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ISOLATION AND IDENTIFICATION OF *FUSARIUM* SPP. ASSOCIATE WITH POTATO TUBER DRY ROT DISEASE: GROWTH RATE AND PATHOGENICITY IN RELATION TO ELICITATION OF PHYTOALEXINS IN INOCULATED TISSUES.

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ABSTRACT

Potato tubers collected from different stores in Cairo Governorate and stored at laboratory conditions showed the occurrence of dry rot symptoms on high proportion of them accompanied by appearance of whit mycelia mats characterized *Fusarium* spp. Such fungi were isolated, purified and identified according to their morphological characters and identified as *F. solani* (5 isolates), *F. semitectum* (4 isolates), *F. equisiti* (5 isolates), *F. oxysporum* (5 isolates) and one unknown species. Pathogenecity test of such isolates on potato tuber slices Balmivore cv. indicated the presence of different degrees of virulence between isolates either belong to the same species or not. In general *F. solani* isolates showed the higher degree of verulance. Correlation coefficient between fungal growth and disease severity reached – 0.0624 indicating the absence of any relation between both phenomena. The ability of such isolates as phytoalexin elicitors was studied. Some isolates especially related to *F. solani* appeared more active than the others which belong to another *Fusarium* spp. Correlation coefficient between disease index and number of phytoalexin bands reached -0.15. However, it was noticed that *Fusarium solani* isolates which showed high disease index elicit the higher account bands of phytoalexins which mean that these compounds have relation with disease incidence.

KEYWORDS: Balmivore cv., *Fusarium* spp., Phytoalexins, *Fusarium solani, Fusarium sambucinum, Fusarium oxysporum, Fusarium equiseta.*

INTRODUCTION

Potato tubers either under storing condition or during cultivation is subject to attack by different fungal species which belong to Genus *Fusarium* **(Gachango *et al.*, 2012; Falert and Akarapisan, 2019).** Such attack is usually occurred through wounds due to mechanical injures during harvesting which cannot preventing or may through wounds occurred by wireworms and white grubs, these insects can severely damage seed pieces and tubers and through their wound the fungus penetrates into tuber tissues **(Daami-Remadi, 2012).** Potato dry rot is a very destructive disease; it may cause complete loss of tubers leading to serious loss of agricultural economics. In a previous study **(El-Hassan et al., 2007)** have isolated and identified *Fusarium* spp. associate with dry rotted tubers as: *F. sambucinum, F. solani, F. oxysporum, F. culmorum, F. equesiti and F. semitectum.* The present study was planned to investigate and identify species of *Fusarium* associate with the disease which may be changed due to changing of cultivars, study any phenotypic relationship between tested species and their isolates and disease symptoms due to artificial inoculation and their linear growth with special reference to their correlation with disease severity. Moreover, phytoalexins accumulate in inoculated tissues will be extracted and TLC separated in order to study their relation-ship with disease insex.

MATERIALS AND METHODS

Samples

Samples of potato tubers were collected from different markets in Cairo Provence, Egypt during summer seasons of 2017-2018. Tubers were stored in cartoon boxes at room temperature for one month. After that, tubers were inspected for dry rot incidence and diseased ones were photographed. The percentage of diseased tubers after they had been stored reached 30%.

Isolation of *Fusarium* spp. accompanied with rots

Pies of potato tuber tissues contained mycelia mats of *Fusarium* was cut from diseased tissue then suspended in conical flask (50 ml) contained sterilized distilled water (SDW). Shacked for 5 min on rotary vibrator, then one ml contained conidial spores of *Fusarium* was taken in test tube contained 9 ml of SDW to obtain 1/10 dilution. Serial dilution up to 10^{-6} was carried out. One ml. of the

last dilution was spread on the surface of water ager in sterilized Petri dish. Dishes were incubated at 24±1°C for 24 h in the dark. After that, the surface of water agar contained fungal spores was examined under binocular in order to get single spore colony. Every single spore colony was transferred to sterilized Petri dish contained potato sucrose agar medium, and then dishes were incubated at 24±1°C under light fluorescent for 7 days.

Identification of isolated *Fusarium* spp

According to **Booth (1971)** the isolated fungi were identified firstly at genus level according to morphological features in Petri dishes and colony diameter which was measured after 69 h of cultivation. Slide culture technique was used to examine the main features of such Genus. This technique was carried out as described by **(Riddell, 1950).** Phialide type, micro- and macro-conidial size, formation of chlamidospores and all features of *Fusarium* as described by **Booth (1971)** were examined and photographed.

Pathogenecity test of isolated Fusaria

Potato tuber slices of Balmivor cv. (1 cm thick) were prepared with exclusion of apical and basal parts of tubers. Slices were washed several times in SDW then left to dry under aseptic condition, and then transferred to large Petri dishes (15 cm in diameter). Every dish contained 4 slices. From the active edge of fungal growth piece half cm. of fungal growth contained growth medium was cut and transferred to the central part of slice surface then dishes were incubated at 24±1°C in the dark. Diameter of fungal growth on surface slice was measured after 3 and 7 days from incubation. Moreover, after 9 days slide was cut perpendicular to its axis, then the inner tissue colonized by the fungus was examined and photographed.

Disease index

According to our observations on types of infection, infection types were found to lie in 5 categories according to the following figure.

