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Survey of *Fusarium* species in an arid environment of Bahrain. VI. Biodiversity of the genus Fusarium in root-soil ecosystem of halophytic date palm (*Phoenix dactylifera*) community

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Abstract – Date palm (*Phoenix dactylifera*) as an indigenous halophytic tree was selected as a target host to evaluate biodiversity of the genus *Fusarium* in the north-west transect of the main island of Bahrain. A total of 27 villages were thoroughly surveyed and 81 samples of roots, plant debris and soil were collected from the rhizosphere soil of small, medium and large plants. Soils samples were found mostly saline, high in soluble salts, calcareous, gypsiferous, poor in organic matter and sandy to coarse in texture. Data assessment for recovery of *Fusarium* species was based on colony identification and plate counts by direct plating of roots, plant debris and soil on selective media. A sum of 2107 isolates, fluctuating between 1 to 345 per sample, were encountered among all transects, plant heights, media and isolation type. Jid Ali village yielded the highest colony counts (162.56). Mean recovery of *Fusarium* isolates was highest in plant debris (723) followed by roots (540) and lowest in soil samples (410). A gradual increase in colony counts from root samples were observed proportionally related to growth in plant height from small to large. A total of thirteen species were recovered representing eight sections, twelve from roots and six from plant debris and soil samples each. The isolated species were *F. avenaceum, F. chlamydosporum*, *F. equiseti, F. illudens, F. lateritium*, *F. moniliforme*. *var. subglutinans*, *F. oxysporum*, *F. pallidoroseum, F. poae, F. sambucinum, F. solani, F. sporotrichioides* and *F. tricinctum*. Three species, *F. avenaceum, F. illudens,* and *F. poae* are reported for the first time from Bahrain. *F. solani* and *F. oxysporum* were the most dominant and frequently encountered species, a finding confirm previously reported data and validated by chi-square contingency analysis. Northern transect resulted in higher species assemblage and isolate counts compared to west and principally isolation from root samples was superior than other isolation types. Analysis of data supported by diversity indices revealed that roots of small plants were the most diverse (0.588) followed by large plant (0.544). Overall, variations related to locations were much greater than differences attributed to isolation type or plant height. Large-medium plants combinations exhibited the highest species similarity composition (0.80) as determined by Sorensen community coefficient. Species richness among samples fluctuated between 2-10 for soil of medium plants and root of small plants. Overall, that predominance of *F. solani* and *F. oxysporum* in the rhizosphere of date palm is consistent with the hypothesis that these fungi have broad ecological tolerances and tend to be illustrative of hot arid habitats. The influence of soil biotic factors on *Fusarium* diversity and spectra is discussed.

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INTRODUCTION

Kingdom of Bahrain is a small island nation located off the central part of the Arabian Gulf consisting of an archipelago of 33 islands with a total land area of about 706 km^2. These are situated 25 km off the eastern coast of Saudi Arabia between the latitude 25° 32°`` and 26° 20`` N and longitude 50° 20`` and 50° 50`` E. The climate is typical hot Saharo-arid environment characterized by intolerable high temperature during the summer months extending from June to September and mild winter season with scanty rainfalls. Cultivated area is under great stress of drought, salinity and high temperature (Abbas & El-Oqlaha, 1992). Much of the soils are saline, calcareous, gypsiferous and sandy to coarse in texture (Doorenkamp *et al.* 1980). Also, soil is porous, poor in the organic matter content (< 0.05-1.5%) and nutrient level with low fertility and potential. The water holding capacity is low and the available soil moisture is about 2.6%. Such lands are, therefore, not regarded suitable for agriculture purposes, locally as well as worldwide (Mandeel 2002). However, this situation might raise the possibility of using saline water to irrigate salt tolerant plants (Abbas & Mandeel, 1995; Bloomberg & Edler, 1993).

Date palm (*Phoenix dactylifera*) is a commonly cultivated arboresent monocot tree in the hot arid and dry deserted region. It is one of the primary articles of food, uses and commerce in the great desert area extending from the Western North Africa to India. Bahrain is known through centuries as one of the important countries involved in the growing and trading until recent past and is traditionally famous, in particular, for the production of finest quality date palm varieties of this economically important crop in the Arabian peninsula. This practice is still going on but at a narrow scale due to negligence, oil discovery, shift of skilled labors to industrial sectors and the needs of high skills in different plant operations. There are more than 35 commercial varieties of date palm that differ mainly in ripening time, color, size, yield and sweetness. Recently, cultivated lands with dates are estimated to be less than 1500 hectares consisting of about 347000 active and 449000 neglected trees. The average yield has recently fallen to approximately 40-45 kg per plant due to increase in soil and water salinity, low soil fertility, diseases and pests, low ground water table, interference with urbanization and the nonfertilization practice.

The genus *Fusarium* is a common soil inhabitant associated with plant roots and debris, rhizospheric soil and organic matter throughout the world (Lim & Varghese 1977; Burgess 1981; Stoner 1981). *Fusarium* species are usually formed on slimy matrix facilitating dispersal by means of water rather than air indicating that the fungus is relatively uncommon member of the air microflora (Lukezic & Kaiser 1966). *Fusarium* species may persist in soil (Park 1963), as dormant chlamydospores (Nash *et al.* 1961; Nyvall 1970a; Smith & Snyder 1975; Sitton & Cook 1981), hyphae (Nyvall & Kommedahl 1968), or conidia in plant residues (Kreutzer 1972; Manzo & Clafin 1984). Nutritionally, they are active under a broad range of substrates with frequent mutation within population to adapt to specific requirements (Nelson *et al.* 1990; Mandeel 1996). Apart from their parasitic and pathogenic aspects, *Fusarium* species also play a vital role in ecological functions such as nutrient cycling (Dighton 1995), organic matter decomposition (Abdel-Hafez *et al.* 1990; Boddy & Watkinson, 1995), energy flow and synthesis of humic substances (Christensen 1989) and plant-soil inter-relationships (Burgess *et al.* 1975; Gilbertson *et al.* 1985; Kommedahl *et al.* 1987; Farias & Griffin, 1989; Abdul-Wahid *et al.* 1997; Kalc *et al.* 1997). In arid systems, these fungi colonize wide and complex

ecological niches, influencing primarily the function, form, and structure of ecosystem (Stoner 1981). The mechanism of survival of these fungi may be through the decomposition and utilization of wide range of organic substrates (Ronzani 1968; Griffin 1972; Burgess 1985; Marasas *et al.* 1988; Abdel-Hafez *et al.* 1990). The presence of microfungi as heterotrophic microorganisms inside the roots or rhizosphere soil is of ecological importance to escape unfavorable conditions especially extreme high temperature (Saremi *et al.* 1999) and salinity (Bloomberg & Adler, 1993), during summer months (May, June, July and August). Tolerance of such stressful environmental conditions and saprobic ability of *Fusarium* species are important attributes in determining their successful distribution and colonization in the soil niches (Skujins 1984; Sangalang *et al.* 1995b).

Studies on *Fusarium* ecology have focused on either climatic or soil edaphic factors. The former studies led by an Australian group (Burgess 1981; Liddle & Burgess 1985; Burgess *et al*. 1988a, Burgess & Summerell, 1992; Summerell *et al.* 1993; Backhouse & Burgess, 1995; Sangalang *et al.* 1995b; Saremi *et al.* 1999; Backhouse & Burgess, 2002) proposed that climate, especially temperature, seasonal distribution, and rainfall are dominant factors influencing the distribution of *Fusarium* species, supported by *in vitro* studies. Sangalang *et al.* (1995a) hypothesized that such factors indirectly determine water availability and consequently affect both the active and survival phase of these fungi in soil. Accordingly, the genus *Fusarium* has world wide geographical distribution, ranging from arctic tundra to tropical rainforest (Abbas, *et al.,* 1987; Marasas *et al.,* 1988; Kommedahl *et al.,* 1987, 1988; Burgess & Summerell, 1992; Jeschke *et al.,* 1990; Lim, 1974; Lim & Varghese, 1977; Lesile *et al.,* 1990 ; Mandeel, 1996; Onyike & Nelson, 1992). Nonetheless, some species are typical ubiquitous, whilst others have more restricted distribution to certain climatic regions and less prevalent in other zones. For example, *F. longipes* is more common in the tropic and subtropics (Burgess 1981) while *F. sambucinum* is common in cold-temperate and alpine soils (Sangalang *et al.* 1995b) and *F. equiseti* is widely distributed but is more frequently recovered from warm temperate and subtropical areas (Burgess *et al.* 1988). Moreover, Nelson *et al.* (1990) have related the distribution of *Fusarium* species mainly to temperature and soil osmotic potential.

Within particular climate, *Fusarium* species may be restricted in their distribution through adapting to specific soil environmental conditions (Stoner 1981; Mandeel & Abbas 1994; Abbas & Mandeel 1995; Backhouse & Burgess 1995; Mandeel *et al.* 1995; Miller 1995; Mandeel 1996). Rapid increase in soil salinity and low organic matter are most important soil edaphic factors affecting fungal growth and distribution of this arid region. Several reports focused the effect of soil salinity upon various physiological and developmental aspects of fungi (Bloomberg & Adler 1993; Castillo & Demoulin 1997). This increase in salinity has resulted in rapid deterioration of wild vegetation covers which shelter microbial community. Moreover accumulation of salts due to saline marine water may impose an environmental stress not only to plants but also to the microflora of rhizosphere (Rasmuseen & Stanghellini 1988). Pereira *et al.* (1999), have suggested that *F. moniliforme* and *F. subglutinans* can loose their pathogenecity in saline solution of NaCl (0.85%) but were able to survive in distilled water. It has been further, reported that *Fusarium* is one of the common genus in the untreated areas of high spills of brine solution and has shown high tolerance to Na Cl concentration. The ability of fungi to improve drought resistance, and to alleviate salinity stress has also been reported by Pupi & Brass (1999).

Several studies have focused on soil fungi of arid deserts and other extreme environments (Ronzani 1963; Nash & Snyder 1965; Stoner 1981; Cook &

Rayner 1984; Skujins 1984; Moubasher *et al.* 1985; Khodair *et al.* 1991). However, relatively few reports have dealt with fungi of higher desert plant roots (Moubasher *et al.* 1985; El-Abyad *et al.* 1993; Mandeel 2002). Studies on the ecology of the fungi, particularly the genus *Fusarium*, are very limited in the Saharo-Arabian phyto-geographical region. With exception of some investigation on genus *Fusarium* from arid environment of Bahrain (Mandeel *et al.* 1994; Mandeel *et al.* 1995; Abbas & Mandeel 1995; Mandeel *et al.* 1986), no previous reports were documented from this region.

The objectives set forth for the present study was to conduct a comprehensive survey on the virgin island of Bahrain on the biodiversity of *Fusarium* species in roots, soil and plant debris of the indigenous date palm tree of Arabian-Gulf desert. A total of 27 villages representing northern-west transect were extensively surveyed, including cultivated and non-cultivated, irrigated or non-irrigated, neglected, polluted, fertilized or nonfertilized with their geographical location, climate and vegetation description. Various ecological and edaphic parameters have also been considered during this study in relation to plant height to detect any possible correlation with distribution, abundance and assemblage of *Fusarium* species of arid habitats.

MATERIALS AND METHODS

Location and topography

The Island of Bahrain is the largest of the archipelago extending 48 km long from north to south and upto 16 km wide at its maximum point east to west. The general topography is lowlying to smoothly undulating. The land rises gradually from sea shores of the Arabian Gulf towards inland limestone bedrock slopes and gently ending in central rocky core of Jebel Al Dukhan (Smoky mountain), elevated up to 134 m above sea level (Abbas & El-Oqlah 1992). The monotony of the main island landscape can be categorized into five major geomorphologic zones; coastal lowlands, back slopes, escarpment, interior basin and central plateau and jebels (Doorenkamp *et al*. 1980).

Soil

The soils of Bahrain show poor profile development. They have been formed under arid conditions from Holocene and Pleistocene sedimentary rocks with little altered parent material. Five soil groups may be distinguished; cultivated solanchalk, natural solanchalk, regosols, raw mineral soil and rocky areas (Doorenkamp *et al*. 1980). Both types of solonchalk soils are located mainly in the coastal areas, extending inwards to the beginning of the backslopes. The other three soils are mainly found in the central and southern part of the island. Geomorphologically, the dominant soils are similar to other arid soils of the Arabian Peninsula in being mostly saline, slightly alkaline, calcareous, gypsiferous and coarse in texture. Furthermore, soils are poor in organic matter content (< 0.05-1.51), low in fertility, sodium adsorption ratio, total nitrogen and nutrient

Table 1. Coordinate position, date palm (*Phoenix dactylifera*) habitat description and ecological characteristics of sampling sites.

