

Survey of Some Date Palm Pests and Their Associated Fungi in Al- Qassim Region

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ABSTRACT

Date palm pests are considered the most important problem facing the development of date palm plantation. Some of these pests and their associated fungi were surveyed in Al- Qassim region, Saudi Arabia. Lesser date moth (*Batrachedra amydracula*) , Date mites (*Oligonychus afrasiaticus*), Fruit stalk borer (*Oryctes elegans*), White scale(*Parlatoria blanchardi*), Saw toothed Grain Beetle (*Oryzaephilus surinamensis*) and Mealy bug (*Maconellicoccus hirsutus*) were found. Both plant pathogenic and nonpathogenic fungi to date palm trees which associated with these pests were isolated and identified. Nine fungal genera (*Alternaria*, *Aspergillus*, *Cladosporium*, *Fusarium*, *Helminthosporium*, *Mucor*, *Penicillium*, *Rhizoctonia* and *Rhizopus*) were recorded. Among *Fusarium* spp., *Fusarium oxysporum* which considered the most prevalent one was found. On the other hand, the species that may potentially produce mycotoxins were investigated. *Aspergillus flavus* and *Aspergillus parasiticus* that produced Aflatoxins were identified.

INTRODUCTION

Date palm (*Phoenix dactylifera* L.) is considered one of the most important commercial crops in the Arab world (FAO, 1984). In Saudi Arabia, date palm trees are grown on about 90% of the cultivated land (Shaheen, 1990). The fruits of date palm are vital components of the diet in Saudi Arabia and eaten at all stages of the fruit development (Imad, 2003). Fungi attack date palm trees at all stages of ripening on trees as well as during storage and processing (Ahmed and Robinson, 1998). Pests which attack date palm are considered the main sources of these fungi (Saxena et al., 1988). Various pests attack date palm trees in Saudi Arabia. These include: *Derelomus* sp., *Batrachedra amydracula*, *Arenipses sabella*, *Oligonychus afrasiaticus*, *Oryctes elegans*, *Parlatoria blanchardi*, *Phonapate frontalis*, *Rhynchophorus ferrugineus*, *Pseudophilus testaceus*, *Microtermes najdensis*, *Ommatissus binotatus*, *Maconellicoccus hirsutus* , *Schistocerca gregaria* and *Oryzaephilus surinamensis* (Hammad et al., 1982, Al-Ahmadi and Salem, 1995, and Abraham et al., 1998). In addition to the direct damage of these pests to date palm, feeding wounds provide an entry point for plant pathogens that weaken trees making them

becoming susceptible to infection, and these pests may also carry and transmit plant pathogens in many crops such as maize and peanut (Holbrook *et al.*, 1997 and Cardwell *et al.*, 1999). *Aspergillus* spp. are considered the most important genus because they produce aflatoxins, a potent hepatocarcinogenic secondary metabolites (Lynch, and Wilson, 1991). Aflatoxins are mutagenic, carcinogenic, teratogenic and acutely toxic to most animals and human (Phillips, 1980). *Aspergillus flavus* and *A. parasiticus* which produced aflatoxins have been found at all stages of developing date palm and associated with date products (Ahmed *et al.*, 1997). In addition, many genera of fungi were identified as a fungal contaminates of date palm culture and date fruits such as *Penicillium* sp., *Curvularia* sp., *Cladosporium* sp., *Fusarium* sp. and *Alternaria* sp. (Abdul Rahman *et al.*, 2004 and Omamor *et al.*, 2003). The objectives of this study were, 1) to survey and identify the association between insect pests of date palm and fungi , 2) to identify the toxigenic, plant pathogenic fungi and the diseases transmitted to date palm trees.

MATERIALS AND METHODS

Collection of pests from date palm trees :

Pests from various parts of the tree including leaves, stems (trunks), and fruits, were collected from 150 growing date palm trees in 10 orchards located in Al-Qassim region, Saudi Arabia during 2004-2005 seasons, and transferred to the laboratory in closed bags to identify the pests according to (Hammad *et al.*, 1982, Al-Ahmadi and Salem 1995, and Meyer, 1987).

