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Fusarium Species Associated with Corm Rots and Wilt of Banana (*Musa* sp.) under Egyptian Conditions

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Isolation and pathogenicity trails revealed clearly that, *Fusarium* species were the main common fungi associated with corm rot and wilt symptoms and were pathogenic to banana plants with the exception of *F. solani* (Mart.). *F. lateratum* (Nees), *F. moniliforme* (Sheldon) and *F. oxysporum* (Schlecht.) showed the highest percent of disease incidence. Plants artificially inoculated with *F. oxysporum* developed typical symptoms of yellowing and die of outer leaves with brownish rot of the vascular tissues meanwhile, the other *Fusarium* species exhibited a brownish rot of corm tissue extending through the pseudostem tissue without leaf yellowing. Cv. Williams was the highly susceptible cultivar to all tested pathogens followed by cvs. Paradaica and Maghrabi. High performance liquid chromatography (HPLC) indicated that, level of amino acids fractions was increased in the most susceptible cultivar (Williams) inoculated with each of the tested isolates and these extent of increase was depend on fungal species and its virulence. In contrast, a clear reduction in amino acid fractions was noticed in less susceptible cv. Paradaica inoculated with *F. moniliforme*. Also, HPLC revealed both compositional and quantitative differences between phenolic compounds (ferulic acid, salicylic acid, vanilin and benzoic acid) which normally existed in the healthy corms of both cultivars. The concentration of flavones (phytoalexins) was increased in the less susceptible cultivar (Paradaica) inoculated with *F. moniliforme* as compared with that in uninoculated corms. Maxim and Moncut were the best fungicides recorded the highest percent of reduction in disease severity. Meanwhile, moderate effect was obtained with Salnit and carbendazim.

Keywords: Amino acids, banana, corm rot, fungicides and *Fusarium* spp.

Banana (*Musa* spp.) is a large handsome, herbaceous plant native to South-east Asia. In addition to the popularity of ripe banana fruit as dessert, cooked green bananas are important source of starchy food in some tropical countries (Su *et al.*, 1986). In Egypt, commercial cultivation of bananas reached 617764 tons produced from 49282 feddans (Anonymous, 2002) for production of fresh fruits mainly for local consumption. Cavendish is the main commercial cultivar including Dwarf Cavendish (cvs. Hendi and Basrii) and Giant Cavendish (cvs. Williams, Grand Nain and Maghrabi) are triploids of *M. cavendishii*. On the other hand, cvs. Paradaica and Sendehi are triploids of *M. paradisiaca* which is consumed while still starchy.

The tree like plant consists of the basal corm, a pseudostem of leaf sheaths, a terminal crown of leaves, and finally the true stem and inflorescence that arise in the meristem of the basal corm, traverse the pseudostem and emerge through the foliar crown (Su *et al.*, 1986).

During the last decades, bananas and plantains can adapt efficiently to produce high yield in a wide range of climatic extremes. Also, a number of pests and diseases have shown to be major constraints to banana production. Fusarium is a genus of fungi that cause some of the most important plant diseases affecting agriculture crops. In certain banana growing areas of the world, *Fusarium oxysporum* f.sp. *cubense* causes Fusarium wilt of banana, commonly referred to as panama disease. This disease is currently considered as one of the most destructive diseases (Jones, 2000).

Members of the genus *Fusarium* cause important diseases of banana (El-Sheikh, 1989; El-Nasr *et al.*, 1990; Mahdy *et al.*, 1993; Abd-Allah, 1994; Abdel-Hafiz, 1997 and Abdel-Kader *et al.*, 2004). In Egypt, corm and root rots became in the last years a serious problem, caused by variable soil borne fungi in some banana cultivated areas especially in new reclaimed soils (Korra, 2005). In addition, one cultivar grown by smallholder farmers in north delta called Paradaica, sometimes referred to as "local" revealed symptoms somewhat similar to those of Fusarium wilt. This cultivar was infected by Fusarium wilt, while other Cavendish cultivars were not affected by the disease even when intercropped with severely affected cv. Paradaica.

The objectives of this study were to investigate the nature of corm rot and wilt diseases of banana in Egypt. Specific biochemical changes associated with infection and its relation to relative susceptibility of different cultivars were determined.

Materials and Methods

1. Isolation of the causal pathogen:

Banana plants showing yellowing, wilt and death symptoms were used to isolate potential fungal pathogens. Samples were collected from three governorates, *i.e.* Behiera (West Nubaryia): Giza (farms on Cairo Alexandria road and El-Rahway, Om Dienar, Abo-Galeb villages in Imbaba district) and Menofyia. Small pieces (5mm) from corm, suckers, roots, pseudostem and vascular bundles region were surface sterilized with 2% sodium hypochloride for 2 min., washed with sterilized water and dry on sterilized filter paper. Sterilized pieces were placed in plates containing potato dextrose agar (PDA) medium and incubated at 25°C. A hyphal tip culture from each fungus was maintained on PDA medium and keeps at 4 °C till use. The established fungal isolates were identified on the basis of morphological and microscopical characteristics according to Nelson *et al.* (1983).

2. Pathogenicity test and cultivars reaction:

Eleven isolates representing 8 species of *Fusarium* from diseased banana were selected for its pathogenicity screening on the same cultivars from which it was originally isolated.

2.1. Selection of test plants:

Three different cultivars including Williams and Maghrabi, from Giant Cavendish cultivars groups, in addition to cv. Paradaica, the most popular cultivar grown by small-hold farmers in the Delta region were selected for evaluation of cultivars reaction. Suckers of cvs. Maghrabi and Paradaica were selected from healthy plants in farms with no history of disease symptoms and plantlets derived from meristem culture of cv. Williams were then planted in pots No.40 containing a mixture of sterilized sand and soil (1:1 v/v) for two months before inoculation. Three pots each containing one plant were used as replicates for each treatment.