*Location Sites **	*Sample Frequency*	*Longitude N*	*Latitude E*	*Sampling site*	*Habitats*	*Male/ Female*	*No. of. Suckers*	*Manure*	*Irrigated / Fallow*	*Stem Dia-meter(cm)*	*Healthy/ Weak*	*Vegetation Cover*
BUS-S	3	261003	503229	A	NC	M	NIL	FR	IRR	175	H	M
BUS-M	3	261625	503635	W	NC	M _	NFR	FAL	150	W/DL		S
BUS-L	3	261641	503627	A	NC	M	6	NFR	FAL	145	H	S
HID-S	1	261528	503903	W	NC	F	3	NFR	FAL	160	H	M
HID-M	1	261528	503903	W	NC	F	10	NFR	FAL	122	H	S
HID-L	1	261528	503903	W	NC	M	NIL	NFR	FAL	175	H	M
AIR-S	1	261600	503710	S/A	C	M	1	FR	IRR	118	H	M
AIR-M	1	261600	503710	S/A	C	M	6	FR	IRR	113	H	M
AIR-L	1	261600	503710	S/A	C	M	5	FR	IRR	120	H	M
SAQ-S	3	261307	503426	A	NC	M	NIL	NFR	FAL	130	W/DS	S
SAQ-M	3	261320	503438	N	NC	M	NIL	NFR	FAL	134	W/D.S.L	M
SAQ-L	3	261255	503358	N	NC	F	NIL	NFR	FAL	132	H	S
HAM-S	1	260857	502711	A	C	F	NIL	FR	IRR	140	H	S
HAMM	1	260857	502711	A	C	F	NIL	FR	IRR	70	H	S
HAM-L	1	260857	502711	S,A	C	F	2	FR	IRR	180	H	S
TUB-S	3	261256	503332	A	C	F	NIL	FR	IRR	145	H	D
TUB-M	3	261140	503031	A	C	M	NIL	FR	IRR	138	H	D
TUB-L	3	261115	503314	A	C	F	NIL	FR	IRR	130	H	D
JID-S	1	261108	503324	W/N/P	NC	F	NIL	NFR	FAL	160	H	D
JID-M	1	261108	503324	W/N,P	NC	F	2	NFR	FAL	140	H	D
JID-L	1	261108	503324	W,N,P	NC	M	3	NFR	FAL	148	H	D
BUI-S	3	269460	503104	W,N,P	NC	F	NIL	NFR	FAL	150	H	S
BUI-M	3	260938	503037	W,N,P	NC	F	8	NFR	FAL	150	H	D
BUI-L	3	269460	503104	N	NC	F	NIL	NFR	FAL	300	H	D
JAS-S	1	260855	502927	A	C	F	NIL	FR	IRR	100	H	S
JAS-M	1	260855	502927	A	C	F	NIL	FR	IRR	80	W	S
JAS-L	1	260855	502927	A	C	F	NIL	FR	IRR	100	H	D
ABH-S	1	261333	503321	A	C	M	NIL	FR	IRR	300	H	M
ABH-M	1	261333	503806	A	C	F	NIL	FR	IRR	130	H	M
ABH-L	1	26133	503321	A	C	F	NIL	FR	IRR	124	H	M
DMT-S	1	260748	502808	A	C	F	NIL	FR	IRR	130	H	M

Table 1. Coordinate position, date palm (*Phoenix dactylifera*) habitat description and ecological characteristics of sampling sites *(following)*.

*Location Sites **	*Sample Frequency*	*Longitude N*	*Latitude E*	*Sampling site*	*Habitats*	*Male/ Female*	*No. of. Suckers*	*Manure*	*Irrigated / Fallow*	*Stem Dia-meter(cm)*	*Healthy/ Weak*	*Vegetation Cover*
DMT M	1	260748	502808	A	C	F	NIL	FR	IRR	150	H	M
DMT-L	1	260748	502808	A	C	F	NIL	FR	IRR	180	H	M
KAZ-S	1	260707	502815	A	NC	F	5	FR	IRR	300	H	M
KAZ-M	1	260654	502811	N,P	NC	F	5	NFR	FAL	145	H	M
KAZ-L	1	260710	502616	N,P	NC	F	NIL	NFR	FAL	150	H	S
KAR-S	2	261335	503026	A	C	F	NIL	FR	IRR	140	H	S
KAR-M	2	261335	503026	W	C	F	NIL	FR	IRR	120	H	S
KAR-L	2	261335	503026	A	C	F	NIL	FR	IRR	100	H	S
JNU-S	2	261345	502930	A	C	F	NIL	FR	IRR	140	H	M
JNU-M	2	261345	502930	A	C	F	NIL	FR	IRR	140	H	M
JNU-L	2	261345	502930	A/N	C	F	NIL	FR	IRR	140	H	S
BAR-S	2	NO	POSITION	A	NC	F	4	FR	IRR	180	H	D
BAR-M	2	NO	POSITION	W,N	NC	F	NIL	NFR	FAL	120	W	S
BAR-L	2	NO	POSITION	A	NC	F	NIL	NFR	FAL	125	W	S
MAK-S	2	NO	POSITION	A	C	F	1	FR	IRR	190	H	D
MAKM	2	260618	502857	A	C	F	1	FR	IRR	147	H	D
MAK-L	2	260618	502857	A	C	F	2	FR	IRR	126	H	D
SAR-S	1	NO	POSITION	A	C	F	10	FR	IRR	450	H	M
SAR-M	1	NO	POSITION	A	C	F	NIL	FR	IRR	160	H	M
SAR-L	1	NO	POSITION	A	C	F	NIL	FR	IRR	180	H	M
BUD-S	1	NO	POSITION	W,N	NC	M	10	NFR	FAL	300	H	D
BUD-M	1	NO	POSITION	W,N	NC	M	8	NFR	FAL	100	H/D,L	D
BUD-L	1	NO	POSITION	W,N	NC	M	2	NFR	FAL	100	H/D,L	D
SAD-S	2	265022	502919	A	C	F	NIL	FR	IRR	180	H	D
SAD-M	2	265025	502922	A	C	M	NIL	FR	IRR	160	H	D
SAD-L	2	265025	502922	A	C	F	NIL	FR	IRR	145	H	D
ZAQ-S	3	NO	POSITION	A	C	F	10	FR	IRR	200	H	D
ZAQ-M	3	NO	POSITION	A	C	F	1	FR	IRR	127	H	D
ZAQ-L	3	NO	POSITION	A	C	F	NIL	FR	IRR	170	H	D
AWI-S	1	260558	503225	W	C	M	NIL	FR	IRR	80	H	M

Table 1. Coordinate position, date palm (*Phoenix dactylifera*) habitat description and ecological characteristics of sampling sites *(following)*.

Location Sites *	*Sample Frequency*	*Longitude N*	*Latitude E*	*Sampling site*	*Habitats*	*Male/ Female*	*No. of. Suckers*	*Manure*	*Irrigated / Fallow*	*Stem Dia-meter(cm)*	*Healthy/ Weak*	*Vegetation Cover*
AWI-M	1	260558	503225	W	C	F	NIL	FR	IRR	150	H	M
AWI-L	1	260558	503225	W	C	F	NIL	FR	IRR	130	H	M
SAK-S	1	260343	503035	A	C	M	4	FR	IRR	200	H	M
SAK-M	1	260343	503035	A	C	M	NIL	FR	IRR	120	H	M
SAK-L	1	260343	503035	A	C	F	NIL	FR	IRR	140	H	M
NBI-S	1	261102	503455	N,P	NC	F	NIL	NFR	FAL	110	W/D,L,S	S
NBI-M	1	261102	503455	N,P	NC	F	2	NFR	FAL	115	W/D,L,S	S
NBI-L	1	261102	503455	N,P	NC	F	NIL	NFR	FAL	115	W/D,SLS	S
SIT-S	1	260950	503612	N,P	NC	M	2	NFR	FAL	200	W/D,S,L	S
SIT-M	1	260945	503615	N,P	NC	F	NIL	NFR	FAL	140	H	M
SIT-L	1	260945	503615	N,P	NC	F	NIL	NFR	FAL	130	H	M
HIW-S	1	261200	503245	A,S	C	F	4	FR	IRR	300	H	D
HIW-M	1	261200	503245	A,S	C	F	5	FR	IRR	170	H	D
HIW-L	1	261200	503245	A,S	C	F	4	FR	IRR	130	D	D
SNA-S	1	260913	502940	A	C	F	NIL	FR	IRR	180	H	S
SNA-M	1	260913	502940	A	C	M	3	FR	IRR	130	H	S
SNA-L	1	260913	502940	A	C	F	6	FR	IRR	180	H	S
JDF-S	1	261317	503138	A	C	F	NIL	FR	IRR	200	H	M
JDF-M	1	261317	503138	A	C	F	NIL	FR	IRR	170	H	M
JDF-L	1	261317	503138	A	C	F	NIL	FR	IRR	130	H	M

* Refer to table 3, S=Small (0.5-1 m); M = Medium (1-2 m) L = Large (2 m & above).

** A = Agriculture N = Neglected W = wild P = Polluted S=Saline.

*** C = Cultivated D = Desert H = Healthy NC=Non - cultivated.

**** M = Male F = Female.

***** FR = Fertilized NFR = Non fertilized.

****** H = Healthy W = Weak.

******* S = Sparse M = Moderate D=Dense.

level. The water holding capacity is very low and the moisture is about 2.6%, with impervious layers at depths between 75 and 300 cm. Most date palm plants cultivation areas consist of sandy to sandy loam soils where water table is close to surface (37-300 cm). Soil salinity in these locations were further complicated by excessive use of slightly underground irrigated water, absences of good drainage systems, sea water intrusion and water loss through high evapo-transpiration rates, especially during summer months.

Climate

Bahrain, like mainland Arabia, falls within the north African–Euroasian climate province. The main island has a typical Saharo-Arabian climate, generally described as mild, scanty rainy winters and hot humid rainless summers. Mean monthly rainfall, temperature, evaporation, and relative humidity in the main island is illustrated in Table 2. The annual climatological summary was obtained for the years 1999 and 2000 from the Civil Aviation affairs at Bahrain International Airport. The mean annual rainfall is 9.28 mm. Rainfall is seasonal, but irregular, usually occurring between the months of October to April. Summer rainfall is rather rare. The mean annual temperature is 27.39 °C, with a recorded mean daily maximum from May to August is 34.06 °C, and a December to March mean daily minimum is 19.51 °C. The duration between April to September are usually dry with no rains. Precipitation is low and, similarly is irregular, and mostly in the form of winter rain. Evaporation is generally high; with recorded highest mean in June (12.5 mm) and the lowest in January (4.05 mm). Solar radiation is usually high with sunny days over 300 throughout the year. The average maximum mean daily hours of sunshine is 11.5 in June and the minimum is 11.6 in March. Relative humidity is commonly high, ranging from a daily mean in January (69.5%) and daily mean in May (49.5%). Winds usually are damp, blow from the north and northwest and are known as Shamals, with an average speed of 8.95 knots hr^{-1}. Occasional hot and dry winds known as Quws, blow from southwest with an average speed of 8 knots hr^{-1} (Table 2).

Plant description

Date palm is an evergreen perennial monocot tree with unbranched woody slender stem (40-90 cm in diameter) that grows up to 15-30 m tall. The trunk is surrounded from ground upward in spiral pattern with remains of woody bases from cut leaves. Leaves are clustered together in a maximum number of 20-30 forming a loose terminal crown shaft and pinnately lobed. Each leaf consists of several pinnae (40 cm). The upper leaves are ascending, basal are recurved and the segments are loriaceous, linear, rigid, and sharply pointed dark green pigment. In young plants, the roots are small, delicate fibrous used mainly for water and mineral uptake. Older plant develops cylindrical (0.6-2 cm) usually branched root system growing horizontally 1-6 m below soil level and become hard and corky that function for anchorage. Flowers are unisexual, borne terminally in groups inside axillary spadies on dioecious plants. Fruits are commonly known as dates, are oblong berries, when ripe, up to 50 cm long in cultivated varieties, their flesh is scchariferous, it contains woody seed. The plant is usually surrounded by suckers, (offshoots) and seedlings of its own germinated seeds, which are later used as main propagating methods. The main associated plant community of date palm are *Alhagi species* and, *Zygophyllum qatarense*.

Table 2. Monthly and annual means of some climatic factors in Bahrain over the years (1999-2000)*.

Climatic factors	*Years*	***J*	*F*	*M*	*A*	*M*	*J*	*J*	*A*	*S*	*O*	*N*	*D*	*Mean annual*
Temperature C°	1999	19	20.4	21.8	27	31.3	35.1	35.1	35.7	33.7	30.3	25.2	19.7	27.8
	2000	18	17.5	20.2	27.4	31.2	33.1	35.6	35.8	32.2	29.6	23.1	19.5	26.9
	Mean	18.5	18.9	21.0	27.2	31.25	34.1	35.35	35.7	32.95	24.15	24.15	19.6	27.3
Relative Humidity (%)	1999	60	77	64	57	50	49	55	64	63	67	64	74	63
	2000	70	78	59	54	49	48	58	63	62	65	72	75	62
	Mean	69.5	72.5	61.5	55.5	49.5	48.5	56.5	63.5	62.5	66	68	74.5	62.37
Rainfall (mm)	1999	20.3	62.6	11.6										7.87
	2000	14.5									5.2	96.6	12.0	10.69
	Mean	17.4	31.3	5.8							2.6	48.3	6	9.28
Evaporation (mm)	1999	3.6	2.9	5.8	9.3	12	12.5	11.2	10.3	10.6	6.3	6.6	3.7	8.8
	2000	4.5	5.4	7.7	8.7	12.6	11.8	9.7	8.5	7.5	8.6	8.8	8.8	9.1
	Mean	4.05	4.15	5.6	9.0	12.3	12.15	11.5	9.4	9.05	7.45	7.7	6.25	8.21

* = Bahrain civil aviation department.

