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First Record Fungi for Iraq

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Abstract

44fungal species were isolated from plant parts submerged in Al-Huwaiza marsh within Iraqi borders, and 7 new first records fungi in Iraq were isolated too, which have been illustrated and described as follows:-

Carbosphaerella leptosphaeriodes, Curvularia lunata var.aria, Graphium sp., Helicascus kanaloanus, Leptosphaeria obions, Stagnospora sp. and Ulocladium tuberculatum, Carbosphaerella leptosphaeriodes, Curvularia lunata var. aeria, Graphium sp., Helicascus kanaloanus, Leptosphaeria obions, Stagnospora sp., Ulocladium tuberculatum.

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Introduction

Al-Huwaizah marsh is an aquatic ecosystem extend between Iraq and Iran with freshwater body. Al-Huwaizah marsh locates between latitudes 31° 45¯ and 31° 00¯ in the north and longitude 47° 50¯ and 47° 25¯ in the east, passing through Iranian borders, 80 km X 30 km, (Scott, 1995) [25]. Reed plants (Phragmites australis Trin) and Typha (Typha australis Schum & Thonn) are the main main components of the vegetative cover in the marsh ecosystem. (Al-Edany, 1978), Many endemic fungal species play an important role in the biodegradation and bioremediation of marsh environment. Fungi play an important role in biodegradation process of plant debris submerged in marsh and bioremediation occurs during mycoremediation during decomposers (fungi) in the aquatic environment along with some types of bacteria (Mille-Lindblom and Tranvik, 2003; Mohsen and Abdulkadir, 1995; Muhsin and Khalaf, 2002) [21, 22, 23].

Hussein Al-Nasrawi (2006) was confirmed isolation of fungal diversity (fifteen species) as a new record for the first time in Iraq, isolated from the plant remains submerged in aquatic ecosystems in Iraq , in addition to many studies conducted in Iraq for the same mycological puroses (Jaber.,etal.2012; Graca et al., 2016) [11, 16]. Many species of Basidiomycetes were isolated from stems and leaves of the reed plant submerged in salt marshes in Belgium (Van Ryckegem et al., 2005). [28]

Materials and Methods

Collection of Samples

50 pieces of decomposed plants were collected from water body and sediments in Al Huwaizah marsh in Iraq during 2016. Samples were washed gently by tap water and then by distilled water . Plant debris were cut into small parts 7 - 5 cm long and and each 10 pieces were settled in the bottom of petri dish.

Preparation of Culture Media.

Potato Carrot Agar (PCA), which was obtained by weighing 20 g of potato and carrots after washing and peeling, then sliced and boiled with a quantity of distilled water, was sprayed well in a ceramic vase, filtered and placed in a 1 liter flask then added to the prepared mixture Of each of the potatoes and carrots media. the media objected to sterilizing process in autoclave under standard conditions for 20 minutes (250mg 0f chloramphenicol as antibiotic to inhibit bacterial growth.

Insolation and Identification of Fungi

In this study, two methods were used to isolate the fungi: direct isolation from the substrate. The humid chamber method was used to remove the previously prepared vegetable pieces from the beaker using sterile forceps and placed 7 to 5 pieces in a glass bowl of 15 cm diameter Petri dishes Sterilize the filter leaves, then moisten the filter leaves with sterilized distilled water and incubate the dishes under 25° C. The second method is the method of dilution Dilution method to isolate the fungus from the washing of submerged plant parts and summarized the method by withdrawing 10 ml of sterile distilled water, which was washed by the samples previously using a sterile pipette placed in a flask containing 90 ml of distilled distilled water and a well well and withdraw from it 1 ml transferred to A sterile glass dish with a diameter of 9 cm. The food medium, plate roast and incubation were incubated under 25° C. Three replicates were made of each sample.

The isolated fungi were classified under light microscope by using international taxonomic keys published in the following literatures: Arx etal., (1986); Boerema, G.H.(1993); Booth, (1971); Cai etal., (2003); Ellis, (1971); Ellis, (1976) ;Gohand Clement,(2003); Hoog and Guarro,(1995) ; Kohlmeyer and Kohlmeyer, (1979); Kohlmeyer and Kohlmeyer, (1991); Pitt, (979). Sivavanesan, (1987). [3, 4, 5, 6, 8, 9, 12, 14, 18, 19, 24, 26].

