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Osmophilic/osmotolerant and halophilic/halotolerant fungi from mud, salt crusts, and air collected from Wadi El- Natrun lakes

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Abstract. A total number of osmophilic and osmotolerant mycobiota (45 species related to 21 genera) were collected from mud (the most dominant genera were *Aspergillus* (6 species), *Emericella* (3), *Eurotium* (3), *Fusarium* (3), and *Penicillium* (4), salt crusts (*Aspergillus* (8), *Penicillium* (4) and *Emericella* (3) and aerobiota (*Cladosporium* (4), *Alternaria* (3), *Aspergillus* (6) of Wadi El-Natrun lakes on Czapek Dox agar supplemented with 40% sucrose during the seasons of study. The halophilic and halotolerant mycobiota (22 species related to 13 genera) were collected from mud, salt crusts (only *Acremonium strictum*) and aerobiota (*Cladosporium* (3 species), *Aspergillus* (5), and *Alternaria* (3) on Czapek Dox agar supplemented with 10% sodium chloride during the seasons of study.

Keywords: *Osmophilic and osmotolerant, halophilic and halotolerant, mycobiota, Wadi El- Natrun lake*

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1. Introduction

Fungi can grow over a wide range of solute concentrations. Only a few species can grow at the high osmotic pressures characteristic of environments having either supersaturated brine and sugar concentrations (i.e., reduced water activity) (Kim et al., 2014). The osmophilic organisms are microorganisms growing better on media containing high sugar concentration (Alsharjabi and Al-Zubeiry, 2009).

Halophiles are salt-loving organisms that inhabit hypersaline environments. They can be loosely classified according to their salt requirement into 3 categories: slight, moderate, and extreme halophiles which grow optimally at 0.2-0.85 mol/L (2-5%) NaCl; 0.85-3.4 mol/L (5-20%) NaCl and above 3.4-5.1 mol/L (20-30%) NaCl, respectively (Das Sarma and Arora, 2001). In contrast, nonhalophiles can grow optimally at <0.2 mol/L (2% NaCl) (Das Sarma and Arora, 2001). Halotolerant organisms can grow over a broad range in the high and the absence of a high concentration of salts. Moustafa (1975) identified more than one hundred osmophilic species. Of these *A. ochraceus, Alternaria chlamydospora, A. alternata, P. chrysogenum, P. notatum, Cephalosporium recifei, Fusarium equiseti, Stachybotrys atra,* and *Drechslera hawaiiensis* were the broadest prevalent species from salt marshes of Kuwait.

Abdel-Hafez et al. (1977) isolated 95 species which related to 39 genera from the soil of salt marches in Egypt, on Czapek's agar media supplemented with 30% sucrose and on 60% at 28 ºC and 45 ºC and found that at 28 ºC *Aspergillus* and *Penicillium* were most frequent genera. From the preceding genera: *A. niger, A. fumigatus, A. terreus, P. notatum A. flavus, C. herbarum, R. stolonifer, A. nidulans, Alternaria alternate,* and *A. versicolor* were the highest species. The best-studied fungal extremophiles are halophilic and halotolerant fungi, inhabiting hypersaline waters of salt lakes and salterns worldwide (Casamayor et al., 2002; Gunde-Cimerman et al., 2005; Petrovič et al., 2002). Steiman et al. (2004) isolated many species of the genera *Aspergillus*, *Chaetomium*, *Acremonium*, *Alternaria*, *Cunninghamella*, *Ulocladium*, *Embellisia*, *Fusarium*, *Phoma*, *Sporothrix*, *Penicillium*, *Papulaspora*, *Geotricum*, *Curvularia*, *Mucor*, *Phialophora*, *Phaeoramularia*, *Sporormiella,* and *Thelebolus* from an alkaline, saline soda, Mono Lake, California. This work highlights the osmophilic and osmotolerant and the halophilic and halotolerant fungi isolated from extreme environments under extreme conditions.

2. Materials and methods

2.1. Collection of samples

Samples (salt crusts, mud, and air) were collected during January 2009-May 2010 from eight lakes (Fasida, Umm-Risha, Rosetta, Hamra, El El Zugm, Al Beida, Khadra, Al Gaar) of Wadi El-Natrun (depression) region, Egypt. Hands collected mud samples (nearly a depth of 0.5 m) at random from different sites inside and along the shore of lakes hands collected salt crust samples at random from mineral formation present along the shores of the lake.

2.2. Isolation of fungi

From mud and salt crusts: The dilution plate method was used to enumerate different fungal species as described by Johnson et al. (1972) and employed in this laboratory by Moubasher et al. (1977). At least five samples are taken at random from each place, then the five or more samples from each replication were brought into one composite sample, which was mixed thoroughly several times

From the air: Replicate plates of 9 cm diameter containing sterile agar media (5 for each medium type) were exposed to the air for 15 minutes during January 2006-May 2007. The plates were sealed, brought back to the laboratory, then incubated at 28 ºC for 7-21 days, during which the developing fungi were identified and counted.