2.2. Inoculum preparation and inoculation procedure:

Purified isolates were sub-cultured onto PDA plates to serve as starter colonies. The plates were incubated at 25°C for 7 days. A 5mm disk from each resulting colonies was used to inoculate 100 ml sterilized PD broth medium in 250ml flasks and incubated for 15 days at 25-30 °C for further studies. The plants were inoculated two months after planting. A deep trench was dug around the base of the pseudostem enough to reach the corm base. Equal amounts (200ml/pot) of a suspension containing mycelial growth and spores (4x 10^6 cfu/ ml) of each isolate separately were poured in the trench and buried with soil (Su *et al.*, 1986 and Sanchez-Hernandez *et al.*, 1998). Sterilized PD broth medium was added to the soil in the pots in a similar manner to be used as control. Pieces of inoculated plants were removed from the sites showing disease symptoms and were used to reisolate the pathogen originally inoculated to satisfy Koch's postulates.

3. Disease assessment:

The disease was assessed one month after inoculation and the final assessment was carried out after two months. Scale adopted by Moore *et al.* (1993) was used to assign scores to various levels of external and internal disease symptoms.

4. Biochemical changes:

Fresh tissue of each diseased sample of cv. Williams corms inoculated with *F. moniliforme, F. lateratum, F. oxysporum* and cv. Paradaica inoculated with *F. moniliforme* (*F. lateratum* and *F. oxysporum* were not pathogenic) as well as uninoculated control samples from two cultivars were used for studying the biochemical changes. Each fraction was quantitively measured as mg/g fresh weight.

4.1. Fractioning of amino acids:

Free amino acids were extracted according to the proposed method of Shad *et al.* (2002). Two grams of each sample were soaked separately in 75% ethanol (100ml). After 24 hr. the sample was ground and filtered through Whatman No. 1 filter paper. The residue was washed with a few ml of 75% ethanol and the volume was made up to 100 ml. Several amino acids were examined using a HPLC system (HP1050) with a UV detector at 254 nm. The separation was accomplished with a ODS, C18 (5µm.4 x 250mm) column.

The mobile phase consists of 32 % (acetonitril/tetrahydrofuran, 90/10 v/v); and 64 % (tetrahydrofuran/water, 5/95 v/v) with 0.3 ml acetic acid and pH adjusted 5.15 with 1 M NaOH. The flow rate was 1.5 ml/min. the temperature of column was 60°C, while the injection volume was 10 µl according to the method of Gertz (1990).

4.2. Fractioning of Phenolic compounds:

Ten grams of fresh tissues of each sample were homogenized in 40% methanol and stirred on a shaker. The extract was filtered through a Whatman No. 1 filter paper and the solvent was evaporated by a vacuum. The dried residue containing phenols compounds was dissolved in a solution consisting of methanol/water/acetic acid, 40/59.3/0.7, v/v) and stored in vials. The method suggested by Gertz (1990) was used as follows: HPLC analysis was used to detect and determine the phenolic compounds from the plant tissue. The extract was passed through micro-filter 0.45µm. the analysis of phenolic compounds was performed on a model (*hp* 1050). HPLC equipped with UV detector. The separation and determination were performed on C18 column (150 x 4.6mm). The mobile phase yielded results of methanol: water: acetic acid, 40:59.3:0.7, v/v/v). The wave length in the UV detector was 254 nm. Total run time for the separation was approximately 25 min at a flow rate of one ml/min.

4.3. Phytoalexin content:

Phytoalexin content was determined (as flavones compound) according to the method described by Gertz (1990). One gram of fresh tissue was homogenized in 40% methanol and stirred on a shaker for two hours. The extract was filtered through Whatman No.1 filter paper and the solvent was evaporated in a vacuum. The dry residue was dissolved in one ml of solution consisting of methanol: 0.1 acetate buffer PH2 (1:1 v/v) and stored in vials. HPLC analysis was used to detect phytoalexin (isoflavone) from plant tissue. The extract was passed through micro-filter 0.45µm. the analysis of these compounds was performed on a model (HP1050). HPLC equipped with UV detector. The separation and determination were performed on C18-SAX Colum (250 x 4.6 mm). The mobile phase consists of methanol: 0.1 acetate buffer PH2 (1:1 v/v). The wave length in the UV detector was 254 nm. The total run time for separation was approximately 25min at flow rate of 1 ml/min.

5. Disease control:

Banana plantlets (cv. Williams) derived from meristem culture grown in pots (8cm. in diameter) filled with peat-moss and sand (as 1:1 w/w) were used for evaluation of different fungicides on controlling disease under greenhouse conditions. All plantlets were inoculated, in separate treatments with the most pathogenic isolates of any of *F. moniliforme, F. lateratum* and *F. oxysporum* as mentioned from pathogenicity test. One week later, equal amounts from the preparations of any of the tested fungicides, *i.e.* Carbendazim, Maxim, Moncut and Salnit, at the recommended dose were added to each pot in each treatment (one plant for each pot). Disease severity was assessed after one month from fungicidal treatments.

Results

1. Symptoms and development of the disease:

1.1. Wilt symptoms:

Symptoms of banana wilt were noticed firstly in some villages at Imbaba district (El-Rahawy and others) on the local cultivar Paradaica only. The external symptoms were appeared firstly as yellowing of the lower leaves. Leaf margin turns pale green

to yellow which eventually die off and the outer leaves die completely and hang down the pseudostem as a skirt (Fig. 1). Bunches are found to be small with short and thin fingers. The yellowing then progresses inwards towards the leaf midrib and extends to the younger leaves. When the pseudostem of infected plant is cut longitudinally, the vascular tissue shows brownish dots and streaks which can be continuous along the pseudostem. When the rhizome is split open, the infected tissues shows brown spots (Fig. 1).