** = Months of a year.

Study area

The Green belt of date palm plantation area is situated in north–western part of main island, restrained by limitations on water supplies from artesian wells or de-salination stations. Study sites are located in the main island of Bahrain and other small islands of Muharraq, Sitra, and Nabi Saleh. The distance of surveyed area is about 45 km extending from west at Hammllah village to north at Zallaq village. The study sites include, cultivated, noncultivated, fertilized, nonfertilized, irrigated, neglected, wild and air-polluted locations (Fig. 1). The physiographic zone of the sampling transect falls within back slopes about 0.5-2 km from sea shore and an elevation of approximately 3-4 m above sea level. The soil group of this zone varies from cultivated solonchalk to natural solonchalk (Doornkamp *et al*. 1980). According to Abbas & El-Oqlah (1992), soils of cultivated solonchalk is distributed in the northern coastal lowlands extending from Jurdab in the east, through Manama, Diraz, Sarr, Hamlah, to Dar kulaib in the west. The soils are composed of loamy, sandy and clay subgroups. Although such soils are featured by having high water table and absence of lowlands and marshes yet, categorized as cultivated. Moreover, it is also considered to be the oldest agricultural area. All these natural characteristics emphasize the point that formation of the northern plains by means of deposits are carried by the valleys, proceeded the process of the formation of the coastal areas, in other areas of Bahrain. It is believed that Buri valley contributed to the deposits from inner basin to the northern parts of the Bahrain island. Natural solonchalk soils are found in the area located between the interior of the northern coastal lowland and the base of the backslopes, extending from Sanad in the east to Karzakan in the southwest. The soils contain gypsiferous sand, saltpans and marine mud flats. They too are characterized by having high water table.

Soil Sampling

A transect from western to northern Bahrain was established through a major date palm cultivated regions (Fig. 1). A total of 81 samples were collected representing 27 villages where date palm was dominant, irrespective of being cultivated or ornamentally grown on roadsides. Sampling of villages was performed during the period between October 1999 to March 2000. Latitude and longitude coordinates of field location were determined by, GPS 38 Personal Navigator Positioning System. Geographical locations of the sampling sites along with their ecological and cultural practices are listed in Table 1. For collection of soil samples, the surface soil (0-2 cm) was removed aside with a sterile hand trowel, and soil was obtained at a depth of 2-14 cm of the soil profile using another sterile trowel. This depth was chosen for mycological considerations since it encounters the majority of soil microfungi. Three plant heights were considered to represent the various growth stages i.e. small (0.5-1m), medium (1-2 m), and large (> 2 m) from each village.

Whenever possible and accessibility to sampling were permitted, for each plant height, a minimum of one (range 2-3) composite soil sample, each about 2 kg, were obtained. Each sample consisted of about 5 further random subsample (approximately 400g), collected as above using a clean trowel, approximately 30-40 cm around stem base. Special attention was paid to collecting immediately around root balls and loose ectorhizosphere soil from root using a clean paint brush. Plants in fields were sampled randomly, but at least 4 m apart. The subsamples were

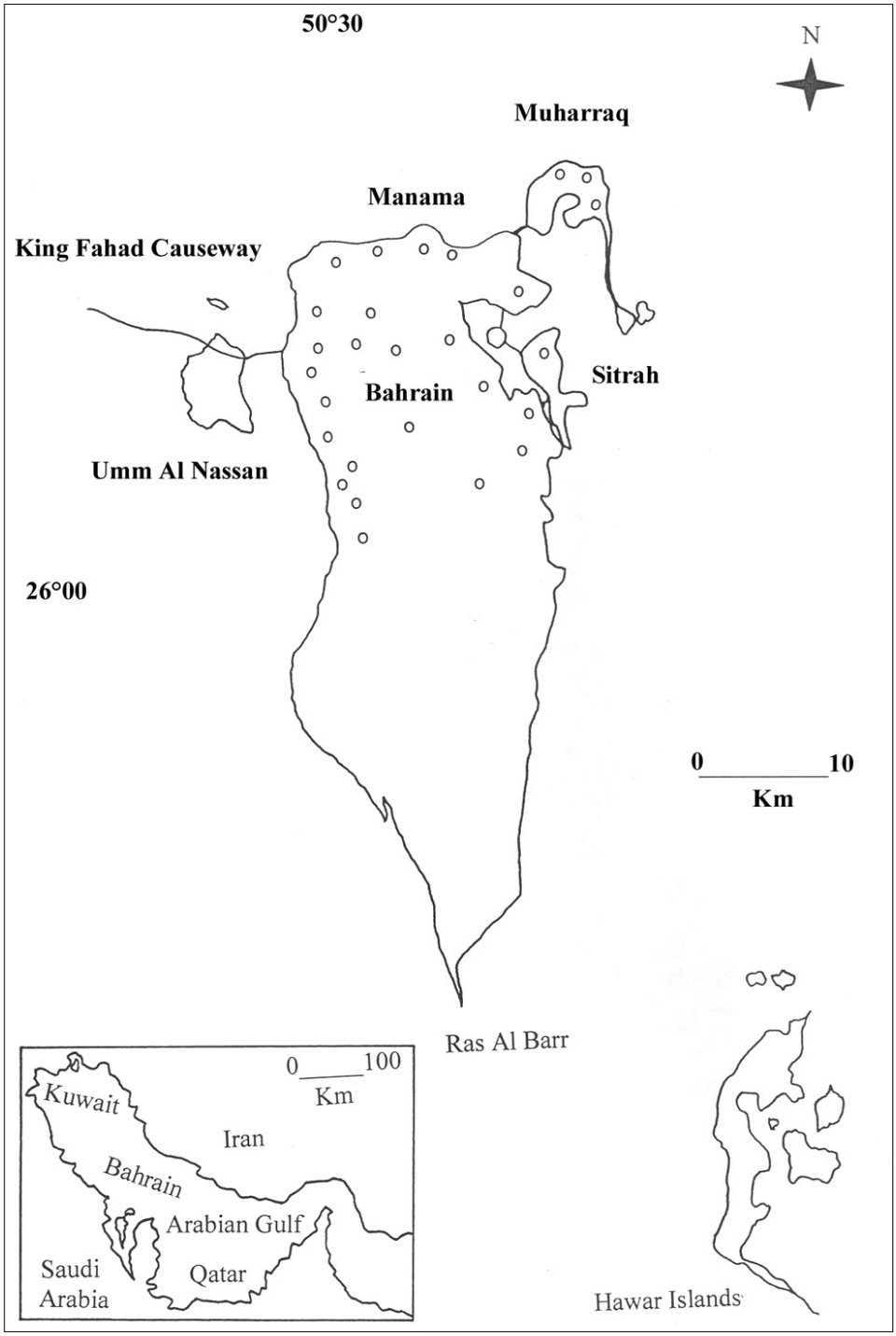

Fig. 1. Location map of Bahrain representing collection sites.

then combined and hand mixed thoroughly in polyethylene bags and labeled properly. The samples were transferred to the laboratory, on the same day, and aseptically air-dried at 25-30 °C on sterile papers for three consecutive days during which large stones, plant parts, soil crumbs and insects were removed and later stored at 5 °C until processed within usually one week (Burgess and Summerell, 1992). For the assays the composite soil samples were further thoroughly hand mixed in plastic bags under aseptic condition. One half was stored at 5 °C until used. The other half was used for physical, chemical and biological analyses.

Isolation from Roots

Root pieces approximately 50 mm × 0.5 × 1.0 mm were either cut from the intact root or recovered from soil sample after sieving at 4 mm mesh sieve. Roots of both types were washed using a fine spray for 15 min, rinsed in a sterile distilled water for 15 min, damp dried on sterile paper towel, and plated on selective medium. Three replicates of root sample per plant height per location were prepared, each having 10 root pieces of 2 cm each in length.

Isolation from plant debris

Debris was identified as non-living plant tissues. Materials retained on the screen were termed as debris, and material that passed through the screen was termed soil (Lesile *et al.,* 1990). A composite soil sample consisting of 100 g from each plant height was suspended in 500 ml sterilized distilled water and mixed thoroughly for 5 min. Soil suspension was poured off thorough a nest of sieves in order of 4 mm, 2 mm, and 0.5 mm mesh size. The first two screens retained the gravel and large pieces of plant residue, which were discarded. Debris retained on 0.5 mm mesh was carefully washed for 2 hrs under a fine mist spray of tap water. Due to small size and porous nature of debris, surface disinfectants were not used. The debris were carefully collected by sterilized forceps and blotted dried on sterile paper tissue aseptically for 24 hrs and under silica gel in a desicator for further 48 hrs before plating on selective media. Debris ranged in size from 0.3 to 0.5 mm in length and 0.1 to 0.2 mm in width were used to place on each plate. Three replicates per plant height were prepared, each having 20 pieces of debris per medium.

Isolation from soil

The second part of the samples were further air-dried, thoroughly homogenized using the mortar and pestle, crushed to a fine powder when necessary and passed through a 0.5 mm soil screen to remove parts of root fragments and other debris (Mandeel 1996). Two techniques were employed to recover *Fusarium* species from composite soil samples.

Soil dilution method and soil direct plating were used on various selective media. In the dilution plate method, 10 g of each composite soil sample was suspended in 90 ml of sterile distilled water in sterile 250 ml glass flasks and were thoroughly hand mixed for 5 min. Soil suspension was allowed to settle slowly for 1 min and a 1 ml aliquot of the 1:10 dilution was aseptically pipetted onto the surface of the below mentioned isolation media and distributed uniformly using a sterile L-shape glass rod. Because of the high variability of *Fusarium* species densities in soils of the date palm fields, preliminary soil plating was necessary to

determine optimal soil dilution. The above dilution was the most suitable since it accounts for 15-20 *Fusarium* colonies per plate.

The direct plating technique was modified from McMullen & Stack (1983). Finally ground powder of each sample of 0.05 g was evenly distributed over the surface of solidified medium. This weight of the soil resulted in almost similar number of *Fusarium* colonies. Due to high variability of soil dilution plate method and high contaimination rates as well as high salinity levels this method was replaced by direct soil plating.

Isolation media

For each composite soil sample, root pieces and organic debris, *Fusarium* species were quantified on three media. A freshly prepared potato dextrose agar (PDA), a general purpose cultivation medium for soil fungi, Komada's selective medium (Komada 1975); *Fusarium* Selective agar medium (Tio *et al*. 1977). The last two are selective for *Fusarium* species. Preliminary tests indicated PDA is not a useful isolation medium as it encounters high levels of contamination.

Komada's medium was recommended for selective isolation of *F. oxysporum* from soil, but in present study it is used for the recovery of all *Fusarium* isolate. Selective *Fusarium* agar medium, is a modified Czapec-Dox medium containing antimicrobial agents and developed for the selective isolation of *Fusarium* species from soil debris (Tio *et al*. 1977). This medium permits slow growth of *Fusarium* species from roots and soil debris. It is highly inhibitory to most fungi and bacteria but allows slow growth of *Fusarium* from small colonies. Potato dextrose agar medium is a carbohydrate rich medium, which contains enough amount of energy to support microbial growth. This medium is used for the identification and purification of *Fusarium* species only. Additionally, two more media were used for identification, 1.5% water agar and synthetic nutrient agar medium (Brayford 1993). All media were autoclaved for 15 min at 121 °C and allowed to cool at 45 °C before the addition of antibiotics, adjustment of pH, or the addition of heat liable ingredients. Fifteen ml of each medium were poured into plastic Petri dishes. Media were stored in the refrigerator for at least five days prior to use.

Incubation Conditions

All plates of roots, plant debris, and soil were incubated at 25 ± 0.1 °C for 7-10 days, under 40 cm cool, white florescent illumination with a 12 hrs photoperiod (Burgess & Summerell 1992). This temperature was selective for the mesophylic components of the population. Number of presumptive *Fusarium* colonies, on the isolation plates identifiable by their extensive fluffy white mycelium and distinctive pigmentation on the underside of the plate were counted under a compound microscope and noted separately. Growth of other fungal genera was usually restricted. The most common genera other than *Fusarium*, were *Aspergillus spp*, yeast, *Penicillium spp*, and *Alternaria spp*. All resultant *Fusarium* colonies that developed on the isolation plates were transferred individually, using hyphal tip method to standard 2% PDA slants and incubated as described above for 10-14 days.

Identification

Pure cultures were subsequently established on the three identification media using a germinated single conidium technique and further incubated under

conditions described above. Identification was based on, colony morphology, pigmentation, growth rate and morphology of conidia, and phailads, conidia produced on slide mounts. Presence and absence of chlamydospore with their shape was another important criteria of identification to delimit species. Identification of single conidial culture were made according to Nelson *et al*. (1983), Burgess *et al*. (1988b) and Brayford (1993). Representative sporulating structure of each species was permanently mounted on slide with Amman's preservative on glass slides and was deposited in the herbarium collection of the Department of Biology, University of Bahrain.

Soil moisture content determination

Fresh soil (100 g) was placed in a sterile pre-weighed jar, allowed to dry in electric oven at 105 °C for 1 hour. After complete drying, cooled at room temperature, and difference in the weight was calculated in percentage.

Organic matter percentage

An air dried (100 g) soil was ashed at 600 °C in an electric furnace. Organic matter was measured by calculating the difference in the weights of the samples before and after ashing.

Soil reaction and electrical conductivity

The pH value and electrical conductivity (μScm^{-1}) were determined in 1: 5 soil water suspension ratio using a Jenway Water Analyser (Model PW 1), fitted with a combined glass electrode.