2.3. Medium used for isolation of osmophilic and osmotolerant fungi

Czapek Dox agar supplemented with 40% sucrose used for isolation of osmophilic and osmotolerant fungi from all sources investigated.

2.4. Medium used for isolation of halophilic and halotolerant fungi

Modified Czapek Dox agar medium (in which glucose, 10g/L, replaced sucrose) supplemented with 10% sodium chloride was used to isolate halophilic and halotolerant fungi.

2.5. Identification of fungi

The fungal genera and species were identification of (purely morphologically based on macroscopic and microscopic features).

3. Results and discussion

3.1. Osmophilic and Osmotolerant fungi in mud collected from Wadi El-Natrun lakes

Twenty-five species related to 9 genera of osmophilic and osmotolerant fungi were recovered on Czapek Dox agar supplemented with 40% sucrose. Twenty-three species related to 10 genera were recovered on the control medium. *Aspergillus* (6 species), *Emericella* (3), *Eurotium* (3), *Fusarium* (3), and *Penicillium* (4) were the most common genera on 40% sucrose medium (Table 1). *Aspergillus* was the most dominant genera in the mud from Wadi El- Natrun lakes (Ismail et al., 2017; Moubasher et al., 2015; Gouda et al., 2020a, 2020b). *Aspergillus* was the most commonly encountered fungus in the 8 lakes' mud on 40% sucrose-Czapek agar and the control medium. The highest number of *Aspergillus's* CFUs was recorded from the 8 lakes during the 6 seasons of study and was lower on medium with 40% sucrose than those on the control medium.

The peak of *Aspergillus* was recorded from El Zugm during spring 2007. It represented 80.2% of the total fungal counts. *Aspergillus*, *A. terreus* (44.0%), *A. flavus* (12.7%), *A. niger* (8.7%), and *A. fumigatus* (13.7%) were recorded from the mud of all lakes during almost all seasons of study. The remaining *Aspergillus* species were isolated from 2 lakes: *A. candidus* (Al Beida and Al Gaar during winter 2006) and *A. sydowii* (Khadra during spring 2006 and Fasida during summer 2006). Our results correspond to Grishkan et al. (2003), who found that *Aspergillus niger* is the most abundant species, followed by *A. terreus, A. fumigates*, *A. flavus,* and *A. candidus* from the Dead Sea coastal sand and mud. Also, El-Wahid et al. (1982) found that *Aspergillus* (17 spp.)

and *Penicillium* (14) possessed the highest spectrum of species among all genera isolated from the intertidal mud-flats at Kuwait Bay. Among aspergilli, *A. ochraceus, A. niger, A. fumigatus, A. sydowii, A. flavus, A. terreus,* and *A.* oryzae were frequent.

Emericella (7.8% of the total fungi) and its dominant species *E. quadrilineata* (5.4%), were recorded from 5 lakes during almost all study seasons. The peak of *Emericella* was recorded during winter 2007 from Al Gaar. *E. nidulans* (0.7%) were isolated from 3 lakes (El Zugm, Al Gaar, and Khadra during summer 2006 and winter 2007) while *E. nidulans* var. *lata* was recorded from Al Gaar during winter and spring 2007. In this respect, *E. nidulans* was isolated from mangrove mud, India (Rai et al., 1966), and *Emericella acristata* from the Dead Sea coastal sand and mud Grishkan et al. (2003). However, *Emericella nidulans* was isolated with rare occurrence from “Casa Caiada” and “Bairro Novo” beaches, Brazil (Gomes et al., 2008). It was stated that *Emericella nidulans* is a typical soil fungus isolated from the desert and saline soil (Abdel-Fattah et al., 1977). The true osmophilic fungus, *Eurotium,* was recorded from 4 lakes (Hamra, Al Beida, Al Gaar, and Khadra) during 5 out of 6 seasons of study. The peak of *Eurotium* was recorded from Al Gaar and Al Beida during summer 2006 and winter 2007. *E. chevalieri,* which is known as osmophilic nature (Raper and Fennell, 1965), was recovered from 3 lakes (Hamra, Al Beida, and Al Gaar) during 3 seasons of study (spring, summer 2006, and winter 2007) while *E. repens* from 2 lakes (Al Beida and Khadra) and *E. amstelodami* from one lake (Al Gaar, winter 2007). In this respect, the genus *Eurotium* has been recorded in arid and saline zones in Syria (Sizova et al., 1967) and Kuwait (Moustafa, 1975; Moustafa and Al-Musallam, 1975). *E. ruber* was reported from mangrove mud, India (Rai et al. 1966) while *Eurotium chevalieri* from “Casa Caiada” and “Bairro Novo” beaches, Brazil (Gomes et al., 2008) and *E. amstelodami* and *E. herbariorum* from the Dead Sea coastal sand and mud Grishkan et al. (2003).