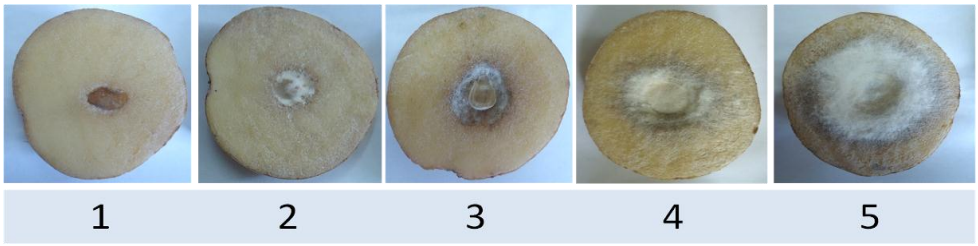


Fig. (1): Different types of disease index on potato tuber slices inoculated by a piece of fungal growth. Where 1: no visible symptoms; 2: growth diameter reached 5 mm; 3: growth diameter reached 10 mm; 4: growth diameter reached 15 mm; 5: growth diameter reached 20 mm.

Extraction and separation of Phytoalexins from potato tuber tissues inoculated by different Fusaria

Potato tuber slices were prepared as previously mentioned, arranged in Petri dishes, then inoculated by different *Fusarium* spp. previously isolated from potato tuber tissues showed dry rot symptoms. Slices were inoculated individually by fungal isolates using spore suspension (5 X 10^5 spore/ml). Inoculated slices were incubated at 24 ± 1° for 48 h. Equal areas of the upper surface of inoculated slices 2 mm in thick and 2 cm in diameter) were gently separated and every treatment was collected together in a glass bottle closed court and sufficient volume of benzene was added to slices in order to extraction of phytoalexins. After 48 h benzene phase was taken and evaporated till dryness under reduced pressure, then residues were dissolved in 100 µl of chloroform. Silica gel-G plates were used to separate potato phytoalexins. Chloroform: acetone (7:3v/v) was used to separation of phytoalexins as immobile phase. After solvent had been reached the front of silica gel plates, they were taken and left to dry then dipped in phosphomolybdic acid in methanol (5%) and heated at 105°C for 5 min. R_f of visualized bands was calculated according the following formula:

R_f of separated band= Ds/Df

Where: Df = the distance of the solvent traveled from the point of start. Ds= the distance of the test spot traveled from the point of start **(Maha H. Mohamed and Mostafa H. Mostafa, 2019).**

Statistical analysis

Standard deviation between averages of 3 distinct experiments was calculating according to **(Lee *et al.*, 2015)** and correlation coefficient between rate of growth and disease severity and the correlation between number of separated phytoalexin bands and disease index were calculated according to **(Schober *et al.*, 2018).**

RESULTS

Disease symptoms of potato dry rot

Storing of potato tubers collected from different markets of Cairo governorates at room temperature for one month showed the appearance of fungal mycelia mats on about 30% of stored tubers, this fungal growth was associated with dry rot as indicate in the following picture.

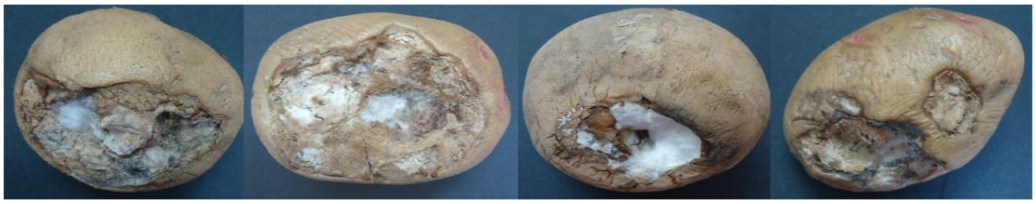


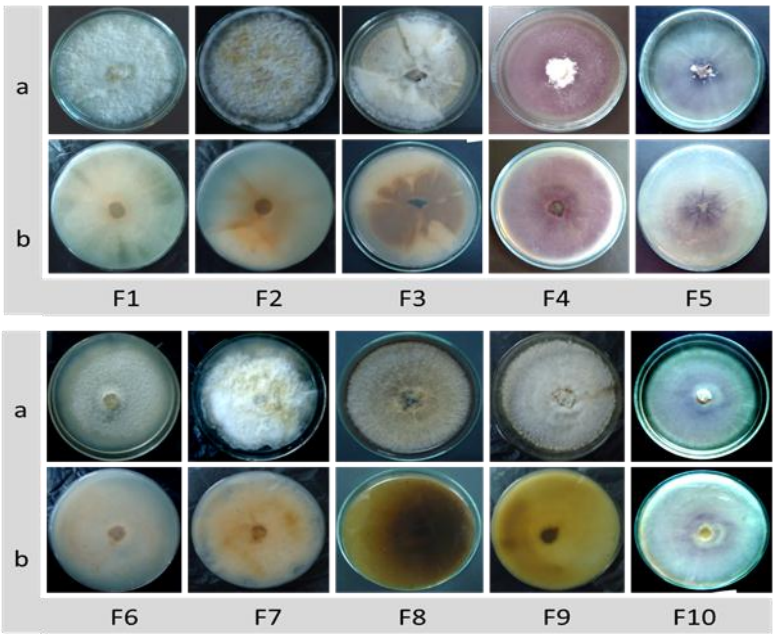
Fig. (2): symptoms of dry rot of potato tubers stored at room temperature for one month.

Isolation and identification of *Fusarium* Spp. isolated from stored potato tubers showed dry rot symptoms

From stored potato tuber showed dry rot and fungal growth, using single spore technique, different *Fusarium* spp. were isolated. All isolates were purified and sub-cultured on potato sucrose agar medium then fungal growth was transferred to Petri dishes contained the same medium for growing the isolates and examine their morphological characters. Morphological features are presented in the following Table (1) illustrated by Figure (3).

