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Utilization of Edible Oil by Fungi Isolated From the Rhizosphere of Saudi Arabian Plants

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ABSTRACT. Fungi were isolated from different types of soils in Saudi Arabia using three different media. Their ability to utilize edible oil as a sole carbon was then examined. Out of a total of 74 fungal species isolated, 29 could utilize edible oil as the sole source of carbon. Initiation of growth was shown by some fungi in a Dox medium containing edible oil and sucrose as carbon sources but these fungi failed to grow on the medium containing edible oil as the only carbon source.

Common fungal genera such as *Aspergillus, Alternaria, Curvularia, Drechslera, Penicillium, Cladosporium* and *Trichoderma* have the capability to grow on hydrocarbons, which are degraded into smaller C-molecules (Llanos and Kjoller 1976, Savitha 1986). Among these compounds petroleum products (Bokhary and Parvez 1993), diesel fuel (Hettige and Sheridan 1984), emulsion paints, cellulose ether solutions (Springle 1978), as well as edible oils (Khalis and Manoharacharya 1985) have been shown to be degraded. Most of these materials could be converted into useful products through biodegradation by fungi (Carrod 1978, Tan and Leong 1986), but sometimes, as in the case of cooking oil, undesirable aromatic compounds might result, which affect the quality of the edible oil (Khalis and Manoharacharya 1985). It is nevertheless, recognized that such fungi could be of great help in ridding the environment of waste hydrocarbons (Arab News Feb. 7, 1992).

Recently, in Saudi Arabia, the degradation of various substrates such as petroleum products (Bokhary and Parvez 1993), starch (Bokhary and Parvez 1992), and cellulose (Bokhary and Parvez, 1994) by fungi have been reported.

In the present study an attempt is made to isolate fungi from various types of soils in Saudi Arabia that have the ability to utilize edible oils as the only source of carbon. this ability might then be exploited to enable the removal of waste oils from the environment.

Materials and Methods

Soil samples, collected from the rhizosphere region of three cultivated (Alfalfa, Date palm and Grape-vine) and two wild plants (*Cuccumis* sp. from sand dunes and *Zizyphus spina christi* from desert). They were used for the isolation of rhizosphere fungi by the dilution plate method previously described (Bokhary *et al.* 1984). To obtain isolates with the ability to utilize edible oils, two of the media were supplemented with this either as a sole or combined source of carbon. In one of these Dox medium to which 1% (v/v) edible oil (corn oil, Afia) was added to replace sucrose as the only source of carbon (Garg *et al.* 1984). In another, the Dox medium was modified to contain 3% (w/v) sucrose and 0.1% (v/v) edible oil. Dox medium with 3% (w/v) sucrose was used as a control. Rose bengal (0.03 g/L) was added to the media to reduce the growth of fast growing fungi, while streptomycin sulphate (0.033 g/L) was added to eliminate bacterial growth for the preliminary isolation of fungi from soil. Those species shown to be able to grow on edible oil containing media were further examined to determine their radial growth rates on these media.

Isolated fungi were identified by reference to the monograph of Raper and Fennell (1965), Zycha *et al.* (1969), Gilman (1971), Ellis (1971, 1976), Pitt (1979), Schipper (1978), Ramirez (1982), Howard (1983) and Nelson *et al.* (1983).

Results and Discussion

A total of 74 fungal species belonging to 17 genera were isolated from the rhizosphere of three cultivated (Alfalfa, Date-palm, Grape-vine) and two wild plants (*Cuccumis* sp., *Zizyphus spina christi*). Generally a larger number of fungal species were found in the rhizosphere soil of cultivated plants than uncultivated ones (Table 1). The rhizosphere soil of grape vine yielded the highest number of species followed by that of date-palm. Fungi of the genus *Aspergillus* were predominant with 23 species followed by *Penicillium* (14 species), *Chaetomium* and *Scytalidium* (4 species each), *Alternaria, Fusarium,*

Mucor and *Ulocladium* (3 species each). Generally the Dox medium with sucrose and 0.1% edible oil as carbon source yielded the same number of fungal species in the case of both cultivated and wild plants but the medium containing 1% edible oil as a sole source of carbon yielded fever fungal species in both cases. *Aspergillus candidus, A. carbonarius, A. ellipticus, A. flavus, A. fumigatus, A. niger, A. terreus and Penicillium chrysogenum* were isolated from the rhizosphere soil of all cultivated and wild plants and were able to grow on all of the three types of media used.