1.2. Corm rot:

The affected plants appeared weak, small and stunted. Rhizome (corm) tissues of the infected banana exhibited discoloration which appeared as a brownish blotch rot areas when transverse or longitudinal cuts are made (Fig. 2). Rapid rotting extends through the entire leaf sheaths of pseudostem and similar discoloration of pseudostem tissues is clearly visible. Leaves do not yellow but remain green and die in severe infection.

2. Isolated microorganisms and their pathogenic capabilities:

Six different Fusarium species including, *F. equiseti* (Corda) Sacc., *F. lateratum, F. moniliforme, F. oxysporum, F. solani* and *F. subglutinans* (Wollenw & Reinking) were isolated from diseased samples of banana collected from affected orchards in three different governorates. Percentage of disease incidence under natural infection ranged from 1% to 10.0% (Table 1). Different *Fusarium* spp. isolates and isolated bacteria were used to investigate the pathogenic capabilities on young banana plants. Data in Table (1) show that, all of *F. equiseti, F. lateratum,* and *F. moniliforme* (isolates F1, F2 and F3), *F. oxysporum* and *F. subglutinans* were able to induce different disease reactions when inoculated in the same cultivars that originally obtained from it under natural infection conditions (Fig. 3). *F. lateratum, F. moniliforme* (isolate F1) and *F. oxysporum* from Behiera governorate were the most virulent one scored 83.33, 72.22 and 66.67% disease severity on rhizome (corm), respectively, and 88.89, 55.56 and 66.67% on pseudostem, respectively. Meanwhile, the percentage of disease severity caused by *F. moniliforme* (isolate F3), *F. equiseti* and *F. subglutinans* were moderate being 22.22, 27.78 and 33.33% on rhizome, respectively, and 33.33, 22.22 and 22.22% on pseudostem, respectively. The other isolated fungi did not show any disease symptoms.

3. Reactions of cultivars to Fusarium isolates:

Three different cultivars of banana were used to study their susceptibility to pathogenic isolates of *Fusarium* sp. Data (Table 2) indicate that the susceptibility of cultivars to test isolates were in a descending order, Williams, Paradaica and Maghrabi. Cv. Williams was the most susceptible cultivar with the tested fungi scored 53.33 & 60.01% disease severity on both rhizome and pseudostem, respectively. While, cv. Maghrabi was the least susceptible one scored 6.67 & 13.33%, respectively. In general, pathogenic isolates originally obtained from cv. Williams were more virulent on the same cultivar than on cvs. Paradaica and Maghrabi. Similarly, *F. oxysporum* from cv. Paradaica was virulent on the same cultivar only. Most Fusarium species, regardless their original hosts were virulent on cv. Williams. On the other hand, *F. equiseti* caused the least disease severity on both rhizome and pseudostem.

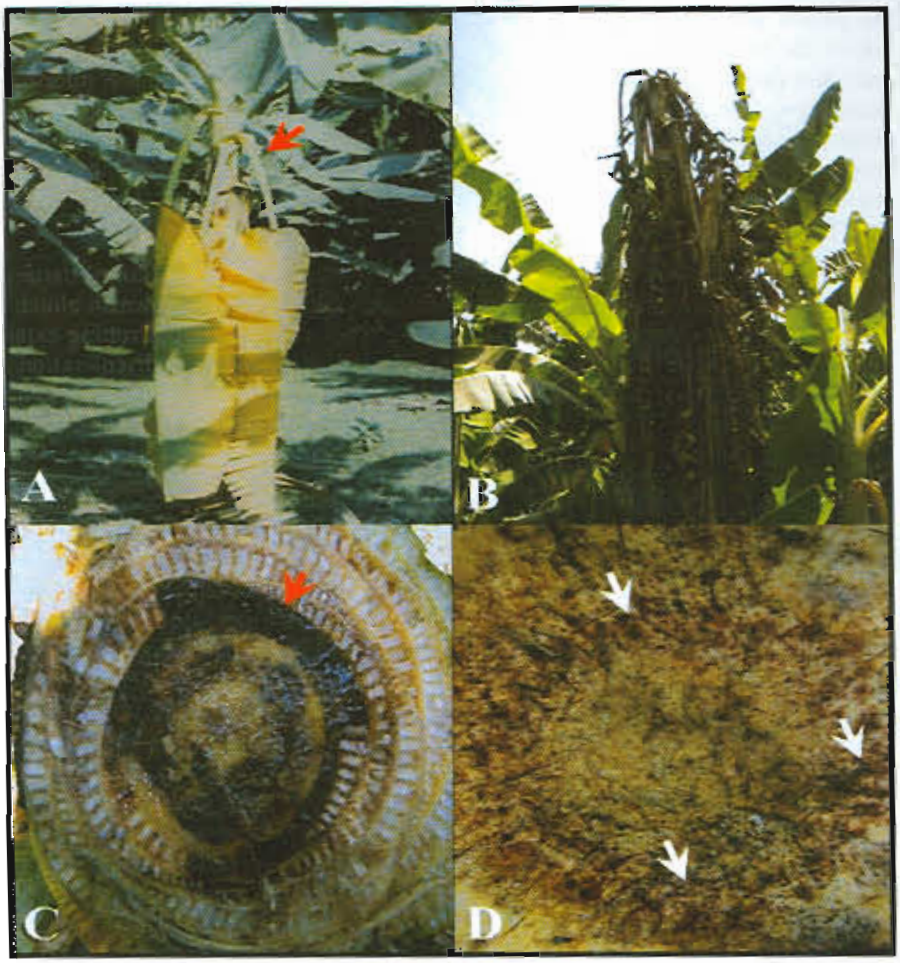

Fig. 1. An early stage of *Fusarium* wilt on cv. Paradaica showing characteristic yellowing and breakdown of the leaves (A, B). Transverse sections showing discoloured vascular bundles of pseudostem (C) characteristic of wilt disease and decayed areas in corm (D).