Table (1): Cultural and morphological characteristics of different *Fusarium* species.

ID no.	Colony diameter (cm)	Colony colour		ID no.	Colony diameter (cm)	Colony colour	
		Upper surface	Lower surface			Upper surface	Lower surface
F1	3.8±0.2	White	Pale to brown	F11	3.1±0.1	White	Peach to brown
F2	3.9±0.1	White to brown	Peach to brown	F12	4.7±0.2	Pale violet	Dark violet
F3	4.3±0.3	White to brown	Dark brown	F13	3.2±0.2	Pale violet	Dark violet
F4	4.1±0.1	Dark purple	Dark purple	F14	2.8±0.0	Tan to brown	Tan to brown
F5	4.1±0.1	Pale violet	Pale violet	F15	3.2±0.0	Peach	Peach to brown
F6	3.7±0.2	White	Pale to brown	F16	2.4±0.1	White	Tan to brown
F7	3.1±0.0	White to orange	Brown	F17	3.4±0.3	White	Dark violet
F8	6.2±0.1	Brown	Dark brown	F18	3.7±0.2	White	Pale to brown
F9	3.8±0.1	Tan to brown	Brown	F19	3.8±0.2	White	White
F10	3.8±0.1	Pale violet	Pale violet	F20	4.0±0.2	White	White
Diameter of colony was taken after 96 hr. of incubation at 28˚C.							



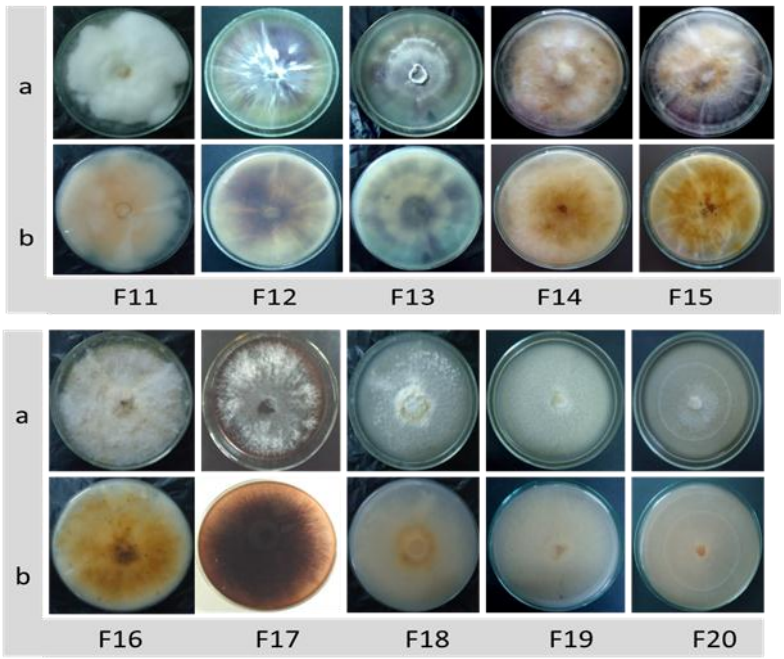


Fig. (3): Morphological features of different *Fusarium* isolates isolated from rotted potato tubers, A: The upper face of culture, B: The lower face of culture.

Moreover slide culture technique was used to appear of fine structures of all isolates in order to accomplish their identification. All features of isolated Fusaria are presented in Table (2) illustrated by Figs. (4, 5,6,7,8,9,10 and 11).

Table (2): Microscopic characteristics of different Fusarium species.

ID no.	Macroconidia						Microconidia				Chlamydospores in hyphae	Species proposed
	Apical cell shape	Basal cell shape	Length µm	Width µm	No. of septa	Chlamydospores	Shape	Length µm	Width µm	No. of septa		
F1	Blunt & rounded	Rounded or foot shaped	35.7±0.30	5.5±0.19	3-4	+	Oval to kidney	12.3±0.13	3.3±0.16	0-1	Singly & in pairs	*F. solani*
F2	Curved & tapering to a point	Foot shaped	39.4±1.30	3.9±0.26	3-5	+	Absent	-	-	-	Singly & in chains	*F. semitectum*
F3	Tapered & elongated	Foot shaped	36.0±0.54	3.7±0.15	3-5	+	Absent	-	-	-	In chains or in clumps	*F. equiseti*
F4	Tapered & curved	Foot shaped to pointed	37.7±0.54	3.3±0.26	3	-	Kidney	9.4±0.32	2.8±0.24	0	Singly	*F. oxysporum*
F5	Tapered & curved	Foot shaped to pointed	33.7±0.79	3.4±0.32	3	-	Kidney	7.1±0.39	2.7±0.13	0	Singly	*F. oxysporum*
F6	Blunt & rounded	Rounded or foot shaped	45.3±0.44	5.8±0.15	3-4	+	Kidney	11.1±0.27	3.2±0.22	0-1	Singly & pairs	*F. solani*
F7	Tapered & elongated	Foot shaped	40.2±1.08	3.5±0.39	3-7	-	Comma shaped	10.5±0.30	3.2±0.15	0	In chains or in clumps	*F. equiseti*
F8	Curved & tapering to a point	Foot shaped	31.6±1.05	2.8±0.26	3-5	+	Absent	-	-	-	Singly & in chains	*F. semitectum*
F9	Curved & tapering to a point	Foot shaped	32.6±0.65	3.7±0.15	3-5	+	Absent	-	-	-	Singly & in chains	*F. semitectum*