Isolation, purification and identification of various fungi:

Czapek's- sucrose agar medium was used for isolating fungi. Chloramphenicol (20 µg/ml) and rose Bengal (30 ppm) were used as bacteriostatic agents. Four plates were used to isolate fungi from each pest, Plates were incubated at 28 °C for one week. The developing fungi were counted and identified according to the following references: Booth, 1971, Ellis, 1976, Raper and Fennell 1977, and Pitt 1979.

Preliminary detection of aflatoxin- producing fungi:

Isolation of *Aspergillus flavus* and *A. parasiticus* recovered from the collected date palm pests were screened for their ability to produce aflatoxins on Sabouraud – yeast extract agar plates, using the fluorescent agar technique (El-Baize *et al.*, 1982). Each isolate was inoculated as a single short streak at the center of the plate surface. Plates were then incubated at 25 °C for two weeks , viewed under UV light (366nm), and the presence of any fluorescence in the medium surrounding the fungal growth

were recorded. A plate of non-inoculated medium was similarly incubated and kept as a control. This control was used to rule out any fluorescence that may be produced by the constituents of the medium.

RESULTS AND DISCUSSION

Nine genus of fungi namely, *Alternaria*, *Aspergillus*, *Cladosporium*, *Fusarium*, *Helminthosporium*, *Mucor*, *Penicillium*, *Rhizoctonia* and *Rhizopus* were identified after been isolated from the collected pests (Fig.1). Many investigators have isolated numerous associating fungi with pests. Aisagbonhi (2003) found *Aspergillus*, *Rhizopus* and *Trichoderma* sp. consistently associated with *Oligonyhus monoceros*. Also, Sitri Rahmlah *et al.*(1993) isolated *Aspergillus* spp. from *Metisa plana* infesting oil palm trees in Malaysia. Our results show that *Aspergillus* spp. was found to be the most abundant genus on date palm trees and has association with all tested pests, followed by *Alternaria* , *Fusarium* and *Cladosporium* sp. (Table 1). The frequency of *Aspergillus* sp. was 71.4% on *Batrachedra amydraula*, 33.4% on *Oligonyhus afrasiaticus*, 50% on *Oryzaephilus surinamensis*, 37.3% on *Maconellicoccus hirsutus*, and 41.8 % on *Parlatoria blanchardi*. *Penicillium* sp. was the most abundant in *Oryctes elegans* where the recorded frequency was 37.5 % , while the frequency of *Fusarium* spp. isolated from *Oryctes elegans* was 25%. Also, *Alternaria* and *Cladosporium* sp. were isolated from the pests which attack date palm leaf such as *Oligonyhus afrasiaticus* and *Maconellicoccus hirsutus* . *Rhizopus* was the least abundant genus, associated with tested pests. Three species of *Aspergillus*, *A. niger* *A. flavus* *A. parasiticus*, were isolated and identified from the tested pests (Table 2). *Oligonyhus afrasiaticus* and *Maconellicoccus hirsutus* were found to contain more isolates of *A. flavus* with 50% in each. On the other hand *Oryctes elegans* and *Parlatoria blanchardi* contained more isolates of *A. parasiticus* where the recorded

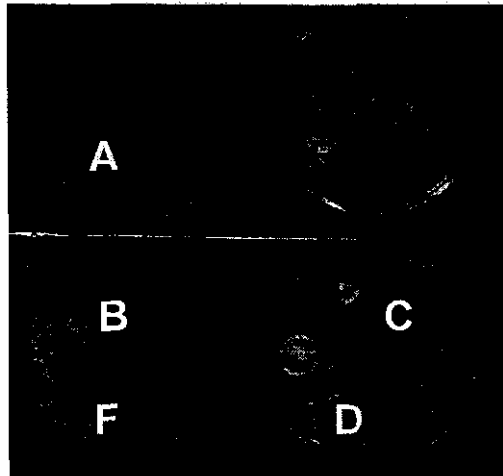


Fig.1: Different fungi isolated from pests of date palm trees showing *Aspergillus* spp.(A, B, C, D) , *Fusarium* sp.(E), *Rhizoctonia* sp.(F) in Petri dishes.