Results

The total fungal species isolated from Al-huwaizah marsh.

1-Carbosphaerella leptosphaeriodes I. Schmidt Nat. Naturschutz Mecklenburg 7,9-10,1969(publ.1971) (Table 1-2)

Description

Ascocarp 90-120 um in diameter, globose to subglobose shape . The Asci 40-45 X 60-80 um with 8

Table 1. Fungal species isolated from plant debris in Al-Audem marsh in Iraq

No.	Fungal species
1	*Alternaria alternata Keissler*
2	*Alternaria sp.*
3	*Aspergills candidus Link*
4	*A.flavus Link & Fries*
5	*A.fumigatus Fres*
6	*A.nidulans (Eidam) vuill*
7	*A.niger van Tieghem*
8	*A.terreus Thom*
9	*Aspergillus sp.*
10	*Aureobasidium pullulans (De Bary) Arnaud*
11	*Bipolaris hawaiiensis (M.B.Ellis) Subram.&Jain*
12	*Carbosphaerella leptosphaeriodes I.Schmidt*
13	*Chaetomium globosum Kunze & Fries*
14	*Chaetomium piluliferum Daniels*
15	*Chaetomium sp.*
16	*Chuppia sarcinifera Deighton*
17	*Cladosporium cladosporoides (Fresen) de Vries*
18	*Cladosporium sp.*
19	*Curvularia lunata var.aeria(Batista ,Lima &Vasconselos)M.B.Ellis*
20	*Curvularia penniseti (Metra)Boedijn*
21	*Emericella nidulans (Eidam) vuill.*
22	*Eurotium sp.*
23	*Exerohilum sp.*
24	*Fusarium oxysporum Schlecht.*
25	*Fusarium sp.*
26	*Graphium sp.*
27	*Graphium putredinis (Corda)Hughes*
28	*Helicascus kanaloanus Kohlm.*
29	*Leptosphaeria obions (Crouan et Crouan)Saccardo*
30	*Monodictys glauca (Cooke & Harken)Hughes*
31	*Mucor sp.*
32	*Mycosphaerella pneumatophora Kohlm,Ber.*
33	*Penicillium sp.*
34	*Phoma herbarum Westend.*
35	*Phoma sp.*
36	*Pleospora pelagica Johnson*
37	*Rhizopus stolonifer (Ehrenb:Fr.)Vuill.*
38	*Savoryella lignicola E.B.G.Johnes et Eaton*
39	*Stachybotrys atra Corda*
40	*Stagnospora sp.*
41	*Torula herbarum (Pers.)Link ex S.F.Gray*
42	*Trichurus spiralis Hasselbring*
43	*Ulocladium botrytis Preuss*
44	*Ulocladium tuberculatum Simmons*

Table 2. First record fungi

No.	Fungal species
1	*Carbosphaerella leptosphaeriodes I.Schmidt*
2	*Curvularia lunata var.aeria(Batista ,Lima & Vasconselos) M.B.Ellis.*
3	*Graphium sp.*
4	*Helicascus kanaloanus Kohlm.*
5	*Leptosphaeria obions (Crouan et Crouan) Saccardo*
6	*Stagnospora sp.*
7	*Ulocladium tuberculatum Simmons*

Ascospores. The Ascospore 15-18X 25-30 um, devided by triseptate, the two mid cells within the ascospore dark to brown color, whereas the terminal cells pale and surrounded by mucous sheath. The present isolate nearly like the isolate of Schmidt, 1969.This species consider as a new record for Iraq. The isolate was illustrated and kept in Basra herbarium under no. BASRA 2011. (Figure 1).

Description

The colony with black to gray color, The hyphae immersed under substrate surface . Conidiophore thicker than fungal filament (macronematous), subhyaline. Conidiogenous cell is polytretic. The conidia with curve shape divided with three septae to form four cells, the two mid cells thicker and darker from the two terminal pale cells . Conidia 10-15X 20-30 um. This species was previously isolated from painted wood and soil whereas our present fungus isolated from reed sample submerged in marsh sediments. Dry culture was kept in Basra herbarium under no. BASRA 2012. (Figure 2).