From the 74 fungal species isolated (Table.1), 50 species belonging to 15 genera (Table 2) were chosen to examine their radial rate of growth on media containing edible oils. Those fungi which exhibited an increase in the number of colonies on medium containing sucrose with 0.1% edible oil as carbon source or could grow on medium containing only edible oil (1 %) as a carbon source were chosen for this experiment. Dox medium containing only sucrose (3%) served as a control.

All the 50 fungi chosen for test showed greater extension of growth rate in the medium containing sucrose and 0.1% edible oil as carbon source than in the control medium.

There was a general decrease in the rate of growth of fungi, as compared to medium containing sucrose +0.1% edible oil, in the medium containing 1% edible oil as sole C-source. Of the 50 fungi tested, 20 completely failed to grow, but the rates of growth of those fungi which could grow on medium containing only edible oil, were generally higher than those observed on the control medium.

Species of *Aspergillus* and *Penicillium* have been previously reported to have some abilities to biodegrade complex carbon compounds including edible oil (Khalis and Manoharacharya 1985), crude oils (Fedorak *et al.* 1984) and petroleum products (Bokhary and Parvez 1993). This study confirms their potential for use as degraders of edible oil. *Fusarium* in contrast failed to grow on edible oil as sole carbon source but did make some growth on 0.1% edible oil with sucrose.

Table 1. Total number of colonies of each fungal species isolated per gram of soil on different media at room temperature (22-25 °C).
a) Dox medium (control), b) Dox medium with 3% sucrose and 0.1% edible oil,
c) Dox medium with 1% edible oil but without sucrose.

Fungal species	Rhizosphere soil														
	Cultivated plants									Wild Plants					
	Alfalfa			Date-palm			Grape-vine			*Cuccumis* sp.			*Z. spina christi*		
	a	b	c	a	b	c	a	b	c	a	b	c	a	b	c
Alternaria alternata (Fr. : Fr.) Keissler	43 ± 5	33 ± 4	--	49 ± 6	39 ± 5	--	30 ± 7	32 ± 6	--	36 ± 5	39 ± 5	--	56 ± 7	53 ± 7	--
A. chlamydospora Mouchaca	--	--	--	52 ± 4	56 ± 7	55 ± 9			--	--	--	--	--	--	--
A. humicola Oudem	--	--	--	--			36 ± 6	39 ± 7	32 ± 4	--	--	--	--	--	---
Aspergillus amylovorus Panasenko ex Samson	49 ± 9	48 ± 8	--	56 ± 8	59 ± 7		62 ± 9	58 ± 8	--	34 ± 5	39 ± 5	--	46 ± 7	43 ± 6	--
A. apica Mehrotra & Basu	--	--	--	--	--	--	36 ± 7	39 ± 7	46 ± 6	--	--	--	--	--	--
A. avenaceus G. Smith	--	--	--	--	--	--	26 ± 5	36 ± 6	43 ± 7	--	--	--	--	--	--
A. caespitosus Raper & Thom	--	--	--	--	--	--	35 ± 4	43 ± 4	49 ± 7	--	--	--	--	--	--
A. candidus Link : Fr.	29 ± 5	42 ± 5	45 ± 6	46 ± 6	52 ± 7	55 ± 6	52 ± 6	53 ± 7	52 ± 5	16 ± 4	19 ± 5	19 ± 4	26 ± 7	25 ± 6	27 ± 6
A. carbonarius (Bain.) Thom	43 ± 6	45 ± 5	47 ± 5	24 ± 5	26 ± 5	24 ± 5	57 ± 7	59 ± 7	55 ± 6	12 ± 4	13 ± 4	15 ± 4	26 ± 4	26 ± 5	29 ± 5
A. carneus (van Tieghem) Blochwitz	22 ± 5	29 ± 5	32 ± 5	26 ± 4	36 ± 4	39 ± 4	39 ± 4	46 ± 5	52 ± 6	--	--	--	12 ± 3	19 ± 5	24 ± 4
A. clavatus Desm.	--	--	--	--	--	--	36 ± 7	46 ± 5	49 ± 5	--	--	--	--	--	--
A. ellipticus Raper & Fennell emend. Al-Musallam	43 ± 5	46 ± 6	49 ± 5	64 ± 4	60 ± 5	63 ± 6	73 ± 6	76 ± 6	83 ± 7	25 ± 4	29 ± 5	25 ± 5	36 ± 5	33 ± 5	42 ± 6
A. flavipus (Bain. & Sartory) Thom & church	--	--	--	46 ± 6	32 ± 4	--	--	--	--	--	--	--	--	--	--
A. flavus Link : Fr.	52 ± 5	59 ± 6	58 ± 5	49 ± 7	56 ± 7	59 ± 6	29 ± 5	32 ± 4	42 ± 6	16 ± 4	21 ± 4	14 ± 5	18 ± 4	26 ± 5	15 ± 3
A. fumigatus Fres.	32 ± 6	43 ± 6	22 ± 5	39 ± 5	40 ± 5	49 ± 4	39 ± 3	49 ± 3	32 ± 5	26 ± 5	42 ± 5	26 ± 4	32 ± 5	49 ± 5	23 ± 4
A. nidulans (Eidam) Winger	--	--	--	--	--	--	--	--	--	21 ± 4	16 ± 4	--	21 ± 5	15 ± 4	--