Fusarium (2.3%) and its dominant species *F. solani* (2.0%), were recorded in mud samples collected from 5 lakes during almost all seasons; *F. sterilihyphosum* and *F. verticillioides* were isolated from only Khadra lake during winter 2006. El-Wahid et al. (1982) reported six *Fusarium* species from Kuwait’s tidal mud-flats, of which only *F. oxysporum* was common. *Penicillium* (4.8%) and its dominant species *P. chrysogenum* (3.2%) were recorded from the mud of all lakes except El Zugm and Umm Risha during almost all seasons of study. The remaining *Penicillium* species were isolated from either 2 lakes: *P. puberulum* (Hamra during winter 2007 and Al gaar during autumn 2006) and *P. crustosum* (Fasida, spring 2007), *P. oxalicum* (Al Beida, summer 2006), and *Penicillium* spp. (Khadra, spring 2006). In the study of Grishkan et al. (2003), *P. aurantiogriseum* was the most common species followed by *P. brevicompactum* and *P. chrysogenum* from the Dead Sea coastal sand and mud. On the other hand, (Rai et al., 1966) isolated only three species of *Penicillium* (*P. nigricans, P. roseopureum,* and *P. stipitatum*) from mangrove mud in India, while El-Wahid et al. (1982) 14 *Pencillium* species from the tidal mud-flats of Kuwait (pH 8.1- 8.6). Of these, only P. *frequentans, P. cyclopium*, and *P. notatum* (= *P. chrysogenum*) were common*. Cladosporium* and its dominant species *C. cladosporioides* were recorded from 4 lakes (Umm Risha, Al Gaar, Khadra, and Fasida) during 3 seasons of study (spring, autumn 2006, and winter 2007). The other species *C. sphaerospermum* was recorded from Al Gaar Lake only during autumn 2006. Other uncommon were recovered from either 2 lakes during winter 2007: *Acremonium* (*A. furcatum*, Hamra and Rosetta), *Acremonium* spp. (Hamra and Khadra), from 1 lake: *Humicola* (*H. fuscoatra*, El Zugm, autumn 2006) and *H. grisea* (Al Beida, winter 2007). *Acremonium* (*A. rutilum*) was frequently isolated from “Casa Caiada” and “Bairro Novo” beaches in Brazil (Gomes et al., 2008) and unidentified *Humicola* species in the Hawaiian mangrove swamp soil (pH ranged from 5.5-6.0) (Lee and Baker, 1972). From our results, *Aspergillus candidus, Emericella lata, E. amstelodami, E. repens, Fennellia nivea, Fusarium sterilihyphosum, F. verticillioides,* and *Humicola fuscoatra* were isolated on medium supplemented with 40% sucrose and were absent on control medium.

Table 1. Summarized of osmophilic and osmotolerant fungi from mud in different lakes of Wadi El-Natrun

Fungal taxa	Hamra	Al Beida	El Zugum	Rosetta	Umm Reisha	Al Gaar	Khadra	Fasida
Acremonium	5			5			5	
A. furcatum	5			5				
Acremonium spp.	5						5	
Aspergillus	123456	12346	3456	23456	246	123456	123456	2356
A. candidus		1						
A. flavus	4	23	3456	235	246	1256	23456	2
A. fumigatus	36	6	3	3456	26	6	36	6
A. niger	2345	3	346	6	4	12356	256	235
A. sydowii							2	3
A. terreus	156	4	3456	2456	6	1346	146	56
Cladosporium					4	4	5	2
C. cladosporioides					4	4	5	2

Table 1. Summarized of osmophilic and osmotolerant fungi from mud in different lakes of Wadi El-Natrun								
Fungal taxa	Hamra	Al Beida	El Zugum	Rosetta	Umm Reisha	Al Gaar	Khadra	Fasida
C. oxysporum						4		
Emericella		134	34	34		1256	2345	
E. lata						56		
E. nidulans			3			5	3	
E. quadrilineata		134	34	34		125	34	
Eurotium	2	236				5	125	
E. chevalieri	2	23				5		
E. amstelodami						5	125	
E. repens		6						
Fennellia nivea			4					
Fusarium			24	3	24		12	56
F. solani			24	3	24		2	56
F. sterilihyphosum							1	
F. verticillioides							1	
Humicola		5	4					
H. fuscoatra			4					
H. grisea		5						
Penicillium	25	34		2		4	12	36
P. chrysogenum	2	4		2		4	1	36
P. duclauxii								6
P. oxalicum		3						
P. puberulum	5					4		
No. of genera (9)	3	5	4	5	3	5	7	4
No. of species (25)	4	16	13	13	9	18	21	13
Note: 1 = winter 2006, 2 = spring 2006, 3 = summer 2006, 4 = autumn 2006, 5 = winter 2007 and 6 = spring 2								

3.2. Osmophilic and Osmotolerant fungi from salt crusts collected from Wadi El-Natrun lakes

Twenty-five species related to 11 genera of osmophilic and osmotolerant fungi were recovered on Czapek Dox agar supplemented with 40% sucrose. *Aspergillus* (8 species), *Penicillium* (4), and *Emericella* (3) were the most common genera that came in their counts at 40% sucrose medium (Table 2). The three genera possessed their peaks in salts collected from Rosetta lake but in different seasons. In this respect, Siegel and Siegel's (1979) studies have shown that *Penicillium* and *Aspergillus* were the best genera grown in moist salt crystals such as salt flats or special water-bodies such as the Dead Sea.

