J. Plant Prot. and Path., Mansoura Univ., Vol. 4 (1): 41 - 47, 2013

EFFECT OF MAGNETIC FIELD ON VIRULENCE AND MORPHOLOGICAL CHANGES OF *Sclerotium cepivorum*

Abd El-Nabi, Heba M.[1] and M. Hafez [2]

[1] Suez Canal Univ., Fac. of Agric., Dept. of Agric. Botany, Ismailia 41522 Egypt.

E-mail: oheba2004@yahoo.com

Email: heba_abdelnabi@agr.suez.edu.eg

[2] Suez Canal University, Faculty of Science, department of Physic, Ismailia 41522 Egypt.

ABSTRACT

The effects of the exposure to Dynamic magnetic field (ACMF) of 7,10 and 15 gauss for periods from 4 to 12 hours on the *Sclerotium cepivorum* were studied in vitro by using microscope (Leica version 1.8, Germany). ACMF made changes in the thickness of the mycelium of the fungus and reduce the production of sclerotia . Data was indicated there is no significant between the three media with average 44.03, 44.38 and 45.00 respectively. Also, there is no significant between the periods of exposure (4, 12 h) with average 41.44, 47.50. the interaction between Media x Hours (M x H) show there are significant difference between them. Finally, I conclude that by increase the period of exposure time of Dynamic magnetic, it will decrease the Disease severity and the Disease incidence.

INTRODUCTION

Sclerotium cepivorum Berk, is the causal agent of white rot of onions and other *Allium* spp. The fungus produces vegetative resting structures known as sclerotia, which enable it to survive in the soil for many years without host. These sclerotia are small (0.25-0.6 mm dia.) black, a brown and spherical (Punja and Rahe 1992).

Electromagnetic field (EMF) is a generic term for fields of force generated by electrical changes or magnetic fields under certain circumstances. EMF can be considered as radiation when they radiate energy from the source of the fields. Electromagnetic waves periodically change between positive and negative. The speed of changes, or the number of change per second, is called the frequency and is expressed in hertz (1Hz=1 fully cycle of change per second).

One of the methods to study the pathogens of plant diseases is EMF. It influence on the various species of microorganism was investigated by different researchers. Some literature reports the effect of a magnetic field on the growth and the germination of plants (Smith and Mays 1984, Ruzic et al. 1993) and rare on fungi (Sadauskas et al. 1987, Broers at al. 1992, Ruzic et al. 1997, Maria et al. 2003) were studied . Ruzic et al. 1997 studied the effect of 50 Hz frequency magnetic field of 0.025 and 0.1mT magnetic flux density on mycelia growth and ergosterol content of mycorrhizal fungi. The cultures were grown in a magnetic field for 28 days and the dry and wet weight as well as the ergosterol content of cultures was measured on the 7^{th} day. The stimulation effect of magnetic field of 0.1mT flux density could be seen more

quickly between the 7 th and 14 th days from treatment, then the effect of magnetic field of 0.025 m T flux density. The ergosterol content slightly increased in the first week of treatment.

Maria Cristina et al. 2003 studied the effect of 0.3± 0.03T static magnetic field on the *Fusarium culmorum* in vitro, SMF inhibition of mycelia growth was accompanied by morphological and biochemical changes and fungal conidia germination and cell viability were also reduced.

Sadauskas et al. 1987 examined the effect of 200mT flux density static and 29 mT flux density pulsating magnetic field on the different species of fungi, according to their examination, morphological changes were observable on the conidia of *Aspergillus puniceus* and *Alternaria alternate.* The pigmentation of the colony of *Aspergillus niger* changed, the cultures remained white.

The aim of the present study was examine the effect of AC magnetic field with flux densities which are not much greater than the flux density of average geomagnetic field on the virulence of the fungus *Sclerotium cepivorum* and morphological changes of mycelium growth.

MATERIALSAND METHODS

Isolation of the fungus

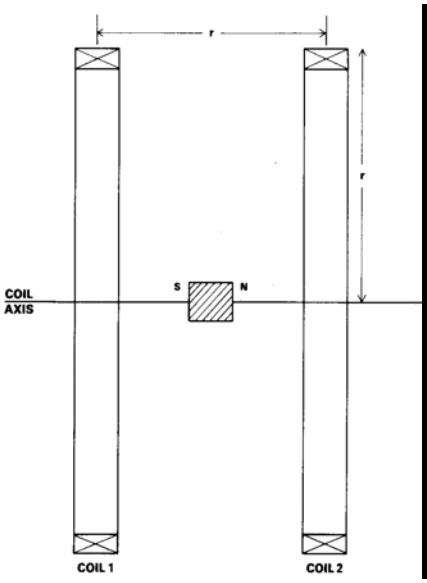
Sclerotium cepivorum was isolated from infested onion plants with the fungus which it cause white rot, then purification on Potato dextrose agar (PDA) plates and *Sclerotium cepivorum* has been preserved on PDA media at 20º c ±2.