Table 1. *Continued*

Fungal species	Rhizosphere soil														
	Cultivated plants									Wild Plants					
	Alfalfa			Date-palm			Grape-vine			*Cuccumis* sp.			*Z. spina christi*		
	a	b	c	a	b	c	a	b	c	a	b	c	a	b	c
A. phoenicis (Corda) Thom	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--
A. raperi Stolk & Meyer	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--
A. restrictus G. Smith	--	--	--	32 ± 4	45 ± 6	46 ± 5	--	--	--	--	--	--	--	--	--
A. rugulosus Thom & Raper	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--
A. terreus Thom	26 ± 4	43 ± 5	45 ± 5	29 ± 5	39 ± 6	43 ± 7	49 ± 7	63 ± 4	40 ± 5	16 ± 6	32 ± 5	14 ± 3	15 ± 5	27 ± 6	10 ± 3
A. ustus (Bain) Thom & Raper	--	--	--	--	--	--	--	--	--	--	--	--	36 ± 5	49 ± 6	20 ± 4
A. versicolor (Vuill) Teraboschi	--	--	--	--	--	--	--	--	--	21 ± 3	16 ± 4	--	--	--	--
Blastomyces brasiliensis (Splendore) Conant	29 ± 5	43 ± 4	22 ± 6	23 ± 5	39 ± 6	20 ± 4	--	--	--	--	--	--	--	--	--
Chaetomium bostrychodes Zopf.	--	--	--	36 ± 5	59 ± 7	30 ± 5	--	--	--	--	--	--	--	--	--
C. carinthiacum Sorgel	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--
C. cochlioides Palliser	--	--	--	17 ± 5	13 ± 6	--	--	--	--	--	--	--	--	--	--
C. globosum Kunze : Fr.	29 ± 4	46 ± 7	18 ± 5	32 ± 5	46 ± 6	23 ± 4	46 ± 6	69 ± 7	47 ± 6	--	--	--	--	--	--
C. murorum Corda	--	--	--	39 ± 5	39 ± 5	--	--	--	--	--	--	--	--	--	--
C. nigricolor Ames	--	--	--	--	--	--	25 ± 5	29 ± 6	--	--	--	--	--	--	--
C. robustum Ames	--	--	--	--	--	--	--	--	--	16 ± 4	39 ± 6	26 ± 5	--	--	--
C. uniporum Aue & B. Muller	--	--	--	--	--	--	--	--	--	26 ± 6	13 ± 4	--	20 ± 3	10 ± 3	--
Drechslera australiensis Scharif ex Lam	26 ± 5	--	--	39 ± 5	13 ± 3	--	46 ± 5	32 ± 6	--	12 ± 2	11 ± 2	--	18 ± 4	6 ± 4	--