Aspergillus (76.0%) of the total CFUs) was the most commonly encountered fungus collected from salt crusts of the 8 lakes by using 40% sucrose medium and control medium (Moubasher et al., 2015; Gouda et al., 2020a, 2020b). The peak of *Aspergillus* was recorded during spring 2006. *A. terreus* (28.6%) and *A. flavus* (32.4%) were recorded from all lakes during almost all seasons. While *A. niger* (9.2%) was recovered from all lakes except Fasida Lake. *A. fumigatus* (4.5%) was isolated in only 6 lakes except for Khadra and Fasida during almost all seasons. The remaining *Aspergillus* species were recorded from only 2 lakes: *A. sydowii* (Khadra during spring 2007 and Fasida during summer 2006), or one lake: *A. candidus* (Al Gaar, winter 2006), *A. japonicas* (Khadra, winter 2007), *A. ochraceus* (Fasida, spring 2006 and winter 2007). Steiman et al. (2004) isolated *Aspergillus fumigatus* and *A. niger* from tufa samples present along Mono Lakeshores, California.

Penicillium (5.5% of the total propagules) was recorded from all lakes except Hamra during almost all study seasons. The peak of *Penicillium* was recorded during autumn 2006. *P. chrysogenum* (3.3%) was recorded from all lakes except El Zugm and Hamra. In comparison, *P. puberulum* (0.8%) was isolated from 3 lakes (Al Beida, Umm Risha, and Fasida during spring, summer, and winter 2006 and 2007). The remaining two *Penicillium* species were isolated from 2 lakes: *P. crustosum* (El Zugm during spring 2007, and Khadra during winter 2006), *P. oxalicum* (Al Beida during spring 2006, and Khadra during summer 2006). In this respect, 3 species of *Penicillium* different from those reported here (*P. citrinum, P. duclauxii,* and *P. waksmanii*) were reported from tufa samples collected from Mono lakes shores, California (Steiman et al., 2004).

Emericella (6.5%) and its dominant species *E. quadrilineata* (6.1%), were recorded from salts collected from 4 lakes. The peak of *Emericella* was recorded during spring 2007. *E. vareicolor* and *E. nidulans* were recorded from one lake Al Beida and Rosetta. *Eurotium* and its dominant species *E. chevalieri* were recovered from 3 lakes (Hamra, Al Beida, and El Zugm). The peak of *Eurotium* was recorded from Al Beida during winter 2007. *E. repens* was isolated in 1 lake (El Zugm during 3 seasons). Shearer (1972) showed that as salinity increased, the rate of Ascomycetes to Fungi imperfect increased, while the number of species collected generally reduction. It was also stated that *Emericella nidulans* is a typical soil fungus isolated from the desert and saline soil (Abdel-Fattah et al., 1977; Moustafa, 1975).

Fusarium (6%) and its dominant species *F. solani* (3.4%), were recorded from all lakes during 2 seasons (spring and winter). The peak of *Fusarium* was recorded from Rosetta during spring 2006. The remaining *Fusarium* species were recorded from one lake: *F. oxysporum* (El Zugm, spring 2007), *F. verticillioides* (Al Gaar, winter 2007), and unidentified *Fusarium* species (Rosetta, spring 2006). *Cladosporium* (*C. cladosporioides*) came behind *Fusarium* infrequency and was recorded from 3 lakes (Hamra, Al Beida, and El Zugm) *Acremonium strictum*, *Botrytis* sp., and *Humicola insolens* were recorded each from one lake during one season only. Steiman et al. (2004) isolated *Acremonium chaticola* from a tufa of Mono lakes shores (California). Also, Cantrell et al. (2006) isolated *Aspergillus* sp., *A. japonicus, Chaetomium globosum, Cladosporium cladosporioides, Penicillium* sp., and *P. variabile*, and many isolates of dark and white sterile mycelia on sediment samples collected from hypersaline environments of solar salterns. Our results show that some species were isolated on medium supplemented with 40% sucrose but not on control medium: *Aspergillus candidus, Emericella variecolor,* and *Fennellia nivea.*

Table 2. Summarized of osmophilic and osmotolerant fungi from salt crusts in different lakes of Wadi El-Natrun