Total soluble salts (TSS)

For the estimation of total soluble salts, 20 g of air-dried soil was suspended in 100 ml distilled water and vortexed for 30 min. The mixture was left overnight to settle. Soil extract was then filtered and the obtained filtrate was evaporated in an oven at 105 °C until total dryness. The dry residue was then weighed and the TSS per gram oven dry soil was calculated. Table 3 shows the average values of three soil replicates.

Data analysis

To quantify the abundance and diversity of *Fusarium* species in the rhizosphere soil of date palm in an arid environment of Bahrain, data was analyzed according to statistical methods as outlined by Mandeel *et al*. (1995) and Mandeel (2002). The abundance of species (n) is expressed as the number of individual occurrences of species. Relative abundance was, however, used to determine species occurrence among all *Fusarium* species isolated from roots, plant debris, and soil. Relative abundance is defined as the number of isolates of a given species divided by the total number of fungal isolates. In order to determine the most dominant species, based on their relative abundance (%), species were arranged in descending order according to their abundance values. To measure the recovery

Table 3. Chemical, physical and ecological characteristics of date palm (*Phoenix-dactylifera*) soil collected from various villages.

*Location Code**	*Cond (μScm^{-1})*	*Soil moisture (%)*	*TSS** (%)*	*pH*	*Organic matter (%)*	*Temp °C (Air – Soil)*	*Relative humidity (%)*
ABH	4306	21.3	3.7	6.6	0.520	21 – 17	85
AIR	3800	05.6	2.2	7.6	0.188	20 – 18	75
AWI	2466	13.3	3.70	7.0	0.319	21 – 19	70
BAR	3666	11.3	3.7	7.0	0.505	20 – 20	70
BUD	3433	07.0	3.1	7.6	0.331	21 – 17	75
BUI	1400	09.3	3.38	7.1	0.521	21 – 19	85
BUS	7900	08.0	3.1	7.6	0.472	20 – 18	75
DMT	4700	10.3	2.47	7.2	0.322	21 – 18	85
HAM	3900	17.0	1.38	7.1	1.335	23 – 19	85
HID	6800	09.0	2.7	6.9	0.394	20 – 18	75
HIW	5166	06.0	4.45	6.93	0.499	21 – 19	85
JAS	7366	13.6	9.8	6.6	0.466	21 – 17	85
JDF	9600	26.0	6.67	6.07	0.801	21 – 19	85
JID	1500	30.3	5.4	7.1	0.801	21 – 19	75
JNU	4466	13.3	3.2	6.7	0.663	20 – 20	70
MAK	7706	11.0	3.8	6.9	0.426	20 – 20	70
KAR	7933	19.0	3.6	6.8	0.702	21 – 18	85
KAZ	11,266	11.3	3.7	7.3	1.281	21 – 18	85
NBI	11,100	14.3	5.65	7.2	0.956	21 – 19	80
SAD	6766	07.3	4.36	6.7	1.370	21 – 19	70
SAQ	4200	19.0	4.2	7.3	0.188	20 – 19	74
SAK	2466	07.0	3.2	7.2	0.301	21 – 19	70
SAR	4033	12.6	3.5	7.0	0.381	20 – 17	70
SNA	4300	16.6	4.95	6.70	0.939	21 – 19	85
SIT	14,833	12.6	4.53	6.6	0.863	21 – 19	80
TUB	3633	19.3	1.49	7.2	0.844	20 – 18	75
ZAQ	4466	13.6	3.91	6.8	0.503	21 – 19	70

* = Location codes (Arranged alphabetically).
BUS = Busaiteen, HID, AIR = Airport areas, SAQ = Saqayia, HAM = Hamlah, TUB = Tubli
JID = Jid Ali, BUI = Buri, JAS = Jasra, ABH = Alburhama, DMT = Dumistan, KAZ = Karzkan,
KAR = Karanah, JNU = Janusan, MAK = Malkyia, BAR = Barbar, SARR = Saar,
BUD = Budayia SAD = Saddad, ZAQ = Zallaq, AWI = Awali, SAK = Sakhir,
NBI = Nabi Saleh, HIW = Highway areas, SNA = Snabis and JDF = Jidahfs.

of the isolate, total number of isolate were computed in roots, debris, and soil among all locations.

The independence of recovery of *Fusarium* sp. by roots, plant debris, and soil was tested using Chi-square contingency table analyses (Steel & Torrie 1980).

To measure the species diversity within each plant height, roots, plant debris, and soil, Simpson's Index of diversity was used (Miller 1995). Diversity was calculated as follows:

Diversity = 1- Simpson's Index

$$\sum_{\mathbf{i}}^{\mathrm{s}} = 1\ P_{\mathrm{i}}^{\mathrm{s}}$$

where Pi is relative density of each species and s is the total number of species recovered. The index values span from 1/s to 1. If every isolate was a different species, the index would be equal to 1/s or small, indicating maximum diversity. If all isolates were of the same species, the index would be equal to 1, indicating minimum diversity.

Shannon and Weiner index is also one of a most widely used biodiversity indices for biotic community (Miller 1995). It describes the average degree of uncertainty by predicting species of an organism picked at random. The calculations were made as follows.

$$H` = -[(Pi)\ (In\ Pi)]$$

where H`= Amount of diversity in ecosystem, Pi = Represents proportion or relative abundance of each individual species measured to total, In Pi = Natural logarithm.

Similarity in species composition among the samples were estimated according to Sørensen's (1948) community coefficient (CC):

$$CC = 2a / b + c$$

where a, is the number of common species in the two samples compared, b is the number of single species in the first plant height and c is the number of single species in the second plant height.

To compare the richness of species using different isolation methods within plant communities based on different sample sizes, Sander (1968) proposed the rarefaction method for achieving the better comparison of such communities. Rarefaction is a statistical method for estimating the expected number of species, in a random sample of individuals (n), taken from a population of N total individuals distributed among S species. Calculations were made using rarefaction calculator statistical program, based on the formula of Hurulbert (1971).

$$E\ (Sn) = \sum_{i=1}^{s} = \{1-[\ \tfrac{N\text{-}n\ i}{n\ n})\ /\ \underline{N})]\}$$

where n, is the number of the individual of the species *i,* S is the total number of species.

RESULTS

Habitat description and soil analyses

A summary of various ecological parameters, vegetation pattern, coordinate position, and plant characters, from 27 villages of Bahrain are listed in Table 1. Villages planted extensively with trees of date palms were properly

surveyed, but due to rapid urbanization, oil discovery and lack of continuous supply of fresh water resources, certain locations, cultivars or even entire farms become neglected. Date palm grown in Sitra and Nabi Saleh villages clearly show symptoms of sulphur pollution from the industrial effluents like soil salinity, in the form of superficial salt crust are also observed in some locations, Airport areas and Highways. Villages like, Barbar, Sarr, Budayia, and Zallaq did not show coordinate position, while all other areas are located within 26°(N) and 50°(E). The absence of these locations is may be due to some unknown navigational or military reasons.

Cultivated areas are those which are properly managed by man-made cultivation of date palm while non-cultivated, were those where date palms are growing naturally. No attempt was made to identify the cultivars or to determine the yield, size or plant age. Traditionally, date palm is not cultivated exclusively alone, but interplanted with fruit trees such as papaya, banana, mango, lemon. Vegetables such as tomato, carrot, water melons, egg plant, lettuce, cabbage, turnip, onion, spinach, radish, parsley… etc and forage crops such as alfalfa are grown in addition to all these plants. Practically, no special fertilization is given to trees in intercropped cultivation. Locally, high quality cultivars are propagated by suckers and are planted randomly at about 5-6 meters apart. In nearby villages and farm, this had resulted in dense, moderate or sparse vegetation cover, which reflect farming activity.

All these factors were taken into consideration during the present survey of the date palm soils. None of the sample was from farm or location treated with *Fusarium* sensitive fungicides.

The sampling transect established for date palm soil collection is located towards the western through northern part of the main land in the physiographic zone of black slope (Fig. 1). The vegetation cover is dense, and is mainly dominated by halophytic flora.

Table 3 shows some edaphic features of the study sites for the 27 villages. The results obtained from the analyses of rhizosphere soils of several soil types of date palm proved to vary considerably without any consistent pattern. Most of the sampled soils were found calcareous, low to moderately saline, variable in texture from loamy to sandy loam, well-drained with low profile and water retention capacity. Soil moisture contents of the soil in present work fluctuated greatly among the villages, ranging from as high as 30.3% in Jid Ali to as low as 5.6% in Airport areas and 6.0% in Highway areas. Soils of other villages (Alburhama 21.3%, Tubli 19.3%, Karanah 19.0%, Hamlah 17.0% and Snabis 16.6%) were considered to be moderately moist. However, since sampling was carried on different dates, and because soil moisture is a variable factor, no undue importance should be given to these differences unless they are substantiated by further periodic and comparable determinations. Total water soluble salts (TSS%) were mostly found to be slightly high, a common feature of cultivated soils during non-rainy periods minimum average value for TSS were reported in soils of Hamlah (1.38%) and Tubli (1.49%), where as the maximum average were encountered in soils of Jasra (9.8%) followed by Jidahfs (6.67%). Sodium and chlorides are the most dominant water soluble salts present in date palm soil. Such increase may be due to repeated irrigation of nearby plants with relatively saline underground water. Soil reaction values (pH) in the present study were slightly acidic to neutral with an average of 6.9 without significant difference between the villages. The highest pH value (7.6) was recorded in soils of Busaiteen, Airport areas, and Budayia and the lowest pH (6.0) was noted in soil of Jidhafs.

Data for salinity expressed in terms electrically conductivity (μScm^{-1}) revealed that most soils are moderately to highly saline with rare exceptions. The maximum conductivity level was observed in soils of Sitra (14,833 μScm^{-1}), followed by Karzkan (11,266 μScm^{-1}) and Nabi Saleh (11,100 μScm^{-1}), whereas the minimum conductivity values were reported in the soils of Buri and Jid Ali in order of 1400 and 1500 μScm^{-1}, respectively. Electrical conductivity in other soil samples as Jidhafs (9600 μScm^{-1}), Karanah (7933 μScm^{-1}) and Busaiteen (7900 μScm^{-1}) were also considered to be high. Organic matter contents of all the samples were generally low due to poor or no fertilization programme. Organic matter contents varied from 0.188% in soils of and Airport areas where as the organic matter was found to be (1.335%), (1.370%) and (1.281%) in soils Hamalah Saddad and Karzkan, respectively. Where data for atmospheric and soil temperature are considered, it can be observed that the former value ranged from 20 to 23 °C while the later value range from 17 to 19 °C. The average relative humidity fluctuate between a minimum 70% to maximum 85%, indicating a humid atmosphere.

Fusarium species recovery and distribution

Occurrence of *Fusarium* species from the soils of 27 villages represented by small, medium, and large date palm trees using different isolation media and methods are presented in Table 4. A total of 13 species were isolated from the designated northern-west transect. The species listed according to the sequence of sections according to Brayford (1993) and Nelson *et al*. (1983), were *F. avenaceum* (FR) Sacc. R.J.Cook (Section Martiella) *F. chlamydosporum* Wollen & Reinking (section Arthrosporiella), *F. equiseti* Corda (section Gibossum), *F. illudens* C. Booth (section Martiella), *F. lateritium* Nees (section Lateritium), *F. moniliforme* var. *subglutinans* Wollen & reinking (section Liseola), *F. oxysporum* Schlecht. (section Elegans), *F. pallidoroseum* Cook (section Arthrosporiella), *F. poae* Perk (section Sporotrichiella), *F. sambucinum* (Section Discolor), *F. solani* Martius (section Martiella), *F. sporotrichioides* Sherb (section Sporotrichella) and *F. tricinctum* (Sporotrichiella).

Collectively, the highest species recovery was recorded from root samples (12 species) and the lowest from plant debris and soil samples (6 species each) using the direct plate method onto selective media. Data in Table 4 revealed unclear pattern of *Fusarium* occurrences among the different plant heights. A sum of 2107 *Fusarium* isolates, fluctuating from 1 to 345 per location and media, were encountered during the present study, among all isolation methods, media, plant heights and locations. The average occurrence of fungal isolates (702.3) was much higher in samples of plant debris (916 isolates), than from root samples (684 isolates) or soil samples (507 isolates). Moreover, Komada selective media yielded higher isolate recovery as compared to Selective *Fusarium* agar media, but not necessarily species diversity. The two most common species were *F. solani* and *F. oxysporum*. The remaining (11) species composed only 6.78% of the total *Fusarium* species isolated.

Comparatively, root samples of small, medium and large plants yielded a higher average species recovery (8 species) than plant debris (5 species) and soil (3.33 species). Soil of medium and large plant resulted in lowest species recovery, being 2 and 3 species, respectively. A cumulative of 276 isolates, ranging from 17 to 50 per plant height was obtained from all isolation type. The occurrence of *Fusarium* isolates, (average 92) were much higher in the root samples (aver-

Table 4. Mean recovery of Fusarium species within roots, plant debris, soil from small, medium and large date palm (*Phoenix dactylifera*).