Table 1. *Continued*

Fungal species	Rhizosphere soil														
	Cultivated plants									Wild plants					
	Alfalfa			Date-palm			Grape-vine			*Cuccumis* sp.			*Z. spina christi*		
	a	b	c	a	b	c	a	b	c	a	b	c	a	b	c
Fusarium chlamydosporum Wollenw. & Reinking	–	–	–	18 ± 4	26 ± 5	–	29 ± 6	36 ± 5	–	–	–	–	14 ± 5	17 ± 5	–
F. flocciferum Corda	26 ± 5	36 ± 5	–	32 ± 5	42 ± 6	–	45 ± 6	52 ± 7	–	–	–	–	–	–	–
F. oxysporum Schlecht.	36 ± 6	26 ± 5	–	43 ± 4	32 ± 4	–	25 ± 5	26 ± 4	–	–	–	–	–	–	–
Geotrichum candidum Link	32 ± 6	43 ± 5	40 ± 5	25 ± 6	32 ± 5	20 ± 4	39 ± 7	46 ± 7	33 ± 5	–	–	–	18 ± 5	29 ± 5	32 ± 5
G. capitatum (Diddens & Lodder) V. Arx	–	–	–	18 ± 4	16 ± 5	–	–	–	–	–	–	–	–	–	–
Mucor circinellioides van Tieghem	52 ± 7	63 ± 6	–	32 ± 6	46 ± 3	–	24 ± 5	40 ± 2	–	10 ± 7	27 ± 2	–	26 ± 5	40 ± 5	–
M. hiemalis Wehmer	16 ± 3	7 ± 2	–	39 ± 7	23 ± 5	–	26 ± 4	14 ± 3	–	–	–	–	–	–	–
M. racemosus Fres.	–	–	–	18 ± 4	6 ± 3	–	–	–	–	–	–	–	–	–	–
Penicillium bravicompactum Dierckx	–	–	–	–	–	–	12 ± 3	32 ± 5	–	–	–	–	–	–	–
P. brunneum Udagawa	–	–	–	–	–	–	16 ± 3	26 ± 5	20 ± 4	–	–	–	–	–	–
P. chrysogenum Thom	27 ± 4	37 ± 4	22 ± 4	24 ± 5	39 ± 6	34 ± 5	36 ± 5	43 ± 5	42 ± 5	18 ± 3	28 ±4	22 ±4	24 ±4	42 ± 7	18 ± 4
P. citreonigrum Dierckx	–	–	–	–	–	–	12 ± 4	6 ± 3	–	–	–	–	–	–	–
P. cyaneum (Bain & Sartory) Biourge	–	–	–	13 ± 2	26 ± 3	14 ± 4	–	–	–	–	–	–	–	–	–
P. dierckxii Biourge	–	–	–	–	–	–	14 ± 4	6 ±3	–	–	–	–	–	–	–
P. expansum Link	–	–	–	17 ± 3	26 ± 5	–	18 ± 3	24 ± 5	–	–	–	–	–	–	–
P. funiculosum Thom	–	–	–	26 ± 4	36 ± 5	–	–	–	–	–	–	–	–	–	–
P. islandicum Sopp	–	–	–	–	–	–	15 ± 4	12 ± 3	–	–	–	–	–	–	–
P. phialosporum Udagawa	–	–	–	–	–	–	17 ± 3	26 ± 4	–	–	–	–	–	–	–