Fungal taxa	Hamra	Al Beida	El Zugum	Rosetta	Umm Reisha	Al Gaar	Khadra	Fasida
Acremonium				4				
A. strictum				4				
Aspergillus	1256	23456	2356	23456	2456	12356	1356	23456
A. candidus						1		
A. flavus	1256	35	23	256	256	1235	35	23
A. fumigatus	6	246	6	3	6	56		
A. japonicus							5	
A. niger	126	26	2	2346	4	3	156	
A. sydowii							6	3
A. terreus	6	24	2356	23456	56	6	356	456
Botrytis sp.	5							
Cladosporium		5	5		2		13	
C. cladosporioides		5	5		2		13	
Emericella	56	23456	23456	23	56			
E. nidulans				56				
E. quadrilineata	56	2456	23	356				
E. variecolor		36						
Eurotium	6	2345	2					
E. chevalieri	6	24	2					
E. repens		235						
Fennellia nivea								25
Fusarium	6	5	56	26	2	5	6	5
F. oxysporum			6					
F. solani	6	5	5	6	2		6	5
F. verticillioides						5		
Fusarium spp.				2				
Humicola							6	
H. grisea							6	
Penicillium		2356	6	46	56	35	13	2
P. chrysogenum		25		46	56	35	1	2
P. duclauxii			6				1	
P. oxalicum		2					3	
P. puberulum		36			5			2
Pencillium spp.		2						
Stachybotrys chartarum					5			
No. of genera (11)	4	6	6	5	6	3	5	3
No. of species (29)	8	13	10	9	9	7	11	7

3.3. Osmophilic and osmotolerant aeromycobiota collected from Wadi El-Natrun lakes

Twenty-nine species related to 18 genera of osmophilic and osmotolerant fungi were recovered on Czapek Dox agar supplemented with 40% sucrose, compared to 27 species to one species variety related to 18 genera recovered on the control medium. *Cladosporium* (4 species, 38.6%), *Alternaria* (333.3%), *Aspergillus* (68.9%), *Penicillium* (21%) and *Fusarium* (1 0.7%) possessed the highest CFUs among fungi on 40% sucrose medium. It is worthy of mentioning that *Eurotium repens* was the only osmophilic fungus encountered from the air. In this respect, Moubasher and Moustafa (1974) found that *Cladosporium* and *Alternaria* to be the most common genera at Assiut, Egypt (Table 3).

In previous studies, the most abundant airborne fungi are *Cladosporium, Alternaria, Epicoccum, Stemphylium, Curvularia, Torula, Aspergillus,* and *Penicillium* (Fischer and Dott, 2003; Ismail et al., 2002; Şimşekli et al., 1999). At the same time, indoor environments, the dominant fungi are species from the genera *Aspergillus, Penicillium,* and *Cladosporium* (Chao, 2002; Tsai et al., 2005). *Cladosporium* and its dominant species *C. cladosporioides* were recorded from the air of 8 lakes during all seasons of study. They were the most commonly encountered fungi among the trapped ones (regarding their count in the air in 6 out of 8 lakes investigated). The peak of *Cladosporium* was recorded during autumn 2006 from Fasida. *C. oxysporum* was found in the air of 4 lakes (El Zugm, Rosetta, Khadra, and Fasida) with a pronounced count in Fasida lake air. *C. herbarum* was recovered from 3 lakes (Hamra, El Zugm, and Umm Risha) during summer, and autumn 2006 while *C. sphaerospermum* was recorded from Fasida lake during autumn 2006. In this respect, *Cladosporium* species were the dominant fungi in indoor air in summer, and *Penicillium,* and *Aspergillus* dominated in winter (Ren et al., 1999). *C. herbarum* was the most common *Cladosporium* species in the air of the Botany Department, Assiut University (Moubasher and Moustafa, 1974).

Alternaria and its dominant species *A. tenuissima* were recorded from all lakes during all seasons of study. It had the highest CFUs in two lakes (Al Beida Lake and Al Gaar Lake). *A. alternata* was recovered from 5 lakes during almost all seasons, while *A. chlamydospora* was recorded from 3 lakes (Al Gaar, Khadra, and Fasida) during winter, autumn 2006, and spring 2007. In agreement with our results, *A. alternata* and *A. tenuissima* were the most common in the air of Assiut University (Moubasher and Moustafa, 1974). Ismail et al. (2002) found that *Alternaria* followed by *Aspergillus* were the most frequent in the atmosphere of Farafra and Bahariya Oasis of Western desert, Egypt. Of *Alternaria*, *A. alternata, A. chlamydospora.* Similarly, *A. alternata* was the most common in the air at Tailf, Saudi Arabia (Abdel-Hafez, 1984). *Aspergillus* was recorded from the air of all lakes except Al Beida during almost all seasons of study. The peak of *Aspergillus* was recorded from Rosetta during winter 2007. *A. niger* (2.5%) was recorded from 5 lakes, while *A. terreus* (3.4%) was the most commonly encountered among Aspergilli. However, it was recovered from 4 lakes (El Zugm, Rosetta, Al Gaar, and Umm Risha) during 4 seasons. *A. falvus* and *A. ochraceus* were recorded from two lakes, while *A. fumigatus* and *A. sydowii* were recorded from one lake during only one season.