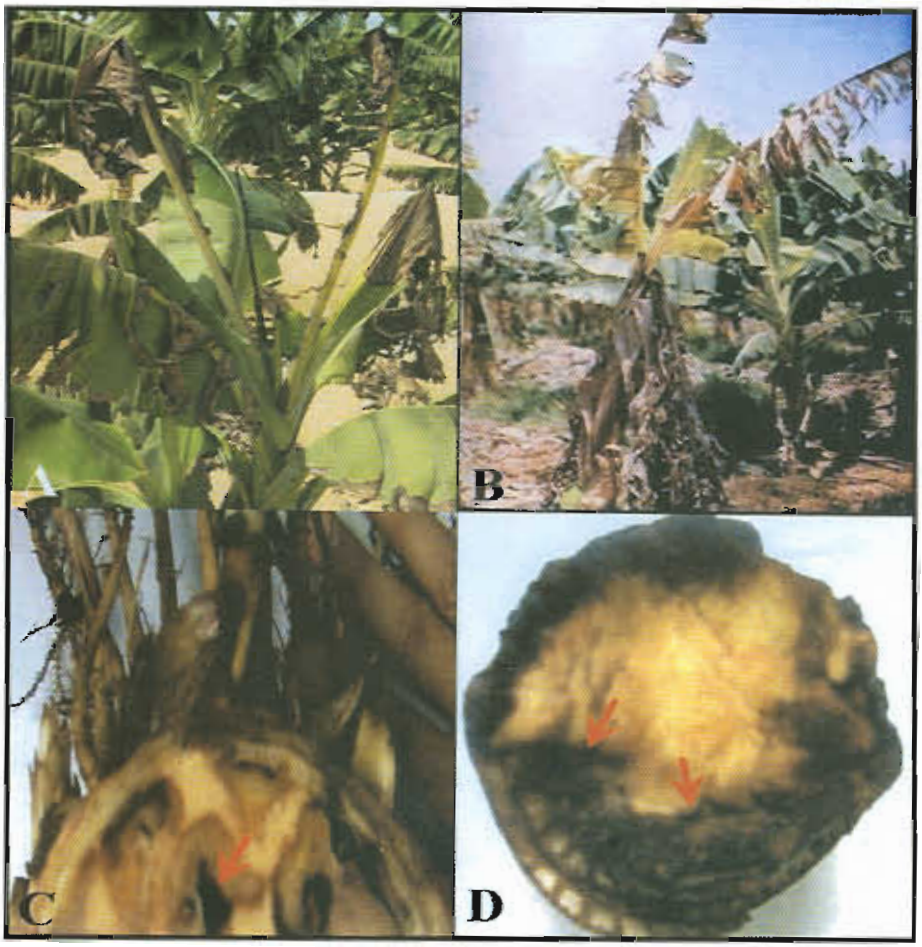

Fig. 2. A. an early stage of corm rots disease. B. most leaves were died with disease progress. C, D. Transverse sections of corm showing decayed areas distributed through corm tissues, note the roots still healthy(C).