F10	Tapered & curved	Foot shaped to pointed	35.4±0.32	3.7±0.14	3	-	Oval or kidney	6.3±0.15	2.2±0.07	0	Singly & in chains	*F. oxysporum*
F11	Tapered & elongated	Foot shaped	47.7±1.05	4.3±0.15	3-7	-	Absent	-	-	-	In chains or in clumps	*F. equiseti*
F12	Tapered & curved	Foot shaped to pointed	34.4±1.30	3.6±0.26	3-4	-	Kidney	9.3±0.26	2.3±0.07	0	Singly	*F. oxysporum*
F13	Tapered & curved	Poorly developed	33.1±1.22	3.3±0.19	3	-	Kidney	7.0±0.27	2.3±0.13	0	Singly & in pairs	*F. oxysporum*
F14	Tapered & elongated	Foot shaped	36.8±0.79	4.1±0.37	3-5	+	Absent	-	-	-	In chains or in clumps	*F. equiseti*
F15	Curved & tapering to a point	Foot shaped	36.1±0.26	4.0±0.13	3-5	+	Absent	-	-	-	Singly & in chains	*F. semitectum*
F16	Tapered & elongate	Foot shaped	44.1±0.75	3.6±0.26	3-5	-	Absent	-	-	-	In chains or in clumps	*F. equiseti*
F17	Curved	Poorly developed	32.0±0.83	5.6±0.07	3	-	Oval	6.8±0.46	2.6±0.19	0	Singly	*Fusarium* sp.
F18	Blunt & rounded	Rounded shaped	38.3±0.79	4.5±0.14	3-4	+	Oval to Kidney	11.1±0.23	3.3±0.13	0-1	Singly & in pairs	*F. solani*
F19	Blunt & rounded	Rounded or foot shaped	45.3±0.83	5.2±0.39	3-4	+	Oval to kidney	12.6±0.39	3.7±0.15	0-1	Singly & in pairs	*F. solani*
F20	Blunt & rounded	Rounded or foot shaped	38.3±1.05	5.3±0.39	3-4	+	Oval to kidney	9.7±0.54	3.1±0.31	0-1	Singly & in pairs	*F. solani*

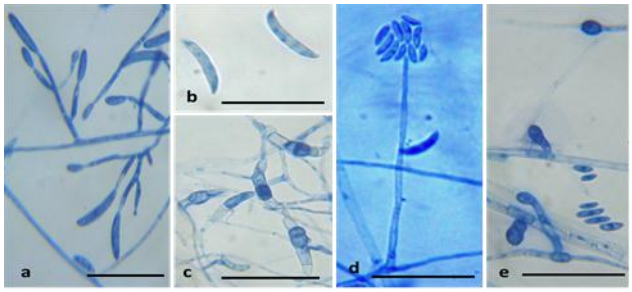


Fig. (4): F1 (*Fusarium solani*); a, branched monophialides conidiophores; b, macroconidia; c, chlamydospores in macroconidia; d, microconidia (formed in false head) produced from elongated conidiophore; e, microconidia and chlamydospores in hyphae; scale bar = 50 µm.

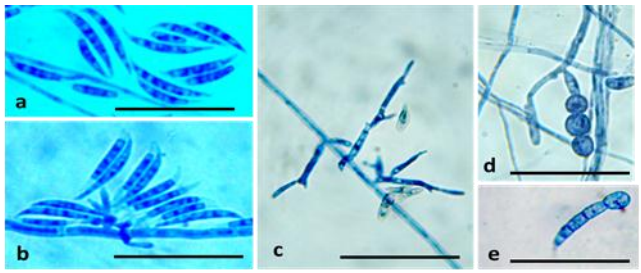


Fig. (5): F2 (*Fusarium semitectum*); a, macroconidia; b, macroconidia formed on sporodochium; c,branched and unbranched polyphialides conidiophores; d, chlamydospores in hyphae; e,chlamydospore in macroconidium; scale bar = 50 µm.

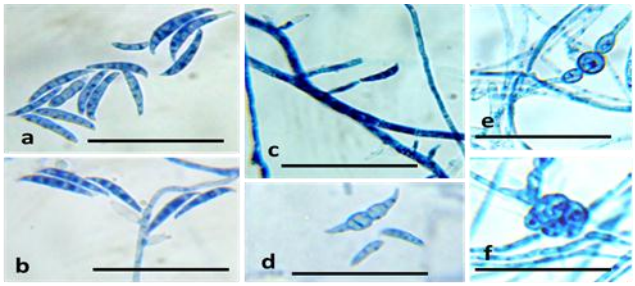


Fig. (6): F3 (*Fusarium equiseti*); a, macroconidia; b-c, macroconidia on simple lateral phialides; d, chlamidospres in macroconidium; e-f, chlamydospores in hyphae; scale bar = 50 µm.

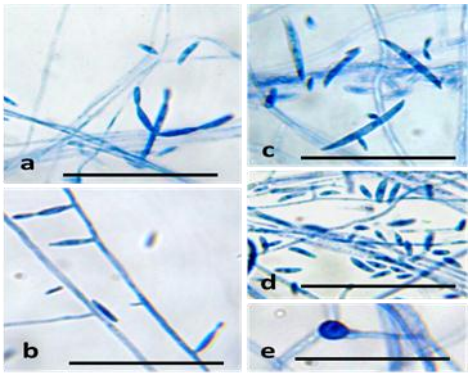


Fig. (7): F4 (*Fusarium oxysporum*); a-b, branched and unbranched conidiophores monophialides; c, macroconidia; d, microconidia; e, chlamydospore in hyphae; scale bar = 50 µm.