Ismail et al. (2002) found that *Aspergillus* to be the most commonly encountered from the air of the Western Desert of Egypt. Of which *Aspergillus flavus, A. fumigatus,* and *A. niger* were highly encountered. Also, Abdel-Hafez (1984) found that the most common genera were *Aspergillus, Alternaria, Cladosporium, Scopulariopsis, Phoma, Drechslera,* and *Penicillium.* Of *Aspergillus, A. sydowii, A. flavus, and A. niger* were the most prevalent at Tailf, Saudi Arabia. *Nigrospora sphaerica* was recorded from all lakes except Al Beida, and Rosetta. On the other hand, *Epicoccum nigrum,* and *Ulocladium botrytis* were recorded from 5 lakes, while *Stachybotrys chartarum* was recorded from 4 lakes (Al Beida, El Zugm, Umm Risha, and Khadra). Other fungal species were recovered from either 3 lakes: *Fusarium solani* (El Zugm, and Fasida during 3 seasons), *Graphium penicillioides* (El Zugm, Rosetta, and Umm Risha during winter and spring 2007), *Scytilidium lignicola* (Hamra, Al Beida, and Khadra during winter, and spring seasons), *Stemphylium botryosum* (Hamra, Al Beida, and Rosetta during winter and spring 2007), 2 lakes: *Acremonium furcatum* (Rosetta, and Umm Risha during winter 2007), *Cochliobolus tuberculatus* (El Zugm, and Rosetta during summer, and autumn 2006), *Penicillium chrysogenum* (Rosetta, and Umm Risha during spring 2006 and winter 2007), or one lake: *Emericella quadrilineata* (Al Gaar, summer 2006), *Emericella* spp. (Rosetta, winter 2006), *Eurotium repens* (Rosetta, winter 2007), *Eurotium* spp. (Rosetta, winter 2007), *Penicillium duclauxii* (Hamra, autumn 2006), *Thermoascus auranticus* (Al Beida, spring 2006, and 2007), and *Trichothecium roseum* (Al Beida, winter, and spring 2007). In this respect, Ismail et al. (2002) isolated species of *Acremonium, Emericella, Epicoccum, Eurotium, Fusarium, Humicola, Scopulariopsis, Scytalidium, Stachybotrys, Ulocladium,* and *Trichothecium* from the air of Western Desert, Egypt. Also, (Moubasher and Moustafa, 1974) reported *Ulocladium, Epicoccum, Fusarium, Scopulariopsis, Stachybotrys, Trichothecium, Humicola, Nigrospora,* and *Stemphylium* in the air of Assiut University. Suchreported species of *Penicillium, Cladosporium,* and Zygomycetes, and *Alternaria* are significantly associated with lower respiratory tract illness (Stark et al., 2003).

Table 3. Summarized of osmophilic and osmotolerant fungi from the air in different lakes of Wadi El-Natrun								
Fungal taxa	Hamra	Al Beida	El Zugum	Rosetta	Umm Reisha	Al Gaar	Khadra	Fasida
Acremonium			5	5	5	5	5	
A. furcatum				5	5			
Acremonium spp.			5			5	5	
Alternaria	26	1256	56	23456	256	1234	15	23456
A. alternata		56		34	26	123456		245
A. chlamydospora						1	1	46
A. tenuissima	26	126	56	356	56	6	5	235
Aspergillus	4		256	23456	345	126	3	4
A. flavus				45	35			
A. fumigatus	4							
A. niger	4		256	45	4		3	
A. ochraceus				25		16		
A. terreus			6	34		26		4
Cladosporium								
C. cladosporioides	123456	156	345	2456	345	3456	1245	2345
C. herbarum	4		4		3			
C. oxysporum			4	5			3	245
C. sphaerospermum								4
Cochliobolus			34	34				
tuberculatus								
Epicoccum nigrum	126		26	236	26		1	
Emericella quadrilineata						3		
Eurotium				5				
E. amstelodami				5				
E. repens				5				
Fusarium solani			6				45	45
Graphium penicillioides			6	6	5			
Nigrospora sphaerica	6		6		246	126	6	6
Penicillium	4			2	5	6		
P. chrysogenum				2	5			
P. duclauxii	4							
Scytilidium lignicola	6	15					26	
Stachybotrys chartarum		15	3		26		3	
Stemphylium botryosum	6	5		6				
Thermoascus auranticus		26						
Trichothecium roseum		56						
Ulocladium botrytis	6		256	6	6			256
No. of genera (18)	9	8	11	10	10	7	9	6
No. of species (29)	11	7	13	16	12	9	10	10

