potassium levels of 0, 75 and 150 kg/ha affected growth and yield of onion (*Allium cepa* L.)by controlling the soil fungi. And also gave the highest yield on onion was recorded with75 kg/ha of potassium fertilizer. And also agree with (Marschner, 1995) who showed that Cu helps in promoting lignifications in plants to develop plants primary defense mechanism against fungal invasion. At the same time Evans and Baber (2010) reported the effect of Cu in promoting the formation of lignin and this resulted in reduced fungal diseases in different plant species.

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دراسة علي تأثير درجات الحرارة المختلفة وتأثير تركيزات مختلفة من الأملاح علي نمو فطر *Sclerotium rolfsii* المعزول من جذور الجزر

سارة سليم المحلاوي [2]، سمير العفيفي[1] و محمد الشثتاوي[1]

[2]قسم نباتات الخضر و الزينة. كلية الزراعة جامعة المنصورة.

[1]قسم أمراض النبات، كلية الزراعة ، جامعة المنصورة .

تمت هذه الدراسة بهدف الحصول علي طريقة فعالة لتثبيط نمو فطر*Sclerotium rolfsii* في المعمل تحت ظروف درجات حرارة مختلفة وهي من 5م20 :ºمº,علي النمو الميلسيومي للفطر الممرض وعلي الأجسام الحجرية التي ينتجها الفطر وكذلك استخدام بعض الأملاح المعدنية كأحد العوامل التي تثبط وتساعد علي وقف نشاط النمو مثل البوتاسيوم (K) والنحاس (Cu) وحمض الستريك (ملح الليمون) بتركيزات 0.05جم و0.1 جم و 0.5 جم/100ملل لتر ماء واختبار تأثيرها علي نمو الفطر علي النمو الميسليومي والأجسام الحجرية. وقد الذي تبين **إن** أفضل نمو لفطر *Sclerotium rolfsii* كان عند 22 درجة مئوية وأعطى أقصى إنتاج من الأجسام الحجرية ، كما يحدث نمو فطري بصوره اقل في درجات الحرارة بين 10 :20 درجه مئوية . باستخدام الأملاح المختبرة كما سبق على الفطر الممرض ، وقد اتضحت قدرتها علي تثبيط نمو الفطر الممرض وتنشيط فسيولوجيا النبات وبالتالي زيادة معدلات نمو النباتات. أعطى استخدام هذه الأملاح نتائج مبشرة في تثبيط نمو الفطر *S.rolfsi*i مثل البوتاسيوم في جميع التركيزات ، لكن النحاس كان جيدًا عند استخدامه بالتركيزات العالية فهو يأتي في المرتبة التالية للبوتاسيوم بينما التركيزات المنخفضة منه لم تتمكن من تثبيط الفطر الممرض بينما لم يكن لحامض الستريك تأثير في أي تركيز.