Description

The colony is gray to Olivaceous brown, Conidiophore thicker than fungal filament (macronematous) appears under dissecting microscope as Synnemata. Fungal hayphae immersed under the epidermis. Conidiogenous cell is monobasic type. Conidia 5-7X 15-20 um. Oval to cylindrical shape, with rounded end, pale color without, unseptated. Our isolated fungus resemble species Graphium putredinis isolated by Huges, 1958 [15]. Our present isolate differentiated by its shape and size (cylindrical 2-4X5-11 um. The species isolated from reed segment submerged in marsh sediment. Dry culture was kept in Basra herbarium under no. BASRA 2013. (Figure 3).

4 -Helicascus kanaloanus Kohlm.Can.J.Bot.47,1471.1969

Description

The Ascocarp globose , immersed , 400-250 um high, 400-800 with ostule. Black to dark brown color. Asci 200-300 um., bitunicate, with 8 ascospores. The ascospore 15-25X35-50 um. Arranged inside ascus as uniseriate. The ascospore devided by septum into 2 dark cells, with funnel shape. cell wall of ascospore surrounded by two layers. There are two germination pores in the ends of ascospore. The ascospore differentiated by gelatinous layer clearly appears when immerse in water drop (disappear with lactophenol stain). The present fungus isolated from Typha segment submerged in marsh sediments , illustrated and kept in Basra herbarium under no. BASRA 2014. (Figure 4)

Description

Ascocarp sub globose, immersed, with high about 100-300 and diameter 200-400 um., black to dark brown color, usually covered by brown filaments. The ascocarp coated by two layers , large dark external layer and pale small internal layer. The asci thick, bitunicate,14-20 X 150-300 um. Each ascus contain 8 ascospores, 8-15X 25-40 um. Arranged inside the ascus as uniseriate in the top of the ascus wherease as biseriate in middle site. The ascospore divided by three septae to form 4 cells, the two middle cells dark brown and larger than the terminal smallest cells. This species was previously isolated from hrbal plants and from mangrove area in Australia .The present fungus isolated from typha segment submerged in marsh sediment,