Preparation of culture fungi

Discs (5mm diameter) from *S. cepivorum* culture isolated from onion were inoculated in the Petri –dishes containing Potato dextrose Agar (PDA) and grown for 2 weeks in a incubator at 18-20˚ c. Petri-dishes were used for ACMF exposure experiments.

Exposure the fungus *Sclerotium cepivorum* to the dynemic magnetic field (ACMF) or the investigations of the effect of a magnetic fields on the mycelia growth and sclerotia formation of the fungus

In our experiment, we use Helmholtz coils to produce our magnetic fields. The strength of recognized and unvarying generation fields is very significant for perfect and quotable usceptible measurements. In the Helmholtz coils configuration, coils of radius 50 cm are separated by a distance 50 cm, which produces a nearly uniform magnetic field between the two coils. The magnetic field strength was measured using German tesla-meter (Model KOSHAVA 5).We fixed the magnetic field at 7, 10 and 15 gauss for periods from 4 to 12 hours as we will explain later.



Cross-sectional view of Helmholtz coils

The samples inside Helmholtz coil

Morphological changes after exposure the fungus to ACMF

One sclerotia from *S. cepivorum* inoculated in Petri-dishes containing PDA media and incubated at 17-20 ˚c for 2 weeks then exposure the culture to DMF for 4,12 h. A part of mycelium growth put on the slide in a drop of glycerol and covered by cover slide to examine under Microscope (Leica version 1.8, Germany) connecting with monitor to capture the photo for the growth of the fungus. To investigate if there any morphological changes of the growth by change the media growth or not, I prepared different media (czapek -Dox Agar,PDA,PFA) and repeated it the previous methods but cultivated the sclerotia in Petri-dishes containing different media.

Effect of magnetic field on the virulence of the fungus *S. cepivorum:*

The experiment layout

Under greenhouse condition, plastic pots filled with sandy loamy soil (1:1) and inoculated with sclerotia of the fungus after exposure to ACMF for (0.4.12 h) by ratio 2% and replicated five times for each treatment , also, five pots filled with sandy loamy soil inoculated with sclerotia of the same fungus without exposure to ACMF and used them as a control. After one week seedlings of onion cultivated in all the previous pots and leaved them for two weeks to determine the disease severity and disease incidence.

Statistical analysis

Analysis of Variance

Data were subjected to a factorial analysis of variance with three media and two exposure time. The experiment was laid out in a completely randomized design (CRD) with five replications the data were statistically analyzed according to (Steel et al*.,* 1997). A Computer program software

Costat was used to analysis the data of this experiment. The means were compared by Least Significant Differences (LSD) at the 0.05 probability level.

RESULTS

First: concerning to the exposure time of Dynamic magnetic on the disease severity using three different media

Data from previous table (1) indicated there is no significant between the three media with average 44.03, 44.38 and 45.00 respectively. Also, there is no significant between the periods of exposure (4, 12 h) with average 41.44, 47.50. the interaction between Media x Hours (M x H) show there are significant difference between them, medium three at time 12 was the best treatment to decreased of D. S. with value 27.22 following by medium 2 at 4 h. with value 30.00. On the other hand, medium 3 and medium 1 at time 4 h. exhibited highly disease severity with average 60.83, 51.66 respectively.

Second: concerning to the exposure time of Dynamic magnetic on the disease incidence using three different media

Data from the previous table (1) showed that the significant between three media, the medium 3 gave the best result in reduction in disease incidence with average 84.4. On the other hand, there is no significant between the periods of exposure time (4, 12 h). The interaction between (M x H) showed that there are significant between them. Medium 3 at time 12 h give high decreased in D. incidence with average 68.88 followed by medium 2 with average 80, reverse with all other media.

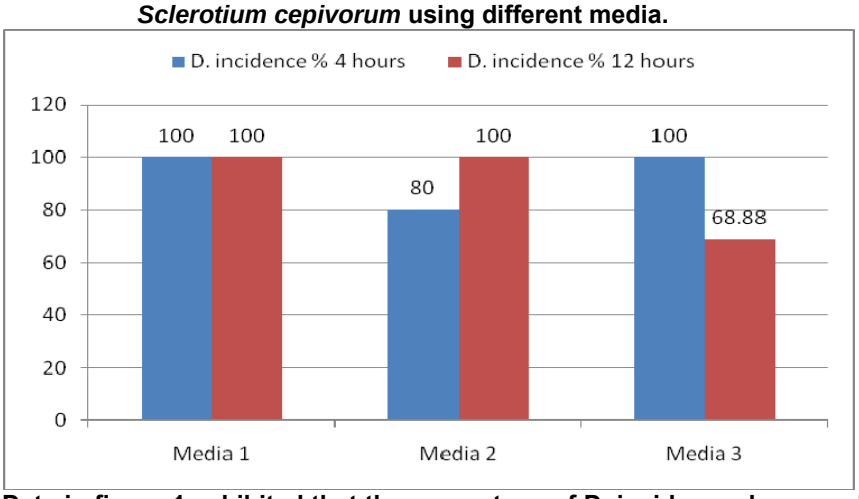
Finally, I conclude that by increase the period of exposure time of Dynamic magnetic, it will decrease the Disease severity and the Disease incidence.