values were 75 and 40%, respectively. Similar studies were performed by many researchers and found existing association between insects and *Aspergillus* spp. (Beti *et al.*, 1995, Setamou *et al.*, 1998 and Wilson *et al.*, 1984). The production of Aflatoxins by *A. flavus* and *A. parasiticus* increased when plants are contaminated with insects. Lynch and Wilson (1991), found that the amount of aflatoxin in peanut pods which penetrated by insects is thirty to sixty times greater than the aflatoxin levels found in undamaged peanut pods. On the other hand Fatih *et al.* (2005) found that many species of *Aspergillus* isolated from tomato plants may potentially generate mycotoxins. These species that produce mycotoxins include *Aspergillus flavus* (aflatoxin, sterigmatistin and derivatives), *A. parasiticus* (aflatoxin), *A. terreus* (Sitrinin, patulin) and *A. fumigatus* (fumitremorgen). The ability of toxins produced by *Aspergillus flavus* and *A. parasiticus* on the growth and formation of aflatoxins fungi were tested using Sabouraud – yeast extract medium, and incubation period of two weeks at 25 °C. This temperature has been reported to be the optimal condition for aflatoxins production (Lieu and Bullerman, 1977). The detection of aflatoxins was confirmed using the fluorescent agar technique. All tested isolates of *A. flavus* and *A. parasiticus* gave fluorescence in the medium, indicating that these isolates produce aflatoxins. Pests may also carry and transmit pathogens to human. In the present study, *Aspergillus*

niger was found in almost of all tested pests (Table 1). *A. niger* infects human causing many diseases such as Otomycosis ear infection and Keratomycosis eye infection (Everett and Alvin, 1996). Also, tested pests found to transmit many plant pathogens, especially to date palm trees (Table 2). These plant pathogens include *Fusarium oxysporum* which causes Fusarium wilt disease in date palm (El-Meleigi *et al.*, 1993), *Alternaria alternate* which causes the Rectangular pale brown spots in date palm leaves (Sheir *et al.*, 1981), and *Cladosporium cladosporoides* which causes the Longitudinal brown spots in the date palm leaves (Kassim *et al.*, 1983). It can be concluded that pests can play important role in transmitting dangerous injurious fungi that produce toxic aflatoxins and/or serious diseases to humans or their domestic animal, and economic crops.

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Table 1: Number, type and percentage of micro flora associated with different pests of date palm trees, Qassim, 2004-2005.

Insect pest	No. of sampled insect pests	Associated fungi	% of fungi
<i>Batrachedra amydraula</i>	30	<i>Aspergillus</i> sp.	71.4
		<i>Penicillium</i> sp.	14.3
		<i>Rhizopus</i> sp.	14.3
<i>Oligonyhus afrasiaticus</i>	30	<i>Aspergillus</i> sp.	33.4
		<i>Alternaria</i> sp.	25.0
		<i>Cladosporium</i> sp.	16.7
		<i>Fusarium</i> sp.	8.3
		<i>Penicillium</i> sp.	8.3
<i>Oryctes elegans</i>	18	<i>Rhizopus</i> sp.	8.3
		<i>Aspergillus</i> sp.	12.5
		<i>Fusarium</i> sp.	25.0
		<i>Mucor</i> sp.	12.5
		<i>Penicillium</i> sp.	37.5
<i>Parlatoria blanchardi</i>	30	<i>Rhizopus</i> sp.	12.5
		<i>Aspergillus</i> sp.	41.8
		<i>Alternaria</i> sp.	8.3
		<i>Cladosporium</i> sp.	8.3
		<i>Fusarium</i> sp.	16.7
		<i>Mucor</i> sp.	8.3
<i>Oryzaephilus surinamensis</i>	30	<i>Penicillium</i> sp.	8.3
		<i>Rhizopus</i> sp.	8.3
		<i>Aspergillus</i> sp.	50.0
		<i>Fusarium</i> sp.	12.5
<i>Maconellicoccus hirsutus</i>	30	<i>Penicillium</i> sp.	12.5
		<i>Rhizopus</i> sp.	25.0
		<i>Aspergillus</i> sp.	37.3
		<i>Alternaria</i> sp.	25.0
		<i>Cladosporium</i> sp.	25.0
		<i>Rhizopus</i> sp.	12.5