Fusarium spp*	*Isolation types*											
	Roots			*P. debris*			*Soil*			*Abundance*	*Mean Frequency*	*Relative density*
	**S	M	L	S	M	L	S	M	L	(%)	(%)	(%)
F. solani	7.05	7.2	9	11.89	7.6	9.08	7.68	14.45	15.87	89.82	100	32.60
F. oxysporum	10.6	9	7	5.42	14.2	10	7.87		4	68.09	88.88	24.63
F. avenaceum	3		2.5		4	1	3			13.50	55.55	05.07
F. equiseti	8	1								09.00	22.22	03.26
F. sporotrichioides	4									04.00	11.11	01.44
F. chlamydosporum	3.5	2	2	2	2	2				13.50	66.66	05.07
F. pallidoroseum	8	1					1			10.00	33.33	03.62
F. lateritum	2	7	12			14				35.00	44.44	12.68
F. sambucinum	1									01.00	11.11	00.36
F. illudense	2									02.00	22.22	00.72
F. moniliforme		3	4	4.5	4	1			3	19.50	66.66	07.24
F. poae		1								01.00	11.11	00.36
F. tricinctum								5	3	08.00	22.22	02.89
Total No. of species	10	7	7	4	5	6	5	2	3			
Total No. of isolates	50	30	38	24	32	37	25	17	23	276.0		100.0

* = Species are arranged in descending order.

** = S = Small M = Medium L = Large

age 118), followed by debris (average 93) and lowest in soil (average 65). Likewise, the lowest isolation occurrences were observed in soil of medium (17 isolates) and large (23 isolates) plant heights.

A gradual decrease in *Fusarium* spectra and richness is noted only in roots and soil in relation to increase in plant height from small to large. The largest average abundance was recorded in roots, digressively arranged in order of magnitude; 50, 30 and 38 for small, medium and large plants, in that order. Similarly, in soil a decrease in total fungal recovery was encountered from large (23 isolates), medium (17 isolates) to small plant height (25 isolates). Species richness in roots and soil showed somewhat similar trends. However, plant debris revealed a gradual increase in *Fusarium* colony counts coupled with an increase in the plant heights from small to large resulting in 24, 32, and 37 isolates, respectively. The highest species occurrence was reported in plant debris of large plants (06 species) whereas, the lowest was noted in debris of small plants (04 species). The number of species recovered during the present study per plant height, fluctuate between 2 to 10 (Table 4).

The most common *Fusarium* species that was reported with 5% or more relative density, among all isolation types and plant heights, were *F. solani* (32.60%) and *F. oxysporum* (24.63%). The relative density of other *Fusarium* species varied from 0.36% to 12.68%, for *F. poae* and *F. lateritium*, respectively. The highest frequency of occurrence was displayed by *F. solani* (100%), followed by *F. oxysporum* (88.88%), *F. chlamydosporum* and *F. moniliforme* (66.66%). The frequency of occurrence of remaining species ranged from 11.11% to 55.55%. Some *Fusarium* species showed broad-spectrum recovery while others were restricted to specific isolation type. For example *F. sporotrichioides* was associated only with roots whereas *F. tricinctum* was from soil.

Table 5 lists specific data on the recovery of *Fusarium* isolates from roots, plant debris and soil of various plant heights at each location, regardless of species encountered on particular isolation media. All sites, except Airport area, contain *Fusarium* colonies but the composition of population in soil, plant debris, and roots as well as within each plant height differed substantially. Variations related to location were much greater than difference attributed to isolation type or plant height. A maximum of 1673 isolates with an average of 163 occurrences per plant height were encountered from all locations. The occurrence of *Fusarium* isolates was much greater in plant debris (723 isolates), followed by roots (540) and lowest in soil samples (410 isolate). Fungal occurrences in plant debris revealed a maximum recovery of 291 isolates in small and 231 isolates in medium plants, whereas large plants were lower than both (201 isolates). Isolate composition in soil samples were not similar to plant debris in being high in medium plants (162 isolates), but low in small plants (123 isolates). *Fusarium* occurrences within root samples were observed proportionally related to plant height from small to large, thus differed from that found within plant debris and soil sample. The highest average abundance was recorded in roots, progressively arranged in the order of 163, 160, 217 isolates in small, medium and large plants, respectively. Plant debris of medium plant yield higher occurrences from Karanah village (36) followed by 34 isolate from Buri village. Locations in Table 5 are arranged in descending order according to their frequency of occurrence showed that some sites namely Hamlah, Jid Ali, and Karanah exhibited absolute occurrence of *Fusarium* isolates. The frequency of occurrence of other locations ranged from 11.11 to 88.88%.

To examine the effect of geographical distribution trends on *Fusarium* occurrence in date palm grown in Bahrain, location were divided into north and west transects (Table 6). In general, northern locations yielded higher species (12)

Table 5. Mean recovery of *Fusarium* isolates within roots, plant debris, and soil from small medium and large date palm (*Phoenix dactylifera*).

Location	*Isolation types*											
*Code**	*Roots*			*P. debris*			*Soil*			*Abundance (%)*	*Mean Frequency (%)*	*Relative density (%)*
	**S	M	L	S	M	L	S	M	L			
JID	17	23	29	13	10	8	12.5	25	25	162.5	100	9.71
JAS	8	5	6	27	25	45		12		128	77.77	7.65
HAM	10	10.5	6.66	38	8.33	5.8	8.5	24	9	120.79	100	7.21
SAR	4.5	1	4	16	16	18		35	18	112.5	88.88	6.72
KAR	8.0	20	1	20	36	10	3	4	4.5	106.5	100	6.36
ABH	11	14	3.5	12.6	15.5	16	6.6	15	2.5	97	100	5.79
KAZ	4.33	5.5	22	13.6		44			4	93.43	66.66	5.58
BUI	9	17	17		34		8	4	4	93	77.77	5.55
SNA	17	4.5	18	7	9			14	20	89.5	77.77	5.34
BAR	5	5		35		7	15	7	10	84	77.77	5.02
BUS	5	5		2	9	4	42		5	72	77.77	4.30
DMT	7.5	8	26	6	7	17				71.5	66.66	4.27
JNU	15	5	15	5	1	1	17	3		62	88.88	3.70
TUB	2.6	19	32		2.5				3	59.1	55.55	3.52
AWI	7		7	21	15	9				59	55.55	3.52
SAQ		1		10	16	9	8.5	3	9.5	57	77.77	3.40
MAK	14	5		13	3.5	6		5		47	66.66	2.80
HIW	2	2	14		16					34	44.44	2.03
HID				17				10		27	22.22	1.61
SAK	7	4	4						10	25	44.44	1.49
AIR				24						24	11.11	1.43
ZAQ	9	5.5	2.5	3		1				21	55.55	1.25
BUD				4	4					8	22.22	0.47
JDF				4	3					07	22.22	0.41
NBI			5							05	11.11	0.29
SAD			4							4	11.11	0.23
SIT							2	1		3	22.22	0.17
Total No. of isolates	163	160	217	291	231	201	123	162	125	1673		100

* Location codes (Refer to table 3). ** = S = Small, M = Medium, L = Large. *** = Relative density.

Table 6. Geographical distribution (± mean recovery) of *Fusarium* species within roots, plant debris, and soil from small, medium, and large date palm (*Phoenix dactylifera*), in north and west transects. (West transects are within paranthesis).

*Fusarium spp**	*Isolation types*											
	Roots			*P. debris*			*Soil*			*Abundance (%)*	*Frequency (%)*	*Relative density (%)*
	***S*	*M*	*L*	*S*	*M*	*L*	*S*	*M*	*L*			
F. solani	9.6 (6.6)	10 (8.8)	10.11 (17.25)	21.64 (14)	13.54 (17)	14.36 (7.28)	12.77 (8)	15 (4.5)	13.62 (9)	120.64 (92.43)	100 (100)	27.93 (21.39)
F. oxysporum	9.75 (7)	13	9.5 (10.33)	(6)	3.5 (3.66)	9.83 (8)	2	12.6	5	65.18 (34.99)	88.88 (55.55)	15.08 (8.04)
F. avenaceum	(3)		8 (2)		4	1				13 (05)	33.33 (22.22)	3.00 (1.16)
F. equiseti	8									08 –	11.11 –	1.85 –
F. sporotrichioides	(4)									(04)	(11.11)	(0.92)
F. chlamydosporum	7	2	2							11 –	16.66 –	2.54 –
F. pallidoroseum	7 (1)	1					1			09 (01)	33.33 (11.11)	2.08 (0.23)
F. lateritium	2	7	12			14				35 –	44.44 –	8.10 –
F. sambucinum	1									01 –	11.11 –	0.23 –
F. illudens	2									02 –	11.11 –	0.46 –
F. moniliforme			4 (3)	6 (3)	2	3				15 (06) –	44.44 (22.22) –	3.48 (1.38) –
F. poae	1									01 –	11.11 –	0.23 –
F. tricinctum					3	5				08	22.22	1.85
Total No. of Species	8 (5)	5 (1)	7 (4)	3 (3)	4 (2)	4 (2)	3 (1)	2 (1)	4 (1)			
Total No.of isolates	46.35 (21.66)	33 (8.8)	46.61 (32.58)	31.14 (23)	29.37 (20.66)	31.37 (15.28)	26.37 (8)	18 (4.5)	26.62 (9)	288.52 (143.48)		100

* = *Fusarium* species are arranged in descending order according to their abundance.

*** = S = Small plant M = Medium plant L = Large plant

than western sites (6). Moreover, the highest species recovery was noted from roots of small plants (8 species) of north locations compared to all other isolation types or plant heights. Quantitatively, the highest average number of isolates was recovered from north (288.82) as compared to isolates from west location (143.48). In addition, root sample of various plant heights yielded higher average isolate recovery (125.96) than all other samples, isolation types and plant heights.

Soil samples of different plant heights at western transect resulted in one species only i.e. *F. solani.* While all other species were found differently in roots and plant debris of small, medium and large plants. All other species found in north locations also were found in west location but not vice versa. *F. equiseti*, *F. chlamydosporum*, *F. sambucinum*, *F. illudense* and *F. poae* were present in different plant heights of north location only and mostly from roots and plant debris samples. *F. sporotrichoides,* however, was only found in west locations associated with roots of small plants comprising 11.11% frequency of occurrence and relative density value of 0.92%. *F. moniliforme* was found in roots of large plant and plant debris of small and medium plans in north, while it is also found in roots and plant debris of large and small plants of west locations with total occurrence of frequency of 44.44%. *F. pallidoroseum* was present in roots and plant debris of small and medium plants in north as well as in small plants in west with a total frequency of occurrence 33.33%. *F. solani,* on the other hand, exhibited an absolute occurrence frequency, whereas *F. oxysporum* was present among all isolation types from sites of the north location but not in west in samples and roots soil of medium plants. In soil samples, *F. avenaceum* was found in the west but absent in north with a total of 33.33% occurrence frequency. In root samples, *F. lateritum* was found in north transects, present in the roots of small, medium, and large plants as well as plant debris of large plants only. However it is absent in the west locations. The frequency of occurrence of other *Fusarium* species ranged between 11.11% to 55.55% (Table 6).

To examine the overall effect of soil edaphic factors on the lack of *Fusarium* species recovery from roots in different locations, data in Table 7 were evaluated. All four locations i.e., Hid**,** Airport areas, Budayia and Saddad showed more or less similar level of soil components, except for Jidhafs. The salient features of this location were high soil moisture contents (26%), TSS (6.67%) and also somewhat high salinity value (9600 μScm^{-1}). Factors for other samples were generally low and varied for soil moisture, TSS, salinity and organic matter from 5.6 to 9.0%, 4.36 to 7.0%, 3433 to 6800 μScm^{-1} and 1.37 to 0.188%, in that order. The pH value remains almost neutral ranging from 6.7-7.6. It might be speculated that these sampling sites were poor in overall vegetation cover and soil types of these locations lacking the supporting nutrients for the growth of *Fusarium* species. It might also be due to poor irrigation methods and fertilization patterns. Despite the fact that the above locations showed no *Fusarium* recovery, however, sites of some similar characters yielded *Fusarium* recovery from plant debris and soil. Moreover, because of relatively small samples size, it is difficult to attribute values given in Table 7 for the lack of *Fusarium* recovery to specific soil factors. Two locations did not indicate any isolation of *Fusarium* from plant debris The analyses of soil chemical factors revealed that Nabi Saleh exhibited high moisture contents (14.3%), TSS (5.65%) and conductivity (11, 100 μScm^{-1}) but endeavour low organic matter contents (0.596%). Saddad revealed less values of same parameters. There are eight soil samples from the rhizosphere of date palm from which no *Fusarium* species were isolated (Table 7). A comparative analysis of soils of these locations indicate that all soils were also poor in soil moisture, except Tubli featured by high soil moisture contents (17.60%) and lowest in Highway

Table 7. Locations indicating no Fusarium isolates from roots, plant debris, and soil samples of date palm (*Phoenix dactylifera*) and their correspondent soil analyses.

Isolation Type	*Location code**	*TSS** (%)*	*Soil moisture (%)*	*pH*	*cond (μScm^{-1})*	*Organic (%)*
Roots	HID	2.77	9.0	7.6	6,800	0.394
	AIR	2.20	5.6	7.6	3,800	0.188
	JDF	6.67	26	6.7	9,600	0.801
Roots/ plant debris	BUD	3.10	7.0	7.7	3,433	0.331
Roots/ plant Debris / soil	SAD	4.36	7.3	6.7	6,766	1.370
Plant debris & soil	NBI	5.0	14.3	6.7	9,600	0.801
Soil	TUB	1.49	19.3	7.1	11,000	0.955
	ZAQ	3.19	13.6	6.8	4,466	0.503

* = Location codes (Refer to table 3). ** = TSS (Total soluble salts).

areas (6.0%) location. TSS in Tubli was the highest (5.65%) and lowest in Nabi Saleh (1.49%). Electrical conductivity was high in Nabi Saleh (11,100 μScm^{-1}) and lowest in Awali (2466 μScm^{1}). In general, all samples from these locations showed high conductivity. Organic matter content was found to be high only in Saddad (1.37%), while others were below one. pH was neutral ranging from 6.8 to 7.7 and does not indicate any limiting effect.