Table 1. *Continued*

Fungal species	Rhizosphere soil														
	Cultivated plants									Wild plants					
	Alfalfa			Date-palm			Grape-vine			*Cuccumis* sp.			*Z. spina christi*		
	a	b	c	a	b	c	a	b	c	a	b	c	a	b	c
P. rubrum Stoll	–	–	–	18 ± 5	18 ± 4	–	–	–	–	–	–	–	–	–	–
P. sclerotiorum van Byma	–	–	–	–	–	–	–	–	–	19 ± 4	26 ± 5	–	–	–	
P. thomii Maire	–	–	–	–	–	–	–	–	–	–	–	–	16 ± 4	27 ± 5	–
P. verrucosum Dierckx	–	–	–	–	–	–	17 ± 3	12 ± 4	–	–	–	–	–	–	–
Petriellidium sp.	21 ± 4	29 ± 5	–	–	–	–	–	–	–	–	–	–	–	–	–
Phialophora sp.	–	–	–	24 ± 3	20 ± 4	–	36 ± 4	32 ± 5	–	–	–	–	–	–	–
Rhizopus microsporus van Tieghem	–	–	–	–	–	–	–	–	–	19 ± 3	18 ± 3	–	25 ± 4	20 ± 4	
R. stolonifer (Ehrenb. Fr.) Vuill	–	–	–	–	–	–	–	–	–	19 ± 4	16 ± 4	–	–	–	–
Scytalidium album Beyer & Klingstrom	–	–	–	–	–	–	23 ± 4	29 ± 4	–	–	–	–	16 ± 4	26 ± 4	–
S. aurantiacum Klingstrom & Beyer	–	–	–	39 ± 5	46 ± 4	–	–	–	–	–	–	–	–	–	–
S. lignicola Pesante	–	–	–	–	–	–	41 ± 5	49 ± 5	–	18 ± 4	29 ± 5	–	–	–	–
S. terminale Rao & de Hoog	–	–	–	–	–	–	7 ± 2	6 ± 2	–	–	–	–	–	–	–
Stachybotrys microspora (Mathur & Sankhala) John & Davis	–	–	–	23 ± 4	49 ± 6	40 ± 5	–	–	–	–	–	–	–	–	–
Stemphylium botryosum Wallr.	29 ± 6	34 ± 5	–	–	–	–	–	–	–	–	–	–	–	–	–
Thielavia albomyces (Cooney & Emerson) Malloch & Cain	–	–	–	–	–	–	23 ± 5	20 ± 5	–	20 ± 4	16 ± 3	–	–	–	–
Trichoderma harzianum Rifai	–	–	–	32 ± 5	59 ± 6	50 ± 6	26 ± 6	43 ± 6	39 ± 5	–	–	–	–	–	–
T. viride Pers. :Fr.	16 ± 4	16 ± 3	–	–	–	–	–	–	–	–	–	–	–	–	–
Ulocladium atrum (Preuss) Simmons	39 ± 4	69 ± 7	40 ± 5	32 ± 5	49 ± 5	40 ± 5	26 ± 4	46 ± 5	40 ± 4	49 ± 6	49 ± 5	35 ± 4	–	–	–
U. chlamydosporum Mouchacca	–	–	–	–	–	–	–	–	–	–	–	–	–	–	
U. tuberculatum Simmons	–	–	–	–	–	–	–	–	–	–	–	–	23 ± 6	22 ± 7	

Total: 17 genera 74 species; each reading is the mean of 5 replicates; ± standard deviation.

Table 2. Radial rate of growth of fungi per day on different media at room temperature (22-25°C)

Fungal species	Radial extension (mm/day)		
	a	b	c
Alternaria chlamydospora	14 ± 2	23 ± 3	16 ± 4
A. humicola	13 ± 3	16 ± 4	14 ± 3
Aspergillus apica	24 ± 4	26 ± 5	23 ± 3
A. avenaceus	6 ± 2	8 ± 2	6 ± 2
A. caespitosus	5 ± 2	7 ± 2	7 ± 3
A. candidus	3 ± 1	5 ± 1	5 ± 2
A. carbonarius	16 ± 4	18 ± 4	16 ± 3
A. carneus	6 ± 2	9 ± 2	7 ± 3
A. clavatus	4 ± 2	7 ± 3	5 ± 2
A. flavus	12 ± 3	19 ± 3	14 ± 4
A. fumigatus	7 ± 2	13 ± 4	10 ± 3
A. niger	11 ± 3	16 ± 3	12 ± 4
A. ochraceus	5 ± 1	6 ± 2	4 ± 2
A. raperi	2 ± 1	5 ± 1	5 ± 2
A. restrictus	7 ± 2	11 ± 4	10 ± 3
A. rugulosus	4 ± 1	4 ± 2	4 ± 1
A. terreus	2 ± 1	3 ± 1	3 ± 1
A. ustus	6 ± 2	12 ± 4	16 ± 3
Blastomyces brasiliensis	3 ± 1	7 ± 3	9 ± 2
Chaetomium bostrychodes	4 ± 1	5 ± 2	7 ± 2
C. carinthiacum	3 ± 2	6 ± 1	--
C. cochlioides	4 ± 1	6 ± 2	--
C. globosum	4 ± 2	9 ± 2	7 ± 1