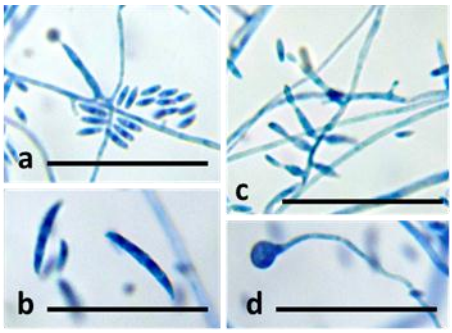


Fig. (8): F5 (*Fusarium oxysporum*); a, microconidia; b, macroconidia; c, unbranched conidiophores monophialides; d, chlamydospore in hyphae; scale bar = 50 µm.

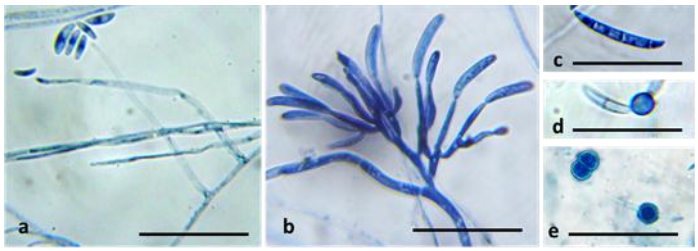


Fig. (9): F6 (*Fusarium solani*); a, microconidia (formed in false head) produced from elongated conidiophore; b, macroconidia on branched monophialides conidiophores; c, macroconidium; d, chlamydospore in macroconidium; e, chlamydospores in hyphae; scale bar = 50 µm.

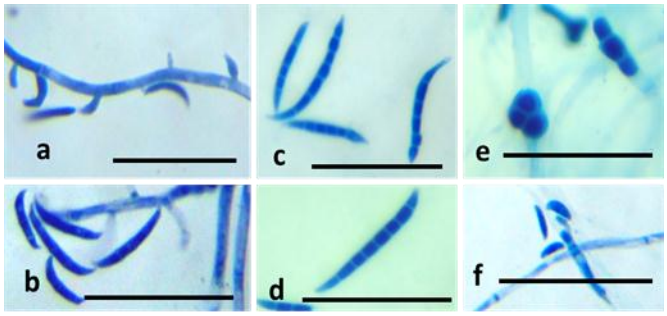


Fig. (10): F7 (*Fusarium equiseti*); a, simple phialides; b-d, macroconidia; e, chlamydospores in hyphae; f, microconidia; scale bar = 50 µm.

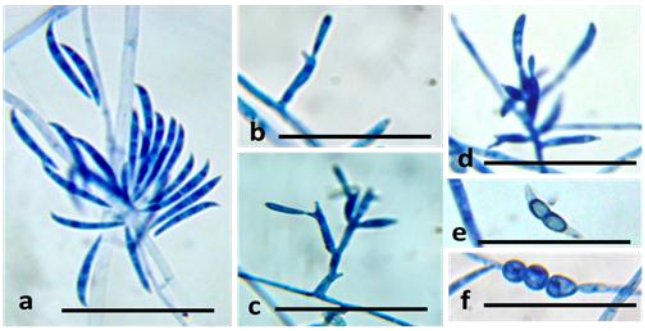


Fig. (11): F8 (*Fusarium semitectum*); a, macroconidia; b-d, macroconidia on polyphialides conidiophors; e, chlamydospores in conidium; f, chlamydospores in hyphae; scale bar = 50 µm.

According to these features *Fusarium* isolates were identified as: *F. solani* (Mart.) Apple & Wollenweber(5 isolates), *F. semitectum* (Robergo) Saccardo (4 isolates), *F. equisiti* (Corda) Saccardo(5 isolates), *F. oxysporum* Schlecht. Emend. Snyder (5 isolates) in addition to one unidentified Fusarium sp. isolate.

Pathogenicity test of isolated *Fusarium* spp

Pathogenecity test of all *Fusarium* isolates isolated from rotted potato tubers was carried out by inoculation of potato tuber slices by a piece of fungal growth putted in the central part of tuber slice. After 3 and 7 days from incubation at 24±1^{o} C the diameter of fungal growth on tuber slice was measured. Data of this experiment are presented in table (3) illustrated by Figs. (12, 13 &14).

Table (3): Pathogenicity test of *Fusarium* spp. isolated from potato tubers showing dry rot symptoms.

	Diameter of growth colony (mm) on slide surface		
Isolates	**after 3 days of inoculation**	**after 7 days of inoculation**	**Genus proposed**
F1	22.5 ± 1.91	43.0 ± 2.16	*Fusarium solani*
F6	20.0 ± 1.63	33.5 ± 2.51	
F18	12.5 ± 1.0	22.75 ± 2.21	
F19	17.5 ± 1.0	30.0 ± 0.81	
F20	6.25 ± 0.5	16.75 ± 2.36	
F4	0.0 ± 0.0	3.0 ± 1.15	*Fusarium oxysporum*
F5	6.0 ± 0.0	12.0 ± 1.63	
F10	12.5 ± 1.0	27.75 ± 2.06	
F12	0.0 ± 0.0	2.25 ± 0.5	
F13	4.0 ± 0.0	5.75 ± 0.5	
F3	0.0 ± 0.0	3.0 ± 0.81	*Fusarium equiseti*
F7	4.5 ± 0.57	8.5 ± 0.57	
F11	0.0 ± 0.0	5.25 ± 0.95	
F14	4.0 ± 0.0	8.75 ± 0.95	
F16	0.0 ± 0.0	4.75 ± 0.5	
F2	3.75 ± 0.5	8.25 ± 0.5	*Fusarium semitectum*
F8	0.0 ± 0.0	0.0 ± 0.0	

F9	5.5 ± 0.57	6.0 ± 0.81	
F15	0.0 ± 0.0	0.0 ± 0.0	
F17	7.75 ± 0.5	12.75 ± 0.95	*Fusarium* sp.
Determination of growth colony diameter on potato slices was determined after 3 or 7 days of inoculation.			