Table 8 lists some of the ecological analyses of *Fusarium* species and isolates distribution from small, medium and large date palm roots, plant debris and soil samples. The highest pattern of mean recovery of *Fusarium* isolates was noted in samples of plant debris of small plants 394, followed by similar samples of medium plants (285). Roots of large plant heights yielded 259 isolates where as the soil resulted in lowest isolate recovery (150). Species richness fluctuated between 2 to 10 for soil samples of medium plants and root samples of small plants, respectively with a total average of 5.5 species. Diversity indices were computed with Simpson index and Shannon and Wiener index. For the combined plant height as shown by Simpson's index, isolation from root samples consistently gave the highest diversity index (0.533) and that isolates from plant debris averaged (0.300). In small plant height, samples of roots and soil resulted in maximum diversity indices of (0.588) and (0.500), in that order. In medium plant heights, soil samples yielded minimum species diversity (0.073). Larger plant heights, however, yield maximum species diversity within root samples (0.544) and isolates from plant debris had the lowermost diversity index (0.218). Species diversity as indicated by Shannon & Weiner indices were found maximum (451.23) in soils of medium plants followed by larger plants (411.67) and plant debris (405.0) of similar plant heights. The lowest diversity was noted in roots of larger plants (356.69) (Table 15). Collectively, the maximum species diversity was found in soils of larger plants (401.42), followed by samples of plant debris (397.0) and roots (351.59).

In order to examine pair wise comparison to determine similarity among *Fusarium* species between the various plant heights of roots, plant debris, and soil samples, Sørenson's coefficient of similarity was used (Table 9). Altogether, with the exception of soil samples, the highest species similarity was observed in large-

Table 8. Some ecological analyses of *Fusarium* occurrence in roots, plant debris, and soil samples of small, medium, and large date palm (*phoenix dactylifera*).

Prameters	*Plant height*	*Sample types*		
		Roots	*Plant debris*	*Soil*
Mean recovery	Small	215	394	195
	Medium	210	285	162
	Large	259	237	150
	** Total	684	916	507
*Species richness	Small	10	04	05
	Medium	07	05	02
	Large	07	06	03
	Total	12	06	06
Species diversity (Simpson index)	Small	0.588	0.224	0.500
	Medium	0.469	0.447	0.073
	Large	0.544	0.218	0.267
	Total	0.533	0.300	0.322
Species diversity (Shannon & Weiner) index	Small	334.61	431.87	377.00
	Medium	373.82	380.67	451.23
	Large	356.69	405.00	411.67
	Total	351.59	397.00	401.42
Equitability (E)	Small	0.058	0.056	0.01
	Medium	0.467	0.089	0.036
	Large	0.077	0.036	0.089
	Total	0.044	0.050	0.053

* = Expressed as number of species recovered from each plant height.
** = Totals are calculated from average data of three plant heights.

Table 9. Species similarity (community coefficient) among various plant heights of roots, plant debris, and soil sample of date palm (*Phoenix dactylifera*).

Isolation Type	*Plant* Height*	*Paired sample*	*Common species*	*Community coefficient (CC)*	*PS (%)*	*PD (%)*
Root	S – M	10 – 7	5	0.58	58	42
	S – L	10 – 7	6	0.70	70	30
	L – M	7 – 7	6	0.85	85	15
Plant debris	S – M	4 – 5	4	0.88	88	12
	S – L	4 – 6	4	0.80	80	20
	L – M	6 – 5	5	0.90	90	10
Soil	S – M	5 – 2	1	0.14	14	86
	S – L	5 – 3	2	0.50	50	50
	L – M	3 – 2	1	0.2	20	80
Total	S –L	11 – 8	6	0.63	63	37
	S –L	11 – 7	3	0.33	33	67
	L – M	7 – 8	6	0.80	80	20

* = Plant height S = small M = medium L = Large
** = CC = Sørenson Co-efficient of similarity.
PS = Percent of similarity.
PD = Percent of dissimilarity.

medium plant combination (0.8) and the lowermost were in pairs of small-large plants (0.33). The maximum community coefficient index was noted in large-medium plant heights (0.90), comparisons in plant debris samples followed by roots of similar plant height (0.85). Plant debris samples of small-medium as well as small-large plants also exhibited high species similarity composition, (0.88) and (0.8), respectively. Community coefficient index revealed that for large-medium plant heights comparison of soil are, distantly related (0.2).

To estimate the highest expected number of species isolated from a theoretical random standardized sample (n), rarefaction indices were computed and curves were constructed (Fig. 2). The highest rarefaction index from a subsample of a standardized size, randomly chosen from 250 occurrences of each sample among all plant heights indicated that root samples were richest in species recovery and soil was the lower most. Root samples of small plant height yielded the highest expected number of species (9) from 140 occurrences, followed by medium and large plants (6) (Fig. 2 A). Plant debris of large plant, (Fig. 2 B) resulted in lower expected number of species than small and medium plants in order of 4.5 and 3.5 respectively. In soil, the highest expected number of species in a random sample of 100 occurrences, were inversely correlated with plant height from small to large in that order (Fig. 2 C).

Fusaria surveyed from various plant heights of date palm plants were not recovered independent of isolation method. Analyses of interaction between species recovered and isolation type proved to be highly significant at $P = 0.05$, as determined by the chi-square contingency Table (10). A greater than expected recovery of *F. oxysporum* and *F. solani* occurred from roots, plant debris and soil samples of all plant heights. In root samples of large plant, recovery of *F. lateri-*

Table 10. Contingency table of *Fusarium* species by plant height interactions, data represents Chi-square statistics.

*Fusarium Species**	*Plant Height*								
	Roots			*P. debris*			*Soil*		
	***S*	*M*	*L*	*S*	*M*	*L*	*S*	*M*	*L*
F. avenaecum	3.14	3.07	3.78	3.87	2.80	2.32	1.51	0.95	0.88
F. moniliforme	2.20	0.92	2.65	6.02	1.24	3.62	1.15	0.95	0.88
F. oxysporum	55	53.7	66.26	51.58	37.02	37.78	31.92	26.52	24.55
F. solani	135.47	132.32	160	324	234.59	195.05	157.30	130.68	127
F. lateritium	6.60	2.14	7.95	6.02	4.35	3.62	–	–	–
F. chlamydosporum	2.20	0.61	4.16	0.86	0.62	0.51	–	–	–
F. equiseti	2.82	0.30	5.6	–	–	–	–	–	–
F. pallidroseum	2.82	0.30	0.56	–	–	–	0.38	0.31	0.29
F. poae	0.31	0.30	0.37	–	–	–	–	–	–
F. sambucinum		0.31	0.30	0.37	–	–	–	–	–
F. sporotrichioides	1.22	1.22	1.51	–	–	–	–	–	–
F. illudens		1.38	1.39	1.25					
F. tricinctum		–	–	–	–	–	0.38	0.31	0.29

* = Species are arranged according to degree of occurrence.

** S = Small M = Medium L = Large

tum was greater than expected as compared to other species. Similar significant chi-square values were obtained in the contingency table using relative density values for species by isolation type and plant heights interactions.

DISCUSSION

Fusarium species spectra and diversity

This report presents the findings of first attempt to examine the occurrence of *Fusarium* species in rhizosphere soil, plant debris and roots of date palm in arid environment of Bahrain. Most of these species representing different sections were isolated previously from desert (Abbas & Mandeel 1995; Mandeel *et al.* 1995; Mandeel 1996) or cultivated habitats (Mandeel & Abbas, 1994). Three species namely, *F. poae*, *F. illudens*, and *F. avenaceum* are newly documented from cultivated habitats and arid desert soils of Bahrain. Maximum colony counts among all plant heights and locations was recorded in plant debris (916 isolate), followed by roots (684 isolates) and soil (507 isolates) (Table 4). However, of the thirteen species isolated, twelve had been recovered from roots alone compared to six species only from either plant debris or soil. *Fusarium* species were found in relatively low densities in all soil samples and were regarded as common soil saprophytes. Comparison of soil *Fusarium* species of date palm in Bahrain with those of other parts of the world is rather difficult, since there are variations in isolation techniques, media, collection season, soil depth or a combination of any of these factors. Abou-Heilah (1985), reported that number of fungi per gram of soil vary within season and soil depth. In addition, disturbance of soil caused by cultivation or human activity, may also have an effect on species spectrum, types and frequency of soil fungi.

Of all the *Fusarium* species recovered, *F. solani* and *F. oxysporum* were the most dominant, occurring in nearly all plant heights, isolation methods and media, as well as from nearly all locations examined with *F. lateritium*, the next most prevalent species (Table 4). Although *F. moniliforme* and F. *avenaceum* and *F. chlamydosporum* occurred scantly over a wide area, they were not frequently isolated. *F. solani* and *F. oxysporum* are prominent members of mycoflora in alkaline desert soils (Abdel-Hafez *et al.* 1990; Khodair *et al.* 1991; Abbas & Mandeel 1995; El-Abyyad *et al.* 1993; Mandeel *et al.* 1995; Sangalang *et al.* 1995b; Mandeel 1996) and cultivated habitats (Nash & Snyder 1965; Kretuzer 1972; Kommedahl *et al.* 1979; Burgess *et al.* 1988a; Farias & Griffin, 1989; Klaasen *et al,* 1991). In similar arid environment, they were among the most prevailing species in dates (Moubasher *et al.* 1988), rhizosphere soil in Saudi Arabia (Bokhary & Parvez, 1995), rhizosphere soil of vegetable crops in Bahrain (Mandeel & Abbas, 1994), and rhizosphere soil of tomato in Egypt (Abdul-Wahid *et al.* 1997).

Fusarium solani was not the predominant species at every sample, however, it preclude in frequency (100%) and abundance (90%) of the total cultures and was isolated approximately in equal frequency from all the plants heights (Table 4). This species is a soil-borne pathogen of many crops in both temperate and in tropical regions where it is associated with root rots, seed decay and seedling blight of beans (Nash & Snyder 1961; Nash & Snyder 1965; Farias & Griffin 1989) wheat (Burgess *et al.* 1975; Klein *et al,* 1990; Klassen *et al.* 1991;

Fig. 2. Rarefaction curves for *Fusarium* species of small, medium, and large plants isolated from roots, plant debris, and soil samples of date palm (*Phoenix dactylifera*) plant. E(s) is the expected number of species in random theoretical samples of (n) population of isolates from each sample.

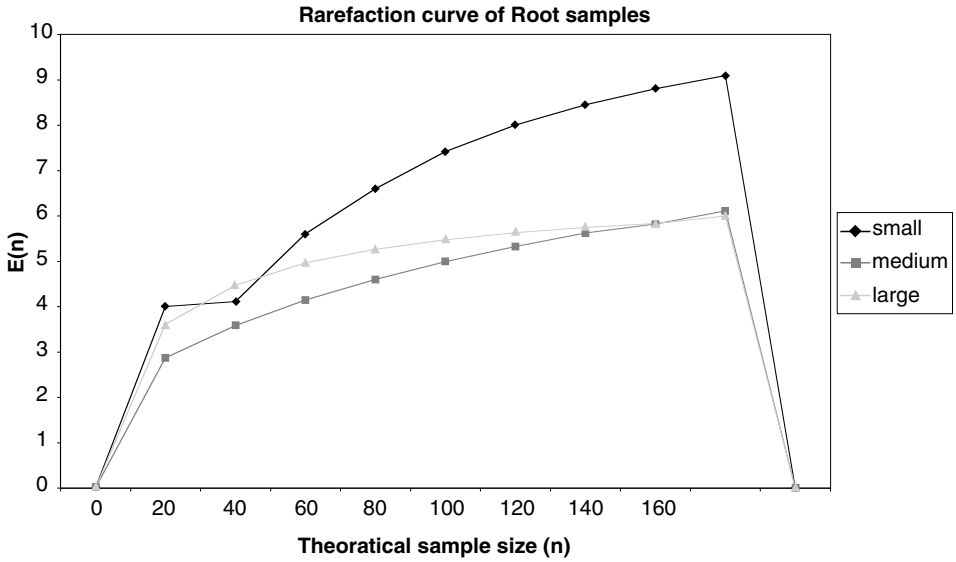

Figure 2a Rarefaction curve of root samples.