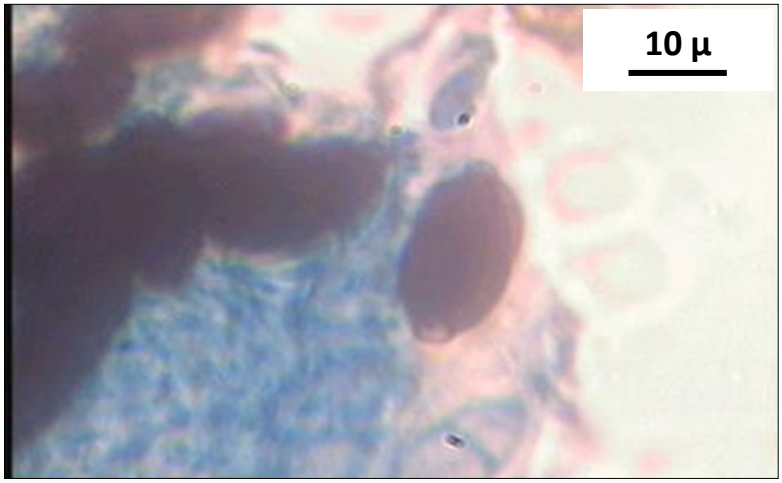

Figure 1- Ascospores of Carbosphaerella leptosphaeriodes

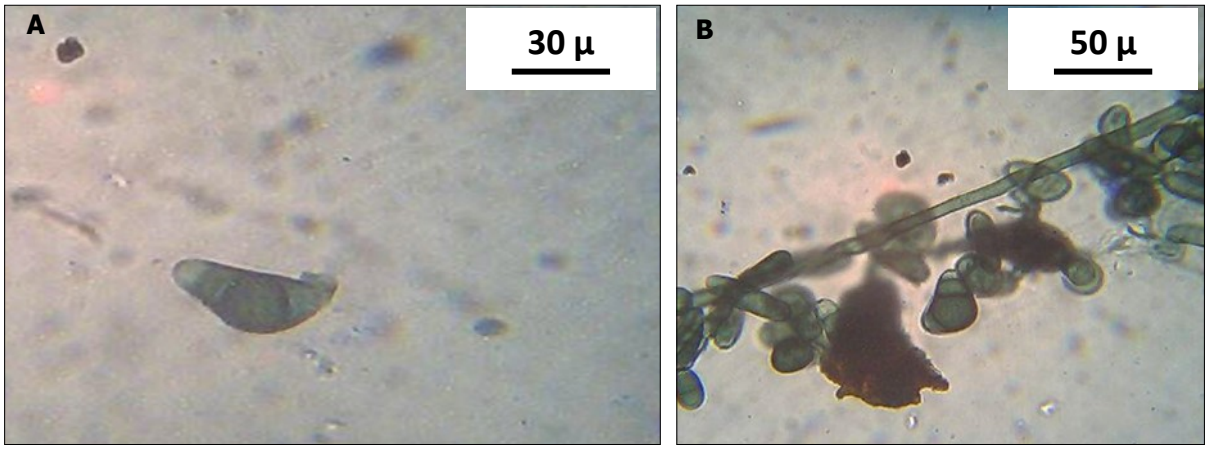

Figure 2. A) Conidia , B) Conidiophore of *Curvularia lunata var. aeria*

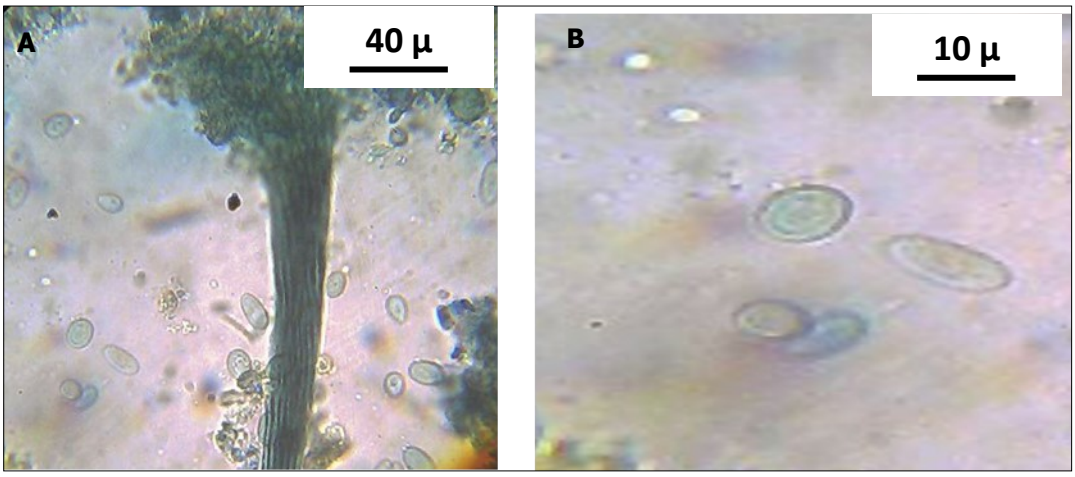

Figure 3. A) Conidiophore, B) conodia of Graphium sp

Figure 4. Ascospores of Helicascus kanaloanus

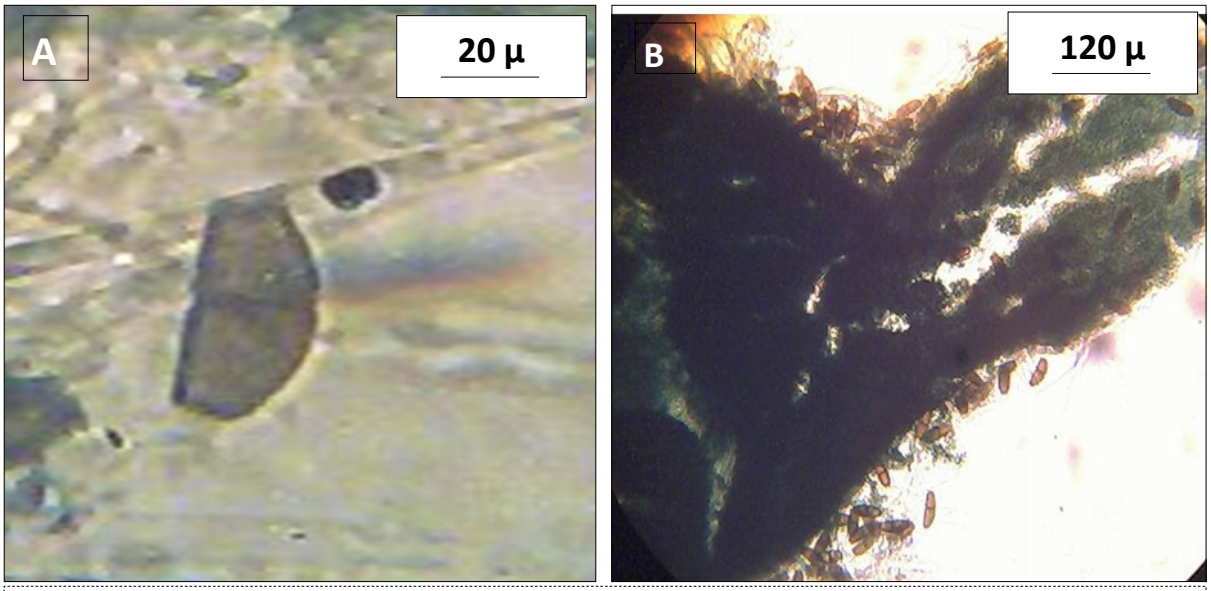

Figure 5. A) Ascospore, B) Ascocarp of Leptosphaeria obions

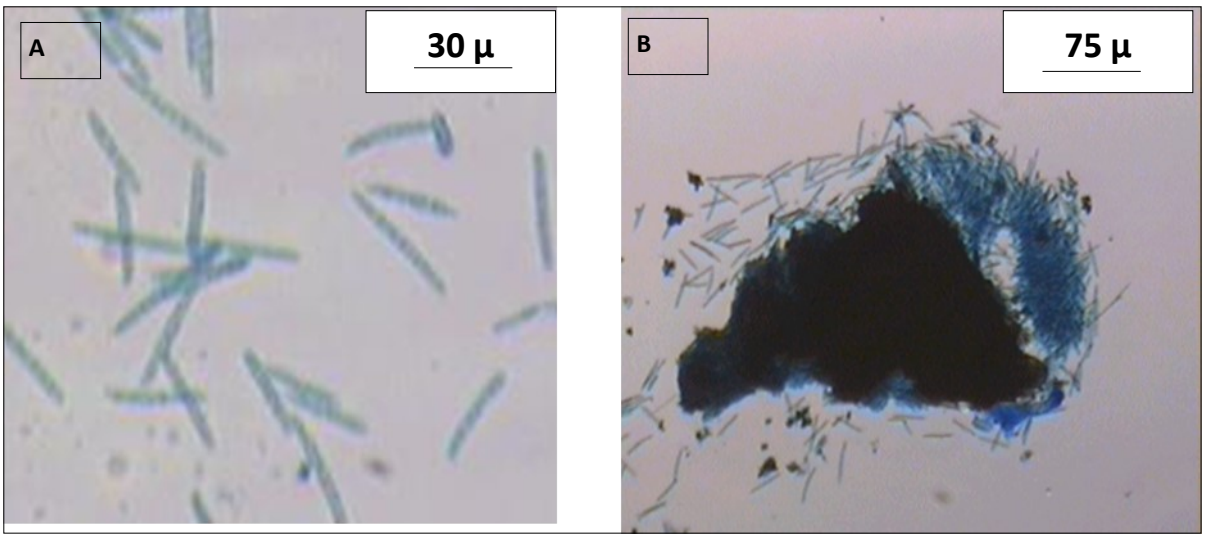

Figure 6. A) Conidia, B) Pycnedium of *Stagnospora sp.*

illustrated and kept in Basra herbarium under no. BASRA 2015. (Figure 5)

The Pycnidium sub globose, partialy immersed, with pale brown ostiole and short papillate.High of pycnidium 150-180 um, 100-200 um diameter with a neck about 10 um diameter. The conidia pale to brown color, cylindrical in shape, 4- 8 X 40-70 um., divided by 5-7 septae .The present isolate ressemble Stagonospora haliclysta which was previously isolated by Kohlm, 1973 (conidia size 3.5-4.5X 20-27 um., smaller than our isolate). The present fungus isolated from reed segment submerged in marsh sediment, and dconsider as first record in Iraq. It was illustrated and kept in Basra herbarium under no. BASRA 2016. (Figure 6)

7 -Ulocladium tuberculatum Simmons , 1967 , Mycologia 59: 83 -84.