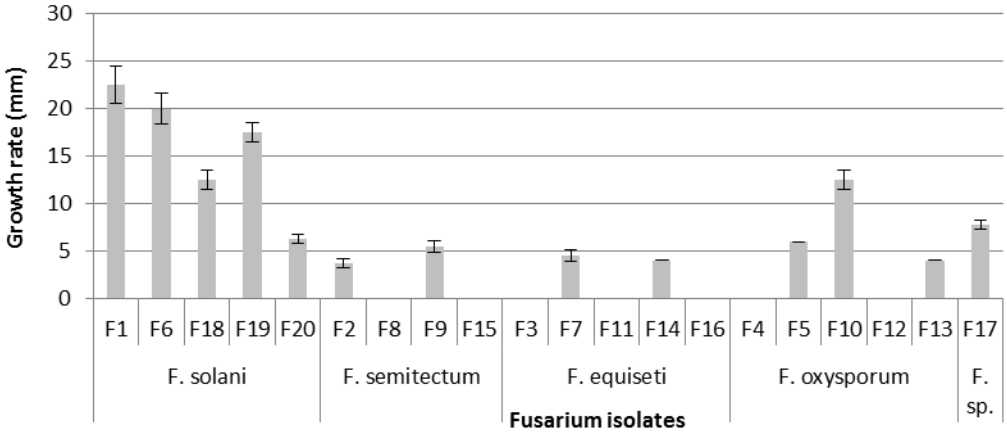


Fig. (12): Pathogenicity test of *Fusarium* spp. isolated from potato tubers showing dry rot symptoms. Determination of growth colony diameter on potato slices was determined after 3 days of inoculation.

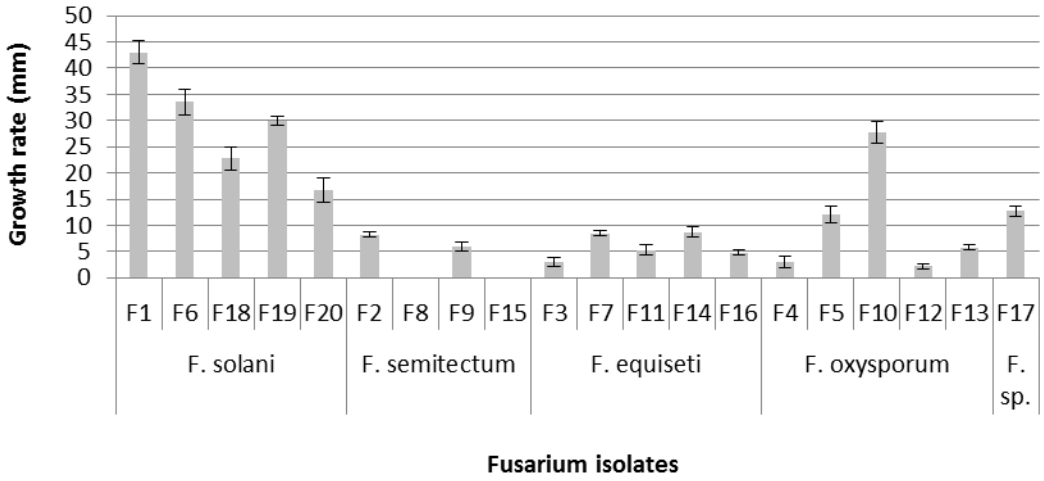
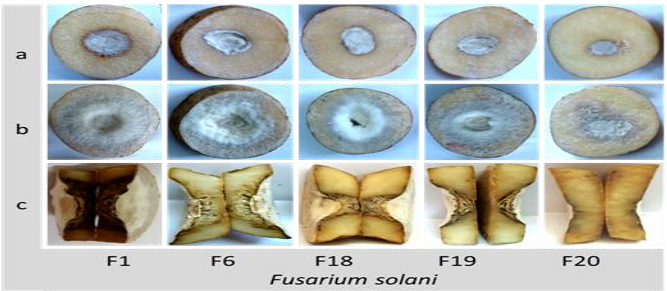


Fig. (13): Pathogenicity test of *Fusarium* spp. isolated from potato tubers showing dry rot symptoms. Determination of growth colony diameter on potato slices was determined after 7 days of inoculation.

According to these results it is clearly shown that all isolates of *F. solani* are pathogenic with different degrees of severity and they contained the most sever isolates on potato tuber slices followed by isolates belong to *F. oxysporum*. On the other hand, isolates belong to *F. semitectum* and *F. equisit* are varied in their ability to cause rots. Some isolates showed low degree of virulence, the others completely none adapted on tuber slices. As a conclusion, *F. solani* isolates are the most aggressive on tuber slices in comparison to other species. Correlation coefficient between rates of fungal growth on potato sucrose agar medium and disease severity reached -.0624.