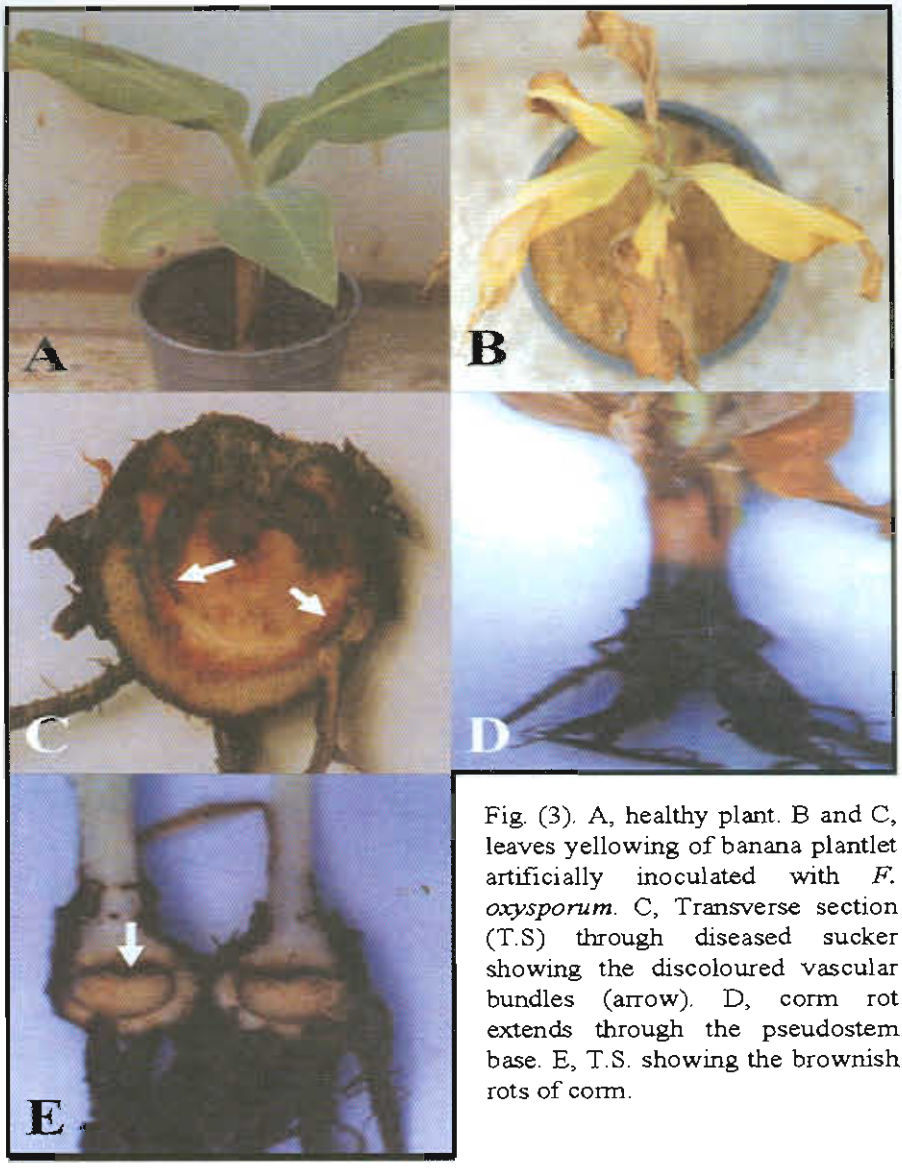

Fig. (3). A, healthy plant. B and C, leaves yellowing of banana plantlet artificially inoculated with *F. oxysporum*. C, Transverse section (T.S) through diseased sucker showing the discoloured vascular bundles (arrow). D, corm rot extends through the pseudostem base. E, T.S. showing the brownish rots of corm.

Table 1. Isolated fungi from diseased banana plants from three locations in Egypt and their pathogenic capabilities under greenhouse conditions

Isolated fungus	Locality	Original cultivar	Natural symptoms	Disease incidence (%)	Disease severity (%)	
					Rhizome	Pseudostem
Fusarium oxysporum	Giza	Paradaica	Wilt – leaf yellowing	10.0	55.56	55.56
F. oxysporum	Behiera	Williams	Plant death	N.D *	66.67	66.67
F. equiseti	Behiera	Williams	Root rot	1.0	27.78	22.22
F. subglutinans	Giza	Paradaica	Wilt – leaf yellowing	N.D	33.33	33.33
F. lateratum	Behiera	Williams	Plant death	N.D	83.33	88.89
F. solani	Behiera	Grand Nain	Corm, pseudostem, heart rot	1.0	0.00	0.00
	Giza	Paradaica	Wilt – leave yellowing	3-4	0.00	0.00
F. moniliforme	Behiera(F1)	Williams	Corm, pseudostem, heart rot	3.0	72.22	55.56
	Behiera(F2)				66.67	66.67
	Behiera(F3)	Grand Nain			22.22	33.33
	Menofyia(F4)				0.00	0.00
Fusarium spp.		Paradaica	Wilt – leaf yellowing	N.D	0.00	0.00

* N.D. = Not detected.

Table 2. Reaction of three banana cultivars against infection by various fungal pathogen isolates, under greenhouse conditions

Fungus	Original cultivar	Disease severity (%)						Mean	
		Paradaica		Maghrabi		Williams			
		Rhiz.	Pseud.	Rhiz.	Pseud.	Rhiz.	Pseud.	Rhiz.	Pseud.
F. oxysporum	Williams	00.00	00.00	11.11	11.11	66.67	66.67	25.93	25.93
F. moniliforme	Williams	50.00	44.44	22.22	55.56	72.22	55.56	48.15	51.85
F. lateratum	Williams	00.00	00.00	00.00	00.00	83.33	88.89	27.78	29.63
F. subglutinans	Paradaica	33.33	33.33	00.00	00.00	16.67	66.67	16.67	33.33
F. equiseti	Williams	00.00	00.00	00.00	00.00	27.78	22.22	9.26	7.41
Mean	--	16.67	15.56	6.67	13.33	53.33	60.01	25.56	29.63

L.S.D. at 5% level for: Fungi (F)= 1.8 . Cultivars (C)= 1.42 , Plant organ (P)= 1.16 . F x C= 3.17 . F x P= 2.59 . C x P= 2.004 . F x C x P= 4.48

4. Amino acids fractionation:

Data of amino acid contents in uninoculated and inoculated corms of two cvs. Williams and Paradaica are depicted on Table (3). According to the wide range of data obtained, it was found that there are 14 basic fractions of amino acids in the inoculated and uninoculated corms in both cultivars. Uninoculated corms of cv. Williams contained low of amino acids fractions being 4 amino acids as compared with inoculated ones or uninoculated corms of cv. Paradaica. This level of amino acids fractions was increased in the most susceptible cultivar (Williams) inoculated with each of the tested isolates of *F. lateratum, F. moniliforme* and *F. oxysporum* and this extent of increase depended on fungal species and its virulence. In contrast, a clear reduction in amino acid fractions was noticed in the less susceptible cv. Paradaica inoculated with *F. moniliforme*. Also, it was found that the concentration of amino acids was decreased in both cultivars as a result of infection compared with the uninoculated corms.