Description

The fungal hyphae immersed, sub hyaline, with thick conidiophore 4-6 um. 160-200 um. length, pale brown color, divided by septae. Conidia 10-16 X 10-20 um. sub globose, like potato fruit, divided into several parts by septae cross shape

The species was firstly isolated in united states. The present isolate ressemble Tetracoccosporium paxianum, which isolated by Szabo, 1905. Our species was isolated from reed segment submerged in marsh sediment, kept in Basra herbarium under no. BASRA 2017. (Figure 7).

Discussion

Fungi Inhabit plant segments submerged in aquatic ecosystems used their enzymic complex system to biodegrade cellulose and produce carbon source, the most important matter for fungal metabolism process (Gessner, 1980; Garraway and Evans, 1984) [10].

Guaro et al., 1996, Abdullah et al., 1996 and Guaro et al., 1997 [13], the pioneers who worked on wetland area in Iraq, they isolated and identified many new fungal species and new record fungi forom plant segments submerged in marsh ecosystem in southern area of Iraq. The present study choosed one fresh and natural premium deep marsh ecosystem called al-Audem in Mysan province to suray fungal diversity and new records. This ecosystem consider a natural, undiscovered mycoflora eniched with organic materials and with highl quality sediments settle in the bottom of water body, that encourage growth of different fungal species. The present study contributed in recording seven new record fungi for Iraq from this marsh environment.

Conclusions

Several marshed in sothern area of Iraq still waiting more studies and novel works to discover more

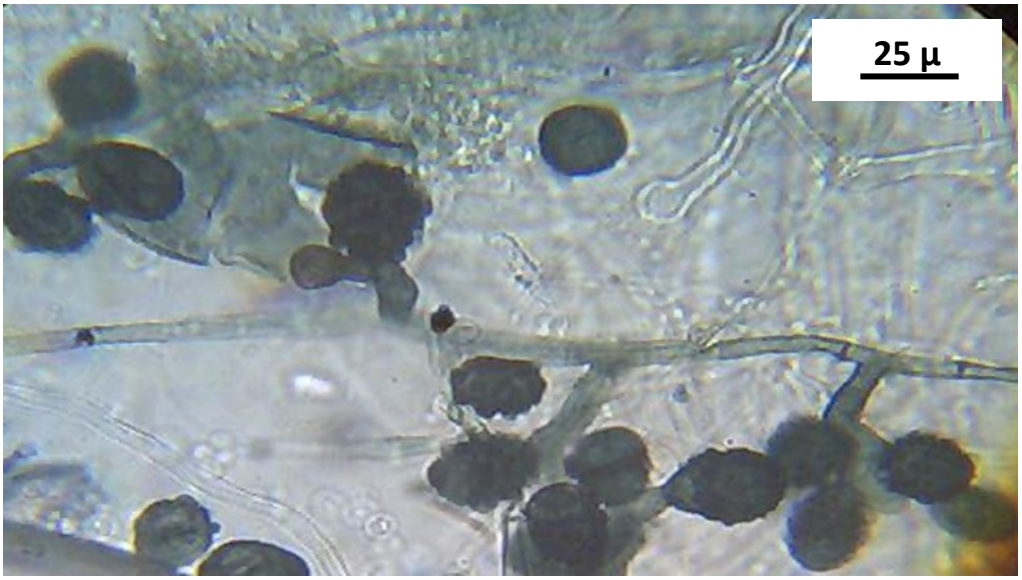

Figure 7. Conidia of Ulocladium tuberculatum

new species and new record fungi. The high quality water parameters of marsh ecosystem with enrichment of plant diversity, leads to establishment a perfect foundation of sediment layers embedded in the bottom of marsh environment, This study open the track for researchers to investigate the ecological niche of fungi in marsh community to detect more aquatic and sediment mycoflora of wetlands.

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