3.4. Halophilic and halotolerant fungi in mud collected from Wadi El-Natrun lakes

Using Czapek Dox agar supplemented with 10% NaCl, only 6 species related to *Aspergillus* (*A. terreus, A. flavus,* and *A. niger*), *Eurotium* (*E. chevelieri*), *Cladosporium* (*C. cladosporioides*), and *Scopulariopsis* (*S. brumptii*) were isolated from mud samples collected during summer and autumn 2006 from Fasida and Al Beida (Table 4). Moubasher et al. (2015) isolated *Aspergillus*, *Penicillium*, *Scopulariopsis*, *Eurotium,* and *Fusarium* from Wadi El Natrun lakes' soil. Aspergilli are one of the most common fungal species and have been recorded regularly from extreme salinity (Molitoris et al., 2000; Kis-Papo et al., 2003). Bronicka et al. (2007) found that too hypersaline environments usually display richness, fungal abundance, and diversity. Wichern et al. (2006) also stated that the soil's hydrogen ion concentration is one of the critical factors that affect the enzymes' activity and, therefore, control microorganisms. The micro fungi more frequent in soils with low alkalinity (pH 7.1 to 8.6), while in high ones, microfungi are more exposed to increasing salt concentration than bacterial populations. This would indicate that microbial activity parameters, such as respiration, N mineralization, and enzyme activity, usually decreased with increasing salinity (Wichern et al., 2020). El-Wahid et al. (1982) found 112 species related to 48 genera in the tidal mud-flats of Kuwait, of which the genera *Aspergillus* (17 species) and *Penicillium* (14) showed the broadest spectrum of species among all genera.

Table 4. Summarized of Halophilic and halotolerant fungi from the air in different lakes of Wadi El –Natrun								
Fungal taxa	Hamra	Al Beida	El Zugum	Rosetta	Umm Reisha	Al Gaar	Khadra	Fasida
Aspergillus		34						34
A. flavus		3						4
A. niger		3						3
A. terreus		34						34
Cladosporium		34						34
C. cladosporioides		34						34
Eurotium		34						34
E. chevelieri		34						34
Scopulariopsis		34						34
S. brumptii		34						
No. of genera		4	0	0	0	0	0	4
No. of species		6	0	0	0	0	0	6
Note: 1 = winter 2006, 2 = spring 2006, 3 = summer 2006, 4 = autumn 2006, 5 = winter 2007 and 6 = spring 2007								

3.5. Halophilic and halotolerant fungi in salt crusts collected from Wadi El-Natrun lake

Using Czapek Dox agar supplemented with 10% NaCl, only *Acremonium strictum* was isolated from salt crusts from Umm Risha Lake during autumn 2006 only. This may be attributed to high salt levels (mainly sodium and chloride) and high alkalinity (pH 8.6-9.8), which is usually not favorable to fungal life and survival. In this respect, Steiman et al. (2004) isolated *Acremonium chaticola* from a mineral form of calcium and carbonate present along Mono Lakeshores (California).

3.6. Halophilic and halotolerant aeromycobiota collected from Wadi El-Natrun lakes

Eighteen species related to 10 genera of halophilic and halotolerant fungi were recovered on Czapek Dox agar with 10% NaCl compared to a higher number of genera (18) and species (27) on the control medium. *Cladosporium* (3 species), *Aspergillus* (5), and *Alternaria* (3) were the most common genera in 10% NaCl medium (Table 5). These genera accounted for 43.6%, 31.5%, and 12.8% of the total fungal catch, respectively. *Fusarium* appeared on the control medium but not on the salted one. Of the preceding genera, only the total fungal catch of *Cladosporium* and *Aspergillus* was higher on the control medium than on the medium supplemented with 10% NaCl. In agreement with the current results, Ismail et al. (2002) found that *Aspergillus,* followed by *Alternaria* was encountered in high frequency from the Western Desert of Egypt's air.

Cladosporium and its dominant *C. cladosporioides* species were trapped from the atmosphere of 4 lakes (El Zugm, Rosetta, Umm Risha, and Fasida) during 4 seasons (summer, autumn 2006, winter 2007, and spring 2006 and 2007). It gave the highest CFUs in the air around Zugm and Fasida lakes. *C. oxysporum* was recorded from the air of 2 lakes only (El Zugm and Rosetta), while *C. herbarum* was recorded from El Zugm during autumn 2006. *Cladosporium* was the most occurrences mycoflora in the outdoor air (Pepeljnjak and Segvic, 2003; Sen and Asan, 2009). Four *Cladosporium* species were encountered (*cladosporioides, C. herbarum, C. oxysporum, and C. sphaerospermum*). The same finding that *Cladosporium* was the dominant organism was reported in indoor environments in Croatia (McGrath et al., 1999), Uganda (Ismail et al., 1999), and Turkey (Sen and Asan, 2009).