Table 3. Fractions of free amino acids of cv. Williams inoculated with *F. lateratum, F. moniliforme* and *F. oxysporum* and cv. Paradaica inoculated with *F. moniliforme*

Amino acids	Concentration (mg /g. fresh weigh)					
	Williams				Paradaica	
	F. lat.	*F. mon.*	*F. oxy.*	Cont.	*F. mon.*	Cont.
Aspartic	0.0322	0.0238	0.0000	0.0000	0.1017	0.0000
Arginine	0.1046	0.0927	0.1011	0.7102	0.0000	0.1459
Alanine	0.0421	0.0000	0.3034	0.2381	0.0000	0.0586
Cysteine	0.0428	0.0391	0.1951	0.0000	0.0000	0.0509
Histidin	0.0421	0.0381	0.0000	0.0000	0.0000	0.0000
Glutamic	0.0425	0.0407	0.2084	0.0000	0.0000	0.0000
Glycine	0.0430	0.0000	0.0000	0.0000	0.0368	0.0000
Cystine	0.0247	0.0298	0.0000	0.0000	0.0259	0.0337
Proline	0.0000	0.0133	0.0000	0.0000	0.0116	0.0000
Methionine	0.0000	0.0114	0.0000	0.0000	0.0000	0.0000
Lysine	0.0000	0.0000	0.0079	0.0000	0.0000	0.0000
Leucin	0.0000	0.0000	0.0020	0.0000	0.0000	0.0000
Valine	0.0000	0.0000	0.0000	0.0037	0.0000	0.0067
Tryptophan	0.0000	0.0000	0.0000	0.0048	0.0000	0.0176
Total No.	8	8	6	4	4	6
Total amount	0.3315	0.2915	0.8179	0.9568	0.1760	0.3134

Generally, the conclusion of amino acids analysis in healthy and diseased corms could be drawn and classified into the following groups: i) some amino acids have disappeared as a result of infection. For example, tryptophan and valine in both cvs. Williams and Paradaica infected with different pathogens and arginine in case of cv. Paradaica infected with *F. moniliforme* (Table 3). ii) Amino acids newly formed in the infected parts and normally have not existed in the healthy corms. The amino

acids cystine and glutamic were detected in corms of cv. Williams infected with different pathogens; aspartic, methionine in case of infection with *F. lateratum* and lysine, leucin in case of *F. oxysporum*. On the other hand, amounts of aspartic, glycine and proline were detected in case of cv. Paradaica infected with *F. moniliforme* being 0.102, 0.037 and 0.012 mg/g fresh weight, respectively. iii) Includes amino acid quantities decreased or increased as compared with the control. The amounts of arginine was decreased in corms of cv. Williams infected with different pathogens, alanine in case of infection with *F. lateratum*. Meanwhile, the amount of cystine was decreased from 0.034 mg/g to 0.026 mg/g in case of infected corms of cv. Paradaica with *F. moniliforme*.

5. Phenolic compounds fractionation:

The accumulation of phenolic compounds in banana corms exposed to different pathogens was investigated. Data in Table (4) indicate that there were six basic fractions of phenolic compounds in inoculated and uninoculated corms of both cultivars. High performance liquid chromatography (HPLC) revealed both compositional and quantitative differences between phenolic compounds (ferulic acid, salicylic acid, vanilin and benzoic acid) which normally existed in the healthy corms of both cultivars.

Also, these differences was found between those constitutively present phenols (coumarin and syringic acid) which were newly formed as a result of infection with different pathogens and was not existed in the healthy plants (Table 4). In general, most of phenolic quantities decreased compared with the control.

Table 4. Phenolic compound fractions of banana cvs. Williams and Paradaica inoculated with *F. lateratum*, *F. moniliforme* and *F. oxysporum*

Phenolic compound	Concentration (mg/g)					
	Williams				Paradaica	
	F. lat.	*F. mon.*	*F. oxy.*	Cont.	*F. mon.*	Cont.
Ferulic acid	0.178	0.082	0.000	0.120	0.000	0.114
Syringic acid	0.000	0.000	0.000	0.000	0.136	0.000
Vanilin	0.000	0.000	0.000	0.000	0.000	0.032
Coumarin	0.012	0.004	0.000	0.000	0.000	0.000
Salicylic acid	0.000	0.000	0.000	0.064	0.000	0.008
Benzoic acid	0.000	0.000	0.043	0.144	0.000	0.000

6. Phytoalexin content:

Data in Table (5) revealed quantities changes were detected in flavonoid content (phytoalexin) according to the cultivar and the fungus used. The concentration of flavones were increased in the less susceptible cultivar (Paradaica) inoculated with *F. moniliforme*(being , 0.010 mg / g fresh weigh) as compared with that in uninoculated corms (being, 0.0039 mg / g fresh weigh). The same result was recorded for cv. Williams inoculated with the most virulent fungus (*F. lateratum*).

Table 5. Flavonoid content (mg/g fresh weight) in two banana cultivars due to infection with different *Fusarium* species

Treatment	Flavones concentration (mg/g fresh weigh)			
	Williams			Paradaica
	F. lat.	*F. mon.*	*F. oxy.*	*F. mon.*
Inoculated	0.017	0.011	0.012	0.010
Uninoculated	0.015			0.0039

7. Disease control:

The efficacies of four different fungicides on both corms rot and wilt disease incidence in Williams cultivar was evaluated under greenhouse conditions. Data presented in Table (6) revealed that all treatments have significantly reduced disease severity when used after one weak from inoculation as soil drench. Maxim and Moncut were the best fungicides recorded the highest percent of reduction in disease severity (ranged from 88.89 to 100 %). Moderate effect was obtained with Salnit and carbendazim against *F. moniliforme* and *F. lateratum*. All fungicides used recorded the full reduction in disease severity when used against *F. oxysporum*.