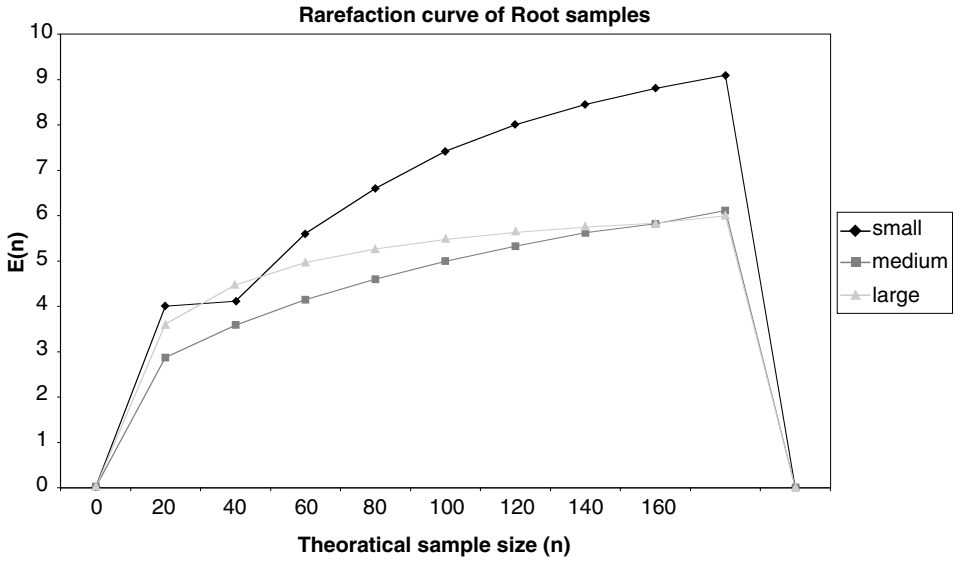

Figure 2b Rarefaction curve of Plant debris samples.

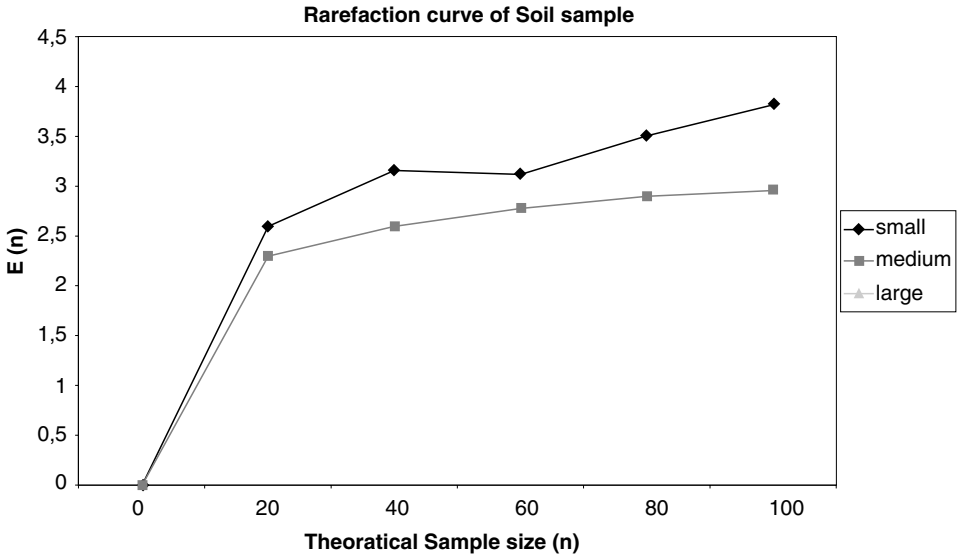

Figure 2c Rarefaction curve of soil samples.

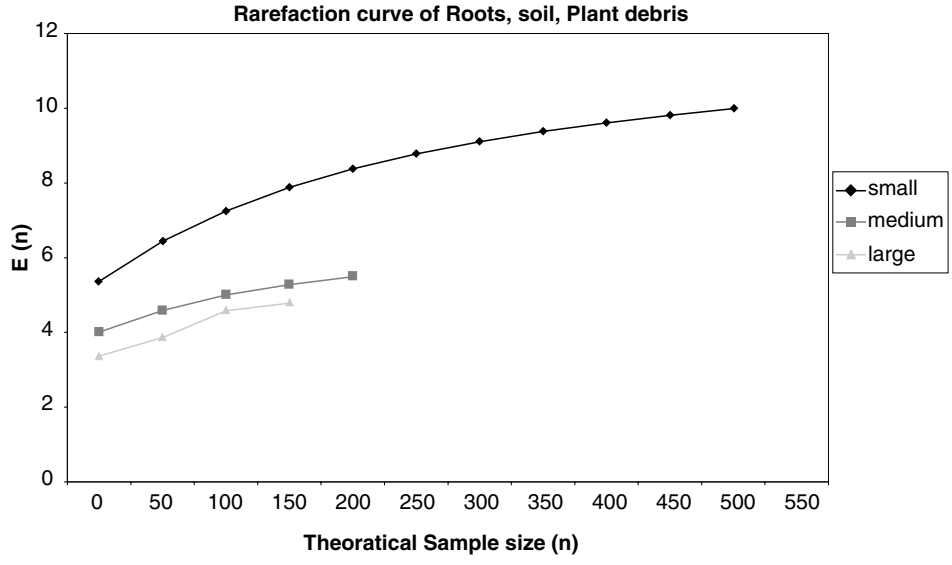

Figure 2d Rarefaction curve of combined samples of roots, plant debris, and soil.

Smiley & Patterson, 1996), corn (Windels & Kommedahl, 1974; Kommedahl, *et al.* 1979; Windels & Kommedahl, 1984; Gilbertson *et al.* 1985; Lesile *et al.* 1990) and other plants (Kommedahl *et al.* 1987; Kalc *et al.* 1997). It may also cause cankers in woody plants. In temperate regions the fungus is mainly soil-borne, but both soil and air borne in tropics (Burgess 1981). Saprophytically, the fungus survives as chlamydospore (Nash & Snyder, 1961; Cook & Bruell 1968; Kretuzer 1972; Cook & Rayner, 1984; Nelson *et al.* 1990). In warmer regions, *F. solani* appears to have an affinity for areas of high rainfall or irrigated soil (Wearing & Burgess 1979; Burgess 1981).

F. oxysporum was also common and accounted for 17.9% of all culture recovered. Like *F. solani*, it was not equally abundant at every sampling site, but attained the second highest frequency (88.88%) of the total species (Table 4). *F. oxysporum* was slightly more abundant in roots than in plant debris and soil. This result was expected since *F. oxysporum* not only produces abundant chlamydospore that can survive as dominant propagules in soil (Pettitt *et al.* 1996; Smith & Snyder, 1975), but also survives as active hyphae in plant debris (Burgess 1981). *F. oxysporum* is widely distributed in both temperate and tropical regions where it has been reported to be a pathogen of many economic crops (Farias & Griffin, 1989; Onyike & Nelson 1992; Abdul-Wahid *et al.* 1997; Kalc *et al.* 1997). It is one of the most common soil-borne *Fusarium* species isolated from roots and crowns of plants and it is active over a wide range of environmental conditions (Burgess 1981). Some forms of *F. oxysporum* are pathogenic causing vascular wilts, seed decay crown and root rots, while others are saprophytes on plant residues in soil. In North Africa, *F. oxysporum* formae species *albeidinis* are responsible for causing date palm wilts commonly known as Bayud disease (Brayford 1993). Pathogenic members of this species once established are very difficult to eliminate (Mandeel & Abbas, 1994).

The predominance of *F. solani* and *F. oxysporum*, in most sites at unequal abundance within specific sites and locations, suggest that these two species may be competing for same niche. *F. solani* was generally more abundant than *F. oxysporum*. The overall predominance of *F. solani* (75.65%) is similar to other findings from soils of Bahrain (Mandeel & Abbas 1994; Abbas & Mandeel 1995; Mandeel *et al.* 1995; Mandeel 1996), Saudi Arabia (Abou-Heilah 1985; Khodair *et al.* 1991; Boukhary & Parvez 1995) and Egypt (El-Abyad *et al.* 1990; Moubasher *et al.* 1985; Moubasher *et al.* 1988; Abdel-Hafez *et al.* 1990). These species clearly exhibits broad ecological tolerances (Sangalang *et al.* 1995a). Marasas *et al.* (1988) concluded that *F. oxysporum* was one of the most predominant species obtained and that this fungus occurred more frequently in cultivated soils.

F. lateritium was the third most prevalent species accounted for 1.66% of the total cultures and was recovered in almost equal frequency from the samples. This species is considered as an air born fungus surviving basically as an active saprophyte on decaying plant parts or soil. Pathogenic forms are well adapted to wet condition in moderate temperature causing twig blight and die back of ornamental trees (Burgess 1981). The fungus persists during the dry seasons as mycelium in plant debris in soil or plant surface but disseminated mainly by rain splashed. Mandeel *et al.* (1994, 1995) previously isolated the fungus from cultivated and non-cultivated habitats of Bahrain.

Other less frequently isolated species in the section Liseola (i.e. *F moniliforme* and *F. smitectum*) are usually associated with various plants as primary pathogens or secondary colonizers, especially in arid regions (Gerlach & Nirenberg 1982), with the rarest exceptions outside their suitable host. Soils from arid region were also commonly featured by the occurrence of *F. chlamydosporum*

and *F. equiseti*, which was supported by other findings of Burgess *et al*. (1988a), Burgess & Summerell (1992), Mandeel *et al.* (1995) and Mandeel (1996).

Diversity indices calculated for *Fusarium* species from roots, plant debris, and soil from various plant heights and among all locations, indicated that overall roots were a source of greater species diversity (0.533%) than either soil or plant debris (Table 8). Our results contradict those of McMullen & Stack (1983) and Jeschke *et al.* (1990) whom found that debris yielded a greater diversity of *Fusarium* species than soil. However the fact that species diversity is higher in roots than plant debris or soil might indicate that diversity may be a subjective measure and may depend on species isolated and mode of survival. Chlamydospore producers may be more readily recovered from soil, while species which survives more as mycelium, are more likely to be isolated from plant debris and roots. Our study has shown that *F. solani* was recovered at greater frequency from soil than plant debris (Table 4). The location with greatest diversity of species present was not the most diverse with respect to the vegetation present (Table 1). Moreover, although certain locations (i.e. Tubli, Dumistan, Saddad, Awali) were characterized by dense vegetation cover and apparently healthy plants coupled with fertilized, irrigated and cultivated soil, nonetheless, no *Fusarium* recovery was possible from such sites. Likewise, no recovery was made from noncultivated, nonfertilized and nonirrigated soils and, locations with sparse vegetation (i.e. Budayia, Zallaq, Nabi Saleh). These observations suggest that there was no apparent relationship of species spectra and the types of vegetation present at each site. A greater diversity of adjacent vegetation might be expected to support a greater diversity of *Fusarium* species, but the data did not provide any evidence to support this hypothesis.

Overall, the abundance and assemblage of *Fusarium* species recovered from cultivated areas were similar to that recorded from vegetable crops in Bahrain (Mandeel and Abbas, 1994) and cereal crops of central and eastern United States (Lesile *et al.* 1990), in that *F. solani* and *F. oxysporum*, were the most abundant species.

A comparison of fungal species of various isolation methods among pairs of plant heights showed a high similarity in their species composition especially between large and medium plant heights (total 0.8) (Table 9). This species similarity suggests a minimum variation among the communities that possibly is a reflection of somewhat similar abiotic and biotic characteristics of these habitats. Recovery from soil samples, however, revealed the most variable compared to roots and plant debris. These findings indicate that soil mycobiota are governed by a-biotic factors different from the other two (Cook & Rayner 1984; Burgess *et al.* 1988a; Jeschke *et al.* 1990; Klaasen *et al.* 1991).

Christensen (1988) proposed that the community is in crisis if an adapted biological community has a disproportionate number of certain species. The alpha diversity values (rarefaction curves) in the present study that was obtained in the samples of various plant heights and isolation types employed were low (Fig. 2). This is characteristic of this type of environment and moreover showed that probably the community of *Fusarium* species in the surveyed soil is stable. In order to determine the appropriate sample size, species are enumerated as a function of the number of occurrences in a random sample size and a curve is plotted of species occurrences. Evidently, an asymptote of the species occurrence is considered indicative of a sufficient sample size (Heck *et al.* 1975). In this study, the asymptote species occurrence curve of the sample of the roots followed by plant debris and soil from different plant heights rose to just slightly below the maximum number of occurrences, which indicate the appropriateness of the sample size.

Species diversity within different plant heights of various samples reveals that roots of small plants are quiet diverse than that of plant debris of large plants. However, soil samples of larger plants appear to be the least dissimilar (Table 8).

Influence of soil abiotic factors

High temperature, scanty rainfall and augmented levels of soil and underground water salinity are ever-recurring themes in Bahrain that have characterized the climate as typical Sahara-arid environment. Rainfall was below average (9.28 mm) (Table 2), in the sampling year from September, 1999 to March 2000 when soil samples from the west-north transect were collected. Drought conditions and high evaporation rates (8.21 mm) also existed during the same period. These conditions undoubtedly influenced the spectrum and diversity of *Fusarium* species recovered from roots, plant debris, and soil of date palm in Bahrain.

