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**Short Communication** 

# Detection of Mycoflora from Marine Fishes of Karachi Fish Harbour and Ormara Region (Pakistan)

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#### ABSTRACT

In the present study we have isolated 5 different species of fungi *Aspergillus niger, Aspergillus flavus, Rhizopus stolonifer, Penicillium* sp. and *Fusarium* sp. from 14 different genera of commercially important marine fishes *Acanthopagrus* sp. *Parastromateus niger, Nemipterus* sp., *Pampus argenteus, Ilisha* sp., *Alepes djedaba, Epinephelus* sp., *Teraponjarbua, Terapon puta, Scomberomorus koreanus, Epinephelus coioides, Lutjanus* sp., *Pomadasys* sp. and *Lutjanus johnii* procured from Kemari fish harbor (Pakistan). Our study shows that fresh fishes were not contaminated by incidence of fungi. *Aspergillus flavus* was the most dominant fungus as compared to other species of fungi but it represents the common mycoflora of this region.

Pathogenic microorganisms mostly attack on frozen sea food during storage. Fungal contamination during storage reduces the quality of food for human consumption due to unpleasant taste, lipolysis and sliminess (Abd El-Fattah, 2002; Hassan and Abdel Dayem, 2004). Different fungi produce mycotoxin and reduce the reproduction of fish in aquaculture (Ali et al., 2011). Mycotoxins in fungi are harmful for human health and in large amount cause many carcinogenic diseases (Hassan et al., 2009). Vandersea and Litaker (2007) isolated six fungi from gills of Channel catfish from which 3 genera belongs to Aphanomyces, Achlya and Phialemonium. Some common fungi such as Aspergillus sp., Penicillium sp. and Absidia sp. were isolated from fish feed (Alinezhad et al., 2011). Fifteen different species of fungi were identified from Coregonus lavaretus holsatus (Czeczuga et al., 2004). Saprolegnia parasitica is the most common fungal species which mostly causes infection in fishes and eggs of Salmon (Ebrahimzadeh et al., 2007). Aspergillus niger is a most common pathogen which causes external and internal infection in fishes and also served as human pathogen (Alinezhad et al., 2011; Firoozbhakhsh et al., 2005; Ebrahimzadeh et al., 2007). Fish served as a better source of high nutritional values due to its amino acids, particularly lysine, vitamins A, D and E, thiamin, riboflavin and niacin (vitamins B<sub>1</sub>, B<sub>2</sub> and B<sub>2</sub>) as well as minerals including iron, calcium, zinc,

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iodine, phosphorus, selenium and fluorine (Sankar, *et al.*, 2013). The present study aims to isolate and identify the fungi of marine fishes from Ormara and Karachi harbor.

## Materials and method

Economically important marine fishes Acanthopagrus sp. Parastromateus niger, Nemipterus sp., Pampus argenteus, Ilisha sp., Alepes djedaba, Epinephelus sp., Teraponjarbua, Terapon puta, Scomberomorus koreanus, Epinephelus coioides, Lutjanus sp., Pomadasys sp. and Lutjanus johnii were collected randomly from Kemari harbour of Pakistan.

Marine fishes of economically important