Table 6. Reduction in disease severity of banana plants (cv. Williams) affected with the recommended dose of different fungicides

Fungicide	Dose/l	Disease severity (D.S.) %			Reduction in D.S. %		
		F. mon.	*F. lat.*	*F. oxy.*	*F. mon.*	*F. lat.*	*F. oxy*
Moncut 25%wp	3g	11.11	00.00	00.00	88.89	100.0	100.0
Salnit 20% wp	3g	44.44	55.56	00.00	55.56	37.50	100.0
Maxim 3.5% FS	3ml	00.00	00.00	00.00	100.0	100.0	100.0
Carbendazim 50%wp	3g	55.56	66.67	00.00	44.44	24.99	100.0
Control	--	100.0	88.89	66.67	--	--	--

Discussion

Currently, there is very little information known about the presence of wilt and corm rot of banana in Egypt. Study constitutes on attempt to assess the presence of both diseases in banana orchards. Observations under field conditions revealed similar symptoms of wilt and corm diseases reported by many workers (Su *et al.*, 1986; Beer *et al.*, 2001and Kangire and Rutherfold, 2001). The results indicate clearly that, *Fusarium* species were the main common associated fungi with disease symptoms. Pathogenicity tests demonstrated that all tested species were pathogenic to banana plants except *F. solani. F. lateratum, F. moniliforme* and *F. oxysporum* showed the highest percent of disease incidence. All the Fusarium species found have been reported in other banana-growing areas in Egypt and worldwide associated with a wide range of symptoms other than corm rot and wilt (El-Sheikh, 1989; El-Nasr *et al.*, 1990; Mahdy *et al.*, 1993; Abd-Allah, 1994;

Abdel-Hafiz, 1997 and Abdel-Kader *et al.*, 2004). Symptoms development varied on banana plants according to the fungal pathogen. Plants artificially inoculated with *F. oxysporum* developed typical symptoms of yellowing and die of outer leaves with brownish rot of the vascular tissues. The other *Fusarium* species exhibited a brownish rot of corm tissue that extend through the pseudostem tissue without leaf yellowing (Kangire and Rutherfold, 2001and Beer *et al.*, 2001).

Most of the affected farms with corm rot have a combination of some stress factors such as poor drainage, salinity and sometimes some farms have a history of disease incidence on the previous crops.

The susceptibility of three banana cultivars to infection with *Fusarium* species indicated significant differences in disease susceptibility among cultivars tested according to fungal pathogen and / or isolate used. Isolate of *F. oxysporum* originally obtained from cv. Paradaica was pathogenic only to the same cultivar. This result confirmed the field observations about the susceptibility of cv. Paradaica among different cultivars grown in El-Rahawy village, Imbaba district, Giza to infect with the causal pathogen of wilt. This result is in agreement with that obtained by Kung'u *et al.* (2001). Farmers in these affected areas report that removed affected banana mats and replanted with the same cultivar (cv. Paradaica) develop similar external and internal symptoms as would normally occur through replanting on areas already affected by the disease. Meanwhile, no symptoms were observed in other cultivars grown in the same areas. Also, these differences caused by different isolates irrespective of host cultivar; suggest differences in isolate virulence (Kung'u and Jeffries, 2001). Generally, cv. Williams was the highly susceptible cultivar to all tested pathogens followed by cvs. Paradaica and Maghrabi. This agrees with previous results on a wide range of symptoms other than corm rot and wilt of banana (Abdel-Hafiz, 1997 and Korra, 2005). Such differences might be due to differences in their genetic structure which probably affect the quality and quantity of excretions released from wounds which may play a role as food base for the pathogen in the inoculated tissues (Blackeman and Fokkema, 1982). So that, the biochemical changes in the components of infected plants were studied. After the infection has been established, plant pathogens grow and multiply on and in host plants by consuming components of host plants as nutrients. When the components are suitable for pathogen nutrients, the disease outbreak may be severe. On the contrary, the plants in which components are unsuitable for the pathogen may be resistant (Oku, 1994). In this study, amino acids analysis in healthy and diseased parts of banana revealed that, uninoculated corms of cv. Williams contained less amino acids as compared with the inoculated ones or uninoculated corms of cv. Paradaica. This level of amino acids was increased in the most susceptible cultivar (Williams) inoculated with each of the tested isolates and the extent of increase depend on the fungal species and its virulence. In contrast, a clear reduction in amino acids was noticed in the less susceptible cv. Paradaica inoculated with *F. moniliforme*. In several cases, susceptibility or resistance seemed to be correlated with the content of one or a few amino acids (Van Andel, 1966). Lakshminarayanan, 1955 found that wilt resistant varieties of cotton were found to contain a considerable amount of cystine in contrast to the susceptible varieties which decrease the effect of wilt toxin,

lycomarasmin. in cotton by chelation with iron.(note the less susceptible cv. Paradaica). Tryptophan and valine were found in both cvs. Williams and Paradaica and disappeared as a result of infection with different pathogens. The amino acid tryptophan promotes the effect of gibberellins produced by *Gibberella* spp. on rice plants (Van Andel, 1966). This finding may explain the weakness of plants and the small bunches appeared associated with infection.

Some amino acids have newly formed in infected parts and normally were not existed in the healthy corms and other quantities decreased, disappeared or increased as compared with the control. Also, it was found that the concentration of amino acids was decreased in both cultivars as a result of infection compared with the uninoculated corms. As the disease progresses the amino acids content of the tissue usually decreased as shown in wheat plants infected by the rust pathogen (Shaw and Colotelo, 1961). The decrease in amino acids content in infected tissue is sometimes attributed to utilization by the pathogen, but disappearing amino acids are not always the ones used most easily by the fungus (Van Andel, 1966). An increase in the content of certain amino acids is often accompanied by decrease of a number of others. Also, this increase may be due to decomposition of host protein or decreased protein synthesis where diseased tissue often shows higher proteolytic activity than the healthy one (Kiraly and Farkas, 1959). Synthesis of amino acids by the growing fungus would also cause an increase in the amino acid content of the invaded cells (Van Andel, 1966). With such varying results of the qualitative and quantitative changes in the composition of amino acids in plant tissue during infection, it is difficult to draw conclusion as to a general trend.