Aspergillus was trapped from the air of 5 lakes during summer, autumn and spring*;* however, it came behind *Cladosporium* in its counts. The counts of aspergilli were higher in the salted medium (31.5% of the total) than the control medium (7.1% of the total). The peak of *Aspergillus* was recorded from El Zugm during the summer of 2006. *A. flavus* (9.9%) and *A. niger* (9.5%) were almost the most commonly encountered species in the air around 4 and 5 lakes on 10% NaCl medium, respectively; however, *A. niger* was recovered during 3 seasons while *A. flavus* during summer 2006 only. *A. terreus* (during autumn and spring 2006) and *A. ochraceus* (during summer 2006) were recovered from 2. However, different lakes. *A. sydowii* was recovered from Rosetta during only summer 2006. In this respect, Ismail et al. (2002) found that *Aspergillus* to be the most common, followed by *Alternaria* in the air of the Western Desert of Egypt with *Aspergillus flavus, A. fumigatus,* and *A. niger* being highly encountered. Also, (de Ana et al., 2006) reported that *Aspergillus* was more abundant in the summer outdoor environment. On the other hand, (Sen and Asan, 2009) found that *Aspergillus* was more frequent in August and September outdoor in Tekirdag City (Turkey). Of *Aspergillus, A. fumigatus, A. niger, A. tamari, A. versicolor,* and *A. wentii* were identified.

Alternaria (3 spp.) was recorded from 3 lakes (Rosetta, Umm Risha, and Fasida) during spring 2006 and 2007. It had the highest CFUs in the air around Umm Risha Lake. *A. tenuissima* was recorded from Rosetta and Umm Risha during both spring seasons. *A. alternata* was recorded from Umm Risha and *Alternaria* sp. (from Fasida) each during spring 2006 only. In this respect, *A. alternata* was highly encountered followed by *A. tenuissima*, *A. chlamydospora* and *A. citri* in an oasis of Western desert Ismail et al. (2002). Also, Sen and Asan (2009) found that the spore concentration of *Alternaria* represented by *Alternaria alternata* and *A. citri* reached its maximum peak in April and October outdoors in Tekirdag City (Turkey). The other remaining 7 fungal species were trapped from one lake only: *Cochliobolus tuberculatus, Scytilidium lignicola,* and *Ulocladium botrytis* (from El Zugm during autumn 2006), *Epicoccum nigrum*, *Nigrospora sphaerica, Penicillium chrysogenum* (from Umm Risha during spring), and *Stachybotrys chartarum* (Rosetta during spring 2007). In this respect, Ismail et al. (2002) isolated species of *Epicoccum, Humicola, Scytalidium, Stachybotrys, Ulocladium,* and *Trichothecium* from the air of Western Desert, Egypt.

Table 5. Summarized of halophilic and halotolerant fungi from the air in different lakes of Wadi El-Natrun

Fungal taxa	Hamra	Al Beida	El Zugum	Rosetta	Umm Reisha	Al Gaar	Khadra	Fasida
Alternaria				6	26			6
A. alternata					2			
A. tenuissima				6	26			
Alternaria sp.								6
Aspergillus	3		34	236	3			6
A. flavus	3		3	3	3			
A. fumigatus	3							
A. niger	3		34	26	3			6
A. ochraceus	3			3				
A. sydowii				3				
A. terreus			4		3			
Cladosporium			34	6	26			5
C. cladosporioides			34	6	26			5
C. herbarum			4					
C. oxysporum			4	6				
Cochliobolus tuberculatus			4					
Epicoccum nigrum					26			
Nigrospora sphaerica					6			
Penicillium chrysogenum					6			
Scytilidium lignicola			4					
Stachybotrys chartarum				6				
Ulocladium botrytis			4					
No. of genera	1	0	5	4	6	0	0	2
No. of species	4	0	9	8	9	0	0	3

4. Conclusion

Forty-five species related to 21 genera of osmophilic and osmotolerant fungi were collected from mud, salt crusts, and aerobiota on Czapek Dox agar supplemented with 40% sucrose during the seasons of study. We isolated twenty- two species related to 13 genera of halophilic and halotolerant fungi were collected from mud (Only 6 species related to *Aspergillus* (*A. terreus, A. flavus,* and *A. niger*), *Eurotium* (*E. chevelieri*), *Cladosporium* (*C. cladosporioides*), and *Scopulariopsis* (*S. brumptii*), salt crusts (only *Acremonium strictum*) and aerobiota (*Cladosporium* (3 species), *Aspergillus* (5) and *Alternaria* (3) on Czapek Dox agar supplemented with 10% sodium chloride during the seasons of study. We found that the dark mycelia on aerobiota were the most dominant genera and species.

Conflicts of interest. There are no conflicts of interest.

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