Table 2. *Continued*

Fungal species	Radial extension (mm/day)		
	a	b	c
C. murorum	3 ± 1	6 ± 2	--
C. nigricolor	5 ± 2	9 ± 2	--
C. robustum	4 ± 1	9 ± 2	8 ± 2
C. uniporum	5 ± 2	7 ± 2	--
Fusarium chlamydosporum	16 ± 4	18 ± 3	--
F. flocciferum	19 ± 3	23 ± 5	--
Geotrichum candidum	15 ± 4	23 ± 4	20 ± 5
Mucor circinellioides	26 ± 5	32 ± 5	--
Penicillium brevicompactum	7 ± 2	9 ± 3	--
P. brunneum	8 ± 3	10 ± 3	5 ± 2
P. chrysogenum	11 ± 2	16 ± 2	6 ± 3
P. cyaneum	6 ± 3	9 ± 3	4 ± 2
P. expansum	5 ± 1	9 ± 3	--
P. funiculosum	7 ± 2	10 ± 3	--
P. sclerotiorum	3 ± 1	5 ± 1	--
P. thomii	4 ± 1	9 ± 2	--
P. petriellidium sp.	7 ± 2	11 ± 3	--
Phialophora sp.	11 ± 3	15 ± 2	--
Scytalidium album	8 ± 2	12 ± 3	--
S. aurantiacum	6 ± 2	9 ± 2	--
S. lignicola	7 ± 3	11 ± 2	--
Stachybotrys microspora	13 ± 4	19 ± 4	10 ± 2
Stemphylium botryosum	9 ± 3	12 ± 4	--

Table 2. *Continued*

Fungal species	Radial extension (mm/day)		
	a	b	c
Trichoderma harzianum	16 ± 4	20 ± 3	12 ± 3
T. viride	12 ± 2	14 ± 3	--
Ulocladium atrum	11 ± 3	19 ± 4	8 ± 2
U. chlamydosporum	12 ± 4	21 ± 5	7 ± 3

Total 15 genera, 50 species of fungi - readings are the means of 5 replicates; ± standard deviation

a) Dox medium control), b) Dox medium + 0.1% edible oil,

c) Dox medium without sucrose + 1% edible oil.

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استخدام الزيوت الصالحة للأكل كمصدر كربوني وحيد للفلورا الفطرية المعزولة من تربة المملكة العربية السعودية

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قسم النبات والأحياء الدقيقة - كلية العلوم - جامعة الملك سعود
ص .ب (٢٤٥٥) - الرياض ١١٤٥١ - المملكة العربية السعودية

لقد تم عزل الفطريات التي تستخدم الزيت الصالح للأكل كمصدر كربوني وحيد والمعزولة من أنواع مختلفة من التربة بثلاث طرق .

لقد وجد تسعة وعشرون نوعاً من بين أربعة وسبعون نوعاً فطرياً استطاعت استعمال الزيت الصالح للأكل كمصدر كربوني وحيد .

كما أوضحت الدراسة أن بعض الفطريات التي أظهرت نمواً بسيطاً على بيئة دوكس محتوية على الزيت الصالح للأكل والسكروز كمصدر كربوني فان هذه الفطريات لم تكن قادرة على النمو في وسط يحتوي فقط على الزيت الصالح للاكل كمصدر كربوني وحيد .