On the other hand, susceptibility and resistance are often found to be correlated not only with amino acids content, but simultaneously with different factors in which case it is difficult to decide whether one factor is more important than the other (Van Andel, 1966 and Weinhold, 1964). Among these factors, phenolic compounds and phytoalexins. In this study, high-performance liquid chromatography (HPLC) revealed six basic fractions of phenolic compounds in inoculated and uninoculated corms of both cultivars. Many workers have proposed that the defensive strategy of plants exists in two stages. The first is assumed to involve the rapid accumulation of phenols at the infection site; secondary responses would involve the activation of specific defences such as the synthesis of phytoalexins or other stress-related substances (Matern and Kneusel, 1988). Numerous studies suggest that low molecular weight phenols such as benzoic acid are formed in the initial response of infection (Nicholson and Hammerschmidt, 1992). A common host response is the esterification of ferulic acid to the host cell wall which has been leads to the formation of lignin-like polymers (Matern and Kneusel, 1988). Ascensao and Dubery (2003) found that the synthesis of cinnamic and benzoic acids derivatives in the root tissue of banana infected with *F. oxysporum* f.sp. *cubense* that were esterified followed by incorporation into the cell wall fraction as part of the anti-microbial defences activated. Métraux (2001) reported that salicylic acid (SA) has an important role in the signalling pathway leading to induced systemic resistance (ISR). After infection, endogenous levels of SA increase locally and systemically, and SA levels increase in the phloem before

ISR occurs (Rasmussen *et al.*, 1991). The inhibitory activity of phenols on pectinolytic enzymes produced by pathogenic fungi was recorded by many workers and might contribute to the resistance of hosts by preventing the growth of the pathogen within the host. (Kosuge, 1969 and Oku, 1994).

In general, most of phenolic fractions quantities decreased as compared with the control. This may be attributed to that the dead cells as a result of infection which accumulate low concentration of phenols because these cells are dead and phenols compounds are oxidized and polymerized ,and hence cannot be analyzed as phenols (Oku,1994and Ascensao and Dubery,2003).

Similarly, phytoalexins which are synthesized in living cells (Oku, 1994) affected by the ingress of pathogens and accumulate in dead or dying cells as disease defence (Hargreaves and Bailey, 1978). Furthermore, although many phytoalexins are phenols, these compounds are diverse and grouped together as a family based on their antimicrobial properties (Nicholson and Hammerschmidt, 1992). Flavonoids were found to accumulate in many plants tissues surrounding lesions of incompatible interactions (Hammerschmidt and Nicholson, 1977).

Maxim and Moncut were the best fungicides recorded the highest percent of reduction in disease severity. Meanwhile, moderate effect was obtained with Salnit and carbendazim. These results are in harmony with those obtained by Abdel-Kader *et al.*, 2004; Korra, 2005 and Shalaby *et al.*, 2007. Narendrappa and Gowda (1995) indicated that losses of Fusarium wilt can be kept to an economic level by following disease management schedules which includes the use of replanting dipping in carbendazim for 45 min and eradication of infected plants together with injection or drenches of carbendazim.

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أنواع الفيوزاريم المصاحبة لأعفان الكورمات والذبول فى الموز تحت الظروف المصرية

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أظهرت عمليات العزل وإختبار المرضية أن أنواع الفيوزاريم كانت هى الفطريات الرئيسية المصاحبة لأعفان الكورمات والذبول فى الموز وكانت قادرة على إصابة نباتات الموز عدا الفطر فيوزاريم سولانى.الفطر فيوزاريم لاتيراتم وفيوزاريم مونيليفورم وفيوزاريم اوكسيسبورم كانت الأعلى قدرة على إحداث الإصابة. أظهر الفطر فيوزاريم اوكسيسبورم الأعراض النموذجية لإصفرار وموت الأوراق الخارجية مع تلون الأنسجة الوعائية باللون البنى بينما سببت باقى الفطريات تلون أنسجة الكورمة باللون البنى والذى إمتد لأنسجة الساق الكاذبة دون إصفرار الأوراق. الصنف وليامز كان أكثر الأصناف قابلية للإصابة بكل الفطريات المختبرة يلية صنفى الباراديكا والمغربى.أظهر التحليل الكيميائى بإستخدام HPLC أن مستوى الأحماض الأمينية قد إزداد فى الصنف وليامز الأكثر قابلية للإصابة نتيجة للإصابة وأن مستوى هذه الزيادة كان معتمداً على نوع الفطر وقدرتة المرضية. فى الجانب الأخر كان هناك إنخفاض واضح فى مستواها فى الصنف باراديكا الأقل قابلية للإصابة والمعدى بالفطر فيوزاريم مونيليفورم. تفريد المركبات الفينولية أظهر وجود إختلافات تركيبية وكمية بين المركبات الفينولية(الفانيلين وأحماض الفيريوليك والساليسيلك والبنزويك) والتى كانت موجودة طبيعيا فى الكورمات السليمة لكلا الصنفين. تركيز الفلافونات (فيتواليكسن) إزداد فى الصنف الأقل قابلية للإصابة (باراديكا) والمعدى بالفطر فيوزاريم مونيليفورم مقارنة بمستواها فى الكورمات السليمة. المبيدين ماكسيم ومون كت كانا الأكثر فعالية فى خفض شدة الإصابة فى حين أعطت مبيدات الساليتيت والكربندازيم تأثيرات متوسطة.