Table 2: Identified fungal species and their frequency in association to pests of date palm trees.

Insect pest	Species of fungi				
	<i>Aspergillus</i> sp.	%of species	<i>Fusarium</i> <i>oxysporum</i>	<i>Alternaria</i> <i>alternata</i>	<i>Cladosporium</i> <i>clasdosporiodes</i>
<i>Batrachedra</i> <i>amydraula</i>	<i>A. flavus</i>	20.0			
	<i>A. parasiticus</i>	20.0	-	-	-
	<i>A. niger</i>	60.0			
<i>Oligonyhus</i> <i>afraasiaticus</i>	<i>A. flavus</i>	50.0			
	<i>A. parasiticus</i>	25.0	+	+	+
	<i>A. niger</i>	25.0			
<i>Oryctes</i> <i>elegans</i>	<i>A. flavus</i>	0.0			
	<i>A. parasiticus</i>	75.0	+	-	-
	<i>A. niger</i>	25.0			
<i>Parlatoria</i> <i>blanchardi</i>	<i>A. flavus</i>	20.0			
	<i>A. parasiticus</i>	40.0	+	+	+
	<i>A. niger</i>	40.0			
<i>Oryzaephilus</i> <i>surinamensis</i>	<i>A. flavus</i>	20.0			
	<i>A. parasiticus</i>	20.0	+	-	-
	<i>A. niger</i>	60.0			
<i>Maconellicoccus</i> <i>hirsutus</i>	<i>A. flavus</i>	50.0			
	<i>A. parasiticus</i>	0.0	-	+	+
	<i>A. niger</i>	50.0			

(-) Not found, (+) Found.

الملخص العربي

حصر لبعض أفات نخيل البلح والفطريات المصاحبة لها في منطقة القصيم

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تعتبر أفات أشجار نخيل البلح من اخطر المشكلات التي تواجه تطور وزراعة أشجار النخيل . تم للحصر والتعرف على بعض من هذه الأفات وكذلك الفطريات المحمولة والمصاحبة لها في منطقة القصيم بالمملكة العربية السعودية. وكان من أهم الأفات التي تم رصدها على أشجار النخيل دودة السبلح الصغرى و حلم الغبار و حفار عذوق النخيل و حشرة النخيل القشرية البيضاء و خنفساء التمر المنشارية (خنفساء سورينام) و حشرة البق الدقيقي . من ناحية أخرى تم عزل كل من الفطريات الممرضة والغير ممرضة لأشجار نخيل البلح من هذه الأفات الحشرية المذكورة سابقا وكان عددها تسعة أجناس وهي : *Rhizoctonia* , *Cladosporium*, *Fusarium*, *Aspergillus*, *Alternaria* *Rhizopus* , *Penicillium*, *Mucor*, *Helminthosporium* . وكان من بين أنواع جنس *Fusarium* التي وجدت الفطر *Fusarium oxysporum* والذي يعتبر من أهم الأنواع الممرضة لأشجار نخيل البلح , كما تم التعرف على الأنواع التي لها القدرة على إفراز سموم فطرية وكان فطري *Aspergillus flavus* و *Aspergillus parasiticus* من بين أنواع جنس *Aspergillus* السائدة والتي لها القدرة على إفراز سموم الافلاتوكسين.