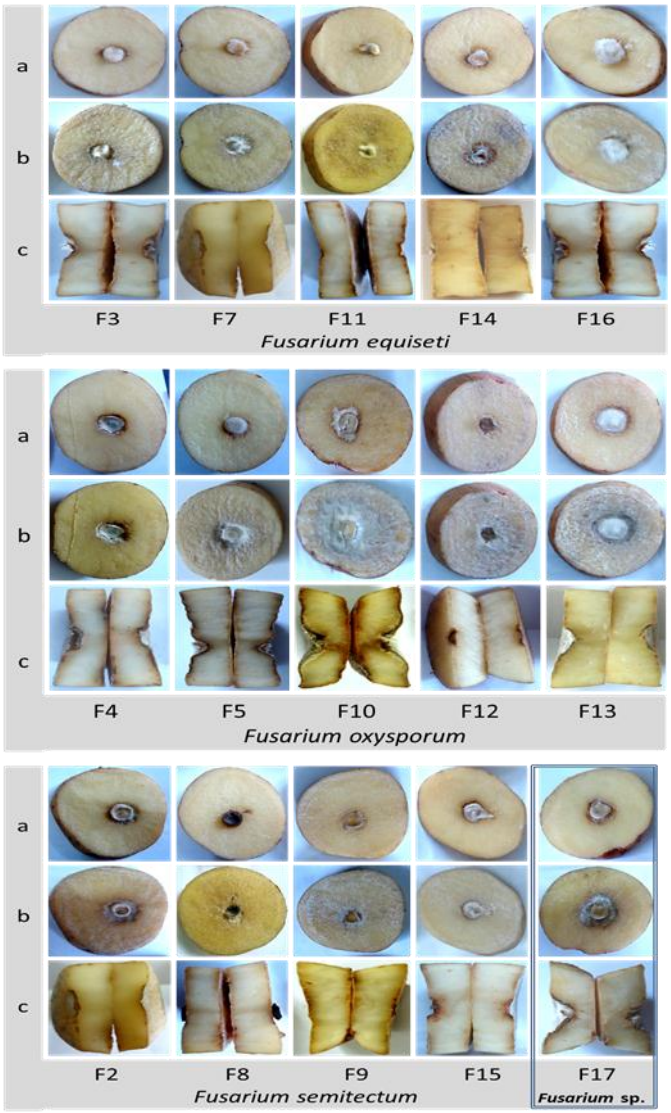


Fig. (14): Pathogenicity test of *Fusarium* isolates on potato slices; a, symptoms after 3 days of potato slices inoculation; b, symptoms after 7 days of potato slices inoculation; c, symptoms after 9 days of potato slices inoculation that showed mycelium growth in inner tissue of potato slices.

Extraction and separation of phytoalexins accumulated in potato tuber tissues inoculated by different *Fusarium* isolates

From potato tuber tissues inoculated by isolates of *Fusarium* spp. phytoalexins accumulated in such tissue were extracted and separated on silica-G plates. Separated bands were visualized by phosphomolybdic acid in methanol. Fig. (15) Indicates all separated bands. Rf values of separated bands are presented in Table (4).

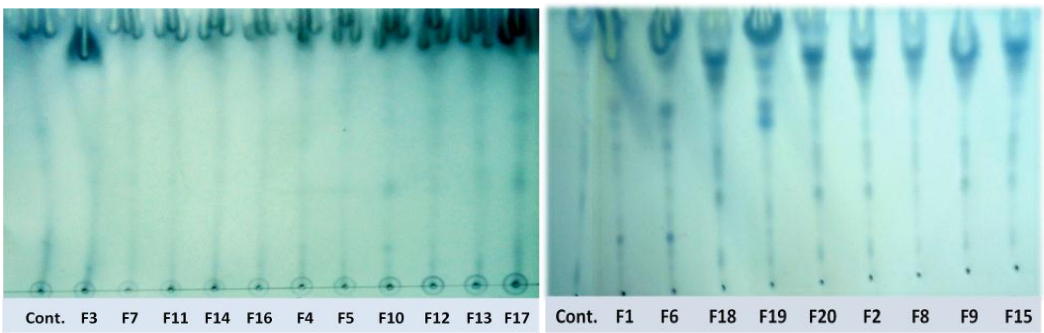


Fig. (15): TLC separation of phytoalexins accumulated in potato tuber tissues inoculated by *Fusarium* spp. isolates.

Table (4): Rf values of separated phytoalexin bands extracted from potato tuber tissues inoculated by *Fusarium* spp. isolates. Phytoalexins were extracted after 3 days of incubation at 24±1° C.

	Isolates																				
	Rf value					*F. semitectum*				*F. equiseti*					*F. oxysporum*					*F.* sp.	Control (healthy potato slices)
	F1	F6	F18	F19	F20	F2	F8	F9	F15	F3	F7	F11	F14	F16	F4	F5	F10	F12	F13	F17	
0.08	-	-	+	+	+	+	+	+	+	-	-	-	-	-	-	-	-	-	-	-	-
0.15	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	+	-	-	-	-
0.16	+	+	-	+	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
0.24	-	-	-	+	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
0.30	-	-	+	-	+	+	-	+	+	-	-	-	-	-	-	-	-	-	-	-	-
0.35	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	+	-	-	-	-
0.37	-	-	-	-	-	-	-	-	+	-	-	-	-	-	-	-	-	-	-	-	-
0.38	-	-	-	-	-	-	-	+	-	-	-	-	-	-	-	-	-	-	-	-	-
0.40	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	+	-
0.42	-	-	-	-	-	+	-	-	+	-	-	-	-	-	-	-	-	-	-	-	-
0.44	-	-	+	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
0.48	-	+	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
0.54	-	-	-	+	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
0.64	-	+	-	+	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	+	-
0.63	-	-	-	-	-	-	-	-	+	-	-	-	-	-	-	-	-	-	-	-	-
0.64	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
0.69	-	-	+	+	+	+	+	+	+	-	-	-	-	-	-	-	-	-	-	-	-
0.73	-	-	-	-	-	-	-	+	+	-	-	-	-	-	-	-	-	-	-	-	-
0.75	-	-	+	+	+	+	+	-	-	-	-	-	-	-	-	-	-	-	-	-	-
0.84	+	+	-	+	+	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
Total	**2**	**4**	**5**	**8**	**5**	**5**	**3**	**5**	**7**	**0**	**0**	**0**	**0**	**0**	**0**	**0**	**2**	**0**	**0**	**2**	**0**
D.I.	**5**	**2.75**	**1.75**	**1.5**	**2.75**	**4.5**	**1.75**	**1**	**1.75**	**3.5**	**2.25**	**1**	**2**	**2**	**1.5**	**1.75**	**2.75**	**3.75**	**4**	**3.75**	**--**