Table (1): Effect of dynamic magnetic field on the disease severity and disease incidence of *Sclerotium cepivorum using* different media.

	D. severity %		Mean	D. incidence %		Mean
	4 hours	12 hours		4 hours	12 hours	
Media 1	51.66 ^{abc}	38.33 ^{abc}	45.0 A	100.0 ^{a}	100.0 ^{a}	100 A
Media 2	30.00 ^{bc}	58.76 ^{ab}	44.38 A	80.0 ^{b}	100.0 ^{a}	90 B
Media 3	60.83 ^{a}	27.22 ^{c}	44.03 A	100.0 ^{a}	68.88 ^{c}	84.4 C
Mean	47.50 A	41.44 A		93.33 A	89.62 A	
L.S.D	M=21 H=17.5 M× H= 30.63			M=5.23 H=4.27 M× H= 7.39		

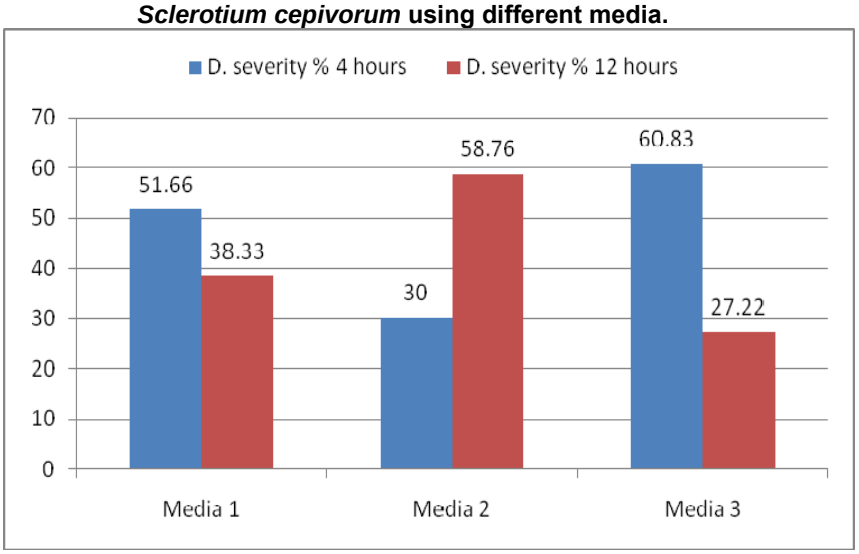
Medium 1 ((czapek), Medium 2 (PDA), Medium 3(PFA)

Figure (1): Effect of Dynamic magnetic field on the disease incidence of *Sclerotium cepivorum* using different media.



Data in figure 1 exhibited that the percentage of D. incidence decreased by increase the exposure time of magnetic. Medium 3 is the best one at time H12.

Figure (2): Effect of Dynamic magnetic field on the disease severity of *Sclerotium cepivorum* using different media.



Data in figure 2 exhibited that the percentage of D. severity decreased by increase the exposure time of static magnetic. Medium 3 and medium 2 were the best of decreased the D. severity. Also, figure shows by increasing time exposure of magnetic decrease the D. severity.

Data in Table (2) show the reduction of growth of the fungus *S. cepivorum* decreased by increasing the exposure time of ACMF with all the different media. Also, data show there are significant different between the kind of media and exposure time, medium with source of carbon sucrose gave the highest reduction of growth with increase time exposure of ACMF then the medium with fructose (4.81,5.62, 6.59) respectively.