Burgess *et al.* (1988a) hypothesized that temperature is the most important climatic factor influencing the geographical distribution of *Fusarium* species. They concluded that temperature might exert a direct influence thereby affecting the fungus itself or indirectly by limiting the distribution of host to definite climatic zones (Burgess *et al.* 1988b; Backhouse & Burgess, 2002). Most *Fusarium* species exist in a wide geographical zone (Gordon 1956), while some species appear to be selectively restricted to specific climatic zones. (Burgess *et al.* 1988; Kommedahl *et al.* 1988; Jeschke *et al.* 1990; Summerell *et al.* 1993; Sangalang *et al.* 1995b). In this regard, *F. solani*, *F. compactum*, and *F. equiseti* are considered cosmopolitan in distribution, adapted to a wide range of environmental conditions and habitats, with particular preference to arid and semi arid regions. Burgess & Summerell (1992) found that *F. equiseti* is more common in plant debris from semi arid rangeland soils of eastern Australia than areas of high rainfall. In contrast, *F. sambucinum* and *F. sprotrichoides* have specific climatic requirements and are restricted more to cold temperature and alpine soils but are less common in tropics (Kommedahl *et al.* 1988). *F. oxysporum* also represents a species of broad occurrence, irrespective of climatic variations. This fungus was isolated from very cold climate of Iceland (Kommedahl *et al.* 1975), near arctic circle (Komedahl *et al.* 1988) tropical zone of southern Brazil (Kommedahl *et al.* 1987), subtropical areas of Nigeria and Zimbabwe (Onyike & Nelson, 1992) and arid habitats of Bahrain (Mandeel *et al.* 1995; Mandeel 1996).

While on a large geographical scale basis, species spectra and diversity could be related to variation in climatic conditions. Nevertheless on a narrow geographical scale, *Fusarium* distribution and recovery is mainly influenced by soil factors. Among the most important abiotic factors influencing microbial diversity and survival in particular soil niche, is the rhizosphere effect, salinity, organic matter content, soil moisture, temperature, nutrient balance and microbial interaction (Griffin 1972; Skujins 1984; Liddle & Burgess 1985; Christensen 1989; Nelson *et al.* 1990; Khodair *et al.* 1991; Mandeel 1996). In the current study, the highest isolate abundance (162.5%) and relative density value of 9.71% occurred in Jidhafs (Table 4), presumably due to low salinity levels (1500 μScm^{-1}) and high soil moisture (30.3%) as compared to other locations. Decrease in colony counts is inversely correlated with increase in salinity levels. It is obvious from the results that high moisture and organic matter contents favors species occurrence, whereas low moisture, organic matter with high total soluble salts, electrical conductivity have an opposite effects. Similar findings were reported elsewhere (Mandeel 1996; Abbas & Mandeel, 1995).

Data reveals inconsistent pattern for the influence of specific soil factors on fluctuation of *Fusarium* population and spectrum (Tables 1, 3, and 4). More

likely, an effect of multi abiotic factors such as high salinity (11,000 μScm^{-1}) and TSS (6.67%) coupled with poor organic matter content (0.188%) have resulted in absolute lack of *Fusarium* recovery from specific locations (Table 7). Furthermore, failure to isolate *Fusarium* species from Saddad location, in particular, using all isolation types, suggest that soil abiotic factors or plant influence and microbial interaction outcome are involved. Occurrence of 5 isolates of *F. solani* in Nabi Saleh locations in roots of large plants (Table 4 and 7) indicates that soil and plant debris are unfavorable media for growth and survival, despite the fact that *F. solani* is chlamydospore producer. Recovery of *Fusarium* species elevated (120.8 isolates) in Hamlah location characterized by moderate salinity levels, (3900 μScm^{-1}) low TSS (1.38%) as well as high organic matter contents (1.335%). Similar observations were also reported by Moubasher *et al.* (1985) for Wadi Bir-El-Ain in Egypt and by Mandeel *et al.* (1995), Mandeel (1996) in desert soils of Bahrain. Although certain species were not recovered by one isolation types, it appears difficult to assess whether recovery was completely favored, or inhibited by plant heights, locations or isolation types. Sangalang *et al,* (1995b) proposed that failure to recover an individual species from specific habitat does not imply the fungus is totally absent, even when present at low frequencies, given the nature of isolation technique used. This absence may reflect the environmental conditions at the time of sampling or may indicate the climate in the sampling locations was not favorable for growth of the species. Stoner (1981) examined soil born-fungi in fragile soil from the forest of Hawaii islands. He attributed the lack of *Fusarium* recovery to changes in plant communities and edaphic factors associated with ecological transition from open areas to closed forest and from rocky or sandy to muck soils. Therefore, for quantitative and qualitative comparison of soil *Fusaria* several isolation media (Mandeel, *et al,* 1994; McMullen & Stack 1983) and techniques (Mandeel, 1996) must be used to avoid any possible bias in the survey results. Variations in pH do not seem to affect species prevalence and diversity, except where possibly in the extremes.

Low occurrence levels and limited species variability of *Fusarium* species was observed in noncultivated locations as compared to cultivated ones (Table 1 and 4). This is in agreement with similar findings of others (Burgess 1981; Burgess & Summerell 1992; Summerell *et al.* 1993; Mandeel & Abbas, 1995), which have shown that number of isolates and species of *Fusarium* are greater in cultivated soils than in desert soils. Expectedly, cultivated soils have high organic matter content, better nutrient availability and low salinity and soluble salts. Windels & Kommedahl (1974) found that the recovery of *Fusarium* species from undisturbed, prairie soils differed significantly from the recovery from similar soils planted with maize. Similar finding were reported elsewhere (Nash & Snyder 1965; Abbas & Mandeel 1995).

Analyses of the relationships between species recovery and isolation types within different plant heights as determined by Chi-square contingency table revealed significant interactions (Table 10). The results suggest that some technique allow greater recovery of *Fusarium* species from plant heights. Unlike *F. oxysporum,* greater than expected recovery of *F. solani* occurred with isolation from plant debris followed by roots. Altogether, with exception of *F. solani* and *F. oxysporum* isolation from roots among all plant heights yielded higher colony counts and species spectra as compared to other methods.

Fusarium species are associated with both diseases (Lukezic & Kaiser 1966; Gilbertson *et al.* 1985; Farias & Griffin 1989; Klein *et al.* 1990; Smiley & Peterrson 1996; Kalc *et al.* 1997), and apparently health crop plants (Kommedahl *et al.* 1979; Thomas & Buddenhagen 1980). Data from this study are similar with

previous findings that *Fusarium* invasion and colonization is host systemic and that the species structure within host plant is somewhat different from that recovered from soil or plant debris. Whether certain species become either systemically distributed or confined to few cortical walls of the roots is largely based on host-fungus biochemical or genetic interactions (Cook & Bruell 1968; Cook & Rayner 1984; Castillo & Demoulin 1997). This interaction renders it difficult to decide if the fungus is a pathogen or saprophyte. For example, several species of *Fusarium* were recovered from vegetable crops (Mandeel *et al.* 1994), tomato (Abdul-Wahid *et al.* 1997), seeds (Thomos & Buddenhagen 1980; Klein *et al.* 1990; Onyike & Nelson 1992), and roots (Kretuzer 1972; Lesile *et al.* 1990; Smiley & Patterson 1996) but were not pathogenic. Although, this study was not designed to characterize host specificities and pathogenecity of the isolated *Fusarium* species to date palm. However, for quantitative comparisons, evaluation of spectra and diversity of *Fusarium* species in diseased and healthy plants is essential.

Predisposing plants by some stress condition is likely to result in poor growth and disturbed metabolic activity that stimulates some opportunistic *Fusarium* species to increase in population and attack (Lesile *et al.* 1990). Stress factors such as those induced by drought, high salinity and soluble salts are known to be conducive to disease incidence. For example, *Fusarium moniliforme* is known to grow and sporulate at very low water potential (Nelson *et al.* 1990) and *F. solani* can tolerate high salinity and temperature (Burgess 1981; Burgess *et al.* 1988a; Saremi *et al.* 1999;). Hence, it becomes apparent that these fungi can selectively escaped from unfavorable soil condition to become established within the host tissue of the roots to induce the disease later.

Recovery of the *Fusarium* species is likely to vary with isolation types used which is dependent on mode of persistence in soil (Mandeel 1996). Also, because composition of the population was analyzed from relatively narrow geographical transect and was not seasonal with saline and poor organic matter (Table 3), isolation types applied were aimed to recover species from these habitats. Maximum colony counts were observed from plant debris (723 isolates), followed by roots (540 isolate) and soil (410 isolates) (Table 5). Isolates from plant debris favors profusely sporulating species or which are basically chlamydospores former (Burgess & Summerell, 1992) and forming thickened hyphae (Nyvall, & Kommedahl, 1968). Recovery from plant roots enhances species that are competitive saprophytes (Cook & Bruell 1968; Nyval 1970a; Cook & Rayner 1984) present as hyphae and micro or macro conidia and weak secondary invaders. The recovery from soil samples, favor species present at low frequencies in discrete locations mostly adhering to humus or mineral particles in the form of chlamydospores (Nash *et al.* 1961; Nyval 1970a; Smith & Snyder 1975) (Tables 4 and 5). Evidently, species present in plant roots and debris are usually more stable once established whereas those found on soil particles are more vulnerable to changes in soil and climatic conditions (Mandeel *et al.* 1995). Higher species density and spectrum in plant debris compared to soil could be a survival mechanism withstanding the long drought and summer season and unfavorable soil condition as dormant spore and thickened hyphae, throughout the year (Lesile *et al.* 1990). Sangalang *et al.* (1995 b) sustained that fungi that occurred in arid zones must be able to survive long dry periods and competitively colonize suitable substrates during the occasional short periods when soil moisture was available for growth. These periods of moisture availability can occur when soil temperature are relatively high.

In their earlier studies, Burgess & Summerell (1992), proposed that plant debris in dry environment is less likely to decompose rapidly and is consequently

more favorable for the survival of fungi. Jeschke *et al.* (1990) reported a higher diversity of *Fusarium* species using plant debris plating technique compared to soil dilution plating. As most of the species recovered in current study are chlamydospore producer, it is more likely that they will survive well in both debris and soil. Species such as *F. moniliforme*, *F. poae*, and *F. avenaceum* that are not chlamydospore producers, rarely recovered from soil samples using the dilution method. These species would only survive as an active or weak saprophytes in plant tissues that they have colonize (Summerell *et al.* 1993).

Findings of this study indicate that high soil moisture in saline habitats supported the growth of *Fusarium* species and presumably developed a mechanism to adopt water stress condition in these areas by producing cytoplasmic osmolytes i.e. polyalcohols to maintain turgor at levels required for growth. Such results are also in accordance with the findings which suggested that intra specific competition for moisture and organic matter are considered more important factors in regulating species abundance (Shearer & Zare-Maniva 1988). Although the specific role of root exudates, chemical components and microbial interactions in the rhizosphere were not determined in this study, it seems obvious that environmental stress may alter exudates composition thereby affecting species occurrence.

An attempt to relate *Fusarium* species recovery from various locations to either north or west transect, based on geographical distribution of all the 27 villages formed the basis of Table 6. The total number of *Fusarium* isolates obtained using various isolation methods was greater in northern transect (288.88) compared to west (143.48). These findings contradict with the previously reported distribution of *Fusarium* species of protected vegetable crops in Bahrain (Mandeel & Abbas, 1994). Despite the fact that both transects fall within relatively small geographical area (< 25 to 30 km) and are affected by almost similar agricultural practices, nevertheless villages of western transect, in current study, are fewer than north. It is probable that dense vegetation cover, availability of irrigation water from well and multi cropping system may be responsible for such high fungal recovery and species diversity in these locations. Comparatively, the frequency *Fusarium solani* was absolute among all locations and isolation types with 27.93%, relative density value, followed by *F. oxysporum* (15.08%) and *F. lateritium* (8.1%). The data presented here substantiated other reports of previous workers (Mandeel *et al.* 1995; Mandeel 1996), in that *F. solani* and *F. oxysporum* dominate both cultivated and natural habitats of Bahrain that form characteristically extremely unfavorable environment for fungal growth.

Summerell *et al.* (1993) argued that population of fungi, which adapted or evolved in association with vegetative or confined climatic features might be expected to have a restricted distribution. Likewise, fungal composition and assemblage in soil is governed directly by age and type of higher plant communities (Stoner 1981; Mandeel 2002). However, in noncultivated soils, no specific saprophytic *Fusarium* species are known to be limited, or always associated with any particular habitat or plant community. Undoubtedly, no clear pattern can be established on the influence of gradual increase in plant heights and *Fusarium* species distribution and spectra in roots, plant debris and soil (Table 4-5). Edaphic factors affecting directly plant growth and soil biotic and abiotic are likely to be more important in determining *Fusarium* abundance and diversity than plant height (Griffin 1972; Christensen 1981; Skujins 1984). Consequently, species composition and population in date palm community may reflect a well-established adaptation and tolerance to arid habitats of Bahrain.

The results of the current study evince that date palm roots, plant debris and soil, were mostly dominated by species of *F. solani* and *F. oxysporum* and to a lesser extent by low occurring species of *F. lateritium, F. moniliforme* and *F. equiseti*. The former species apparently have broad ecological tolerances and cosmopolitan distribution but tend to be illustrative of hot arid zones (Burgess & Summerell 1992). It is possible that the assemblage of these fungi have adapted to the extreme soil and environmental conditions and perhaps the unique vegetation of Bahrain. Moreover, unfavorable soil a-biotic factors such as high salinity and soluble salts coupled with low soil moisture and organic matter content may have accounted for limited species diversity, spectra and distribution. The findings substantiate the results of foregoing studies (Mandeel, 1996, 2002). In subsequent work, in order to better evaluate precisely dynamics of *Fusarium* population in date palm rhizosphere in arid environment, it will be inevitable to contrast both disease and healthy plants in cultivated and noncultivated habitats and the influence of various soil factors on their distribution and survival. Moreover**,** a detailed investigation at biochemical and molecular level is suggested to elucidate broader understanding on mechanisms of survival, biodiversity and growth of the genus *Fusarium* in arid environment.

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