Correlation coefficient between total no. of phytoalexins bands and disease index = - 0.15.

From data recorded in Table (4) it is clearly shown that phytoalexin induction in inoculated tissues depend upon the species of *Fusarium* used to inoculate tuber tissues. All isolates belong to *F. solani* have strong ability to elicit phytoalexins in inoculated tissues. Isolate F19 elicit 8 bands followed by F18 and F20, both of them illicit 5 bands and the least isolate in phytoalexin induction is F2. Isolates belong to *F. sambucinum* elicit several bands ranged from 7 bands (F15) to 3 bands (F8). Only one isolate belong to *F.oxysporum* elicit 2 bands of phytoalexins and all isolates of *F. equisiti* have not any ability to illicit phytoalexins in inoculated tuber tissues. Correlation coefficient between disease index and number of phytoalexins elicited in inoculated tissues reached -0.15. This very low value of correlation coefficient indicate the absence of relationship between phytoalexin induction in inoculated tissues and disease index, neversless, *F. solani* isolates showed the highest level of disease index showed also the highest number of phytoalexins accumulated in infected tissues.

DISCUSSION

Potato tuber dry rot disease is one of the most important storage disease, it cause devastating problem either during storage or during cultivation. All aver the world, it was found that this disease is causing by different species of genus *Fusarium* **(Gachango *et al.*, 2012; Stefańczyk *et al.*, 2016; Falert and Akarapisan, 2019).** In an earlier study in our laboratory on identification of the species of *Fusarium* that cause this disease 33 isolates belong to genus *Fusarium* were isolated and it was found that the most prevalent species of such genus are *F. sambucinum, F. solani, F. oxysporum, F. culmorum, F. equisiti and F. semitectum.* In that study it was found that *F. sambucinum* and *F. oxysporum* were the most damaging fungi on inoculated potato tuber Spunta cv **(El-Hassan *et al.*, 2007).** In the present study, 20 isolates belong to *Fusarium* spp. were isolated from rotted potato tuber tissues. These isolates were identified according to their morphological features as *F. solani* (5 isolates), *F. oxysporum* (5 isolates), *F. equiseti* (5 isolates), *F. semitectum* (4 isolates) in addition to non identified species. Testing the ability of such isolates as dry rotted fungi indicated that all isolates of *F. solani* were the most aggressive isolates followed by isolates of *F. oxysporum* and *F. equiseti* and in the last category only one isolate of *F. semitectum* caused low degree in disease index. These results are in conflict with the earlier study, and this confliction might be due to that the variety of potato tuber tested in the present study was Balmivor cv. Would the change of tested cv. was the

reason of such deviation of disease index, this point need additional study.

Correlation coefficient between rate of growth of isolated species and disease index indicated the absence any relation between both phenomena. **The relationship between pathogenicity and growth is weak (R = -0.0624).**

Phytoalexins are low molecular weight antimicrobial compounds and they are produced in inoculated tissues due to inoculation of plants by an incompatible isolates of microorganisms or by the treatment with suitable elicitor **(Morrissey and Osbourn, 1999).** According to many studies, phytoalexins are plant resistant factors and they play considerable role in disease resistance (**Desjardins *et al.*, 1992; Morrissey and Osbourn, 1999; Delgado *et al.*, 2009).** In some cases inoculation of potato tuber tissues with *Fusarium* spp. resulted in induction and accumulation of sesquiterpenoid stress metabolites (phytoalxins) in inoculated tissues **(Desjardins *et al.*, 1992; Delgado *et al.*, 2009).** The present study was aimed to investigate the relation between disease index due to inoculation by *Fusarium* spp. isolates and accumulation of phytoalexins in inoculated potato tuber tissues. Data obtained showed clearly that inoculation of potato tuber tissues by isolated *Fusarium* spp. resulted in some cases in elicitation of phytoalexins in such tissues and the type of phytoalexin bands depend greatly on isolate used for inoculation. In some cases *F. solani* led to accumulate of huge number of phytoalexin bands. Correlation coefficient between number of phytoalexin bands and disease index reached – 0.15 which indicate that phytoalexin induction in inoculated tissues reversibly correlate with disease severity. This deceptive conclusion does not reflect the truth, because it was noticed that *Fusarium solani* isolates which had sever disease index caused accumulation of the higher number of phytoalexin bands which clearly indicate that phytoalexin induction in such case is correlate with disease severity. In this respect, Mostafa (2018) was postulated that phytoalexin accumulated in inoculated tissues by *Fusarium* spp.may be a factor of susceptibility rather that they are a factor of resistance.

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