Table (2): Effect of exposure time with different media on the growth of *S. cepivorum*

Media	0 time	4 hours	12 hours	**Mean**
Nitrate	5.72 bc	7.80 a	6.26 b	6.59 A
Sucrose	5.517 cd	3.901 e	5.04 d	4.81 C
Fructose	5.52 cd	5.45 cd	5.89 bc	5.62 B
Mean	5.59 A	5.72 A	5.73 A	
L.S.D> 0.5	**M=0.40** **H=0.37** **M× H= 0.63**			

DISCUSSION

Sclerotium cepivorum Berk, is the causal agent of white rot of onions and other *Allium* spp. The fungus produces vegetative resting structures known as sclerotia, which enable it to survive in the soil for many years without host. These sclerotia are small (0.25-0.6 mm dia.) black, a brown and spherical (Punja and Rahe 1992). So, I proposed c dynamic magnetic fields (DMF) as a safe preservative agent substitute for chemical fungicides to help us to control or at least to decreased the populations of sclerotia of the fungus from the soil, especially, The experimental was exhibited the exposure to Dynamic magnetic field decrease percentage of the disease incidence and disease severity of *S. cepivorum* and made of the morphological changes of mycelium of the fungus. These results were agreement with the results was investigated by Nagy Pal , 2005 when studied the effect of ACMF on some plant pathogen fungi and found the magnetic field decreased the growth of colonies by 10% using 0.1, 0.5, 1 m T density region. Maria Cristinia Albertini et al., 2003 found that ACMF inhibition of mycelia growth was accompanied by morphological and biochemical changes of *Fusarium culmorum* . Al. Manoliu, at al. 2006 studied the effect of ACMF on the catalase and peroxidase in some cultures of cellulolytic fungi and was found the magnetic exposure had remarkable effect on the enzyme activity in the mycelium fungi.

REFERENCES

Al. Manoliu, L. Oprica, Z. Olteanu, I. Neacsu, V. Artenie, D. E. Creanga, I. Rusu, I. Bodale 2006. Peroxidase activity in magnetically exposed cellulolytic fungi. J. of Magnetism and Magnetic Materials 300,323-326.

Broers D., Kraeplin G., Lampreeht I., Schultz O. 1992. Mycotypha Africana in low– level athermic ELF magnetic fields. Bioelectro chemistry and Bioenergetics 16:281-291.

Maria Cristina Albertini , Auguso Accorsi, Barbara Citterio, Sabrina Burattini, Maria Piera Piacentini, Francesco Uguccioni, Elena Piatti. 2003. Morphological and biochemical modifications induced by a static magnetic field on *Fusarium culmorum.* Biochimic 85, 963-970.

Nagy Pal 2005. The effect of low inductivity static magnetic field on some plant pathogen fungi. J. of Central European Agriculture Vol 6 , 2 , 167-171.

Punja, Z. K., Rahe, J. E. 1992. Sclerotium In: Singleton, L. L., Michail, J. D., Rush, C. M. (Eds.). Methods for research on soil – borne phytopathgenic fungi. APS press. St. Paul. Pp.166-170.

Ruzic R., Gogala N., Jerman I. 1997. Sinusoidal magnetic fields: Effects on growth and ergosterol content in mycorrhizal fungi. Electro – and Magneto biology. 16: 129-142.

Ruzic R., Jerman I., Jeglic A., Fefer D. 1993. Various effects of pulsed and static magnetic fields on the development of *Castanea sativa* mill. In tissue culture. Electro-and Magneto biology. 12:165-177.

Sadauskas K. K., lugauskas A. Y., Mikulskene A. I. 1987. Vlijanie postojannogo impulsnogo nizkochastotnogo magnitnogo poljana mikroskopicheskie gribi (Effects of constant and pulsating low-frequency magnetic field on microscopic fungi) Mikologija Fitopatologija. 21:160-163.

Smith R. D., Mays R. 1984. Effect of pulsed magnetic fields on root development in plant cuttings. Bioelectrochemistry and Bioenergetics. 12: 567-573.

تـأثير المجـال المغنـاطيس علـي مرضـية ومورفولـوجي فطـر *Sclerotium cepivorum*

هبه محمد عبد النبي[١] و محمد حافظ[٢]

[١] كلية الزراعه – قسم النبات - الإسماعيلية

[٢] كلية العلوم– قسم الفيزياء - الإسماعيلية

بتعـريض ميسـليوم الفطـر *Sclerotium cepivorum* إلـي المجـال المغنـاطيس (ACMF)٧,١٠,١٥ جاوس لمدة ٤,١٢ ساعة متواصلة بالمعمل و أدى ذلك إلي وجود تغيرات في سمك الميسليوم واختزال في أنتاج الأجسام الحجرية للفطر. وأيضا وجد انه عند استخدام ثلاث بيئات مختلفة في مصادر الكربون لم يكن لها اى تأثير علي سلوك الفطر وكذلك مدة التعريض المستخدمة . وفي النهاية وجد انه بزيادة مدة التعريض للمجال المغناطيس يؤدى إلي تقليل في شدة إصابة والمرضية للفطر.

قام بتحكيم البحث

أ.د / محمد عبد الرحمن الوكيل	**كلية الزراعة – جامعة المنصورة**
أ.د / ابراهيم ناجى محمد على	**كلية الزراعة – جامعة قناة السويس**

42

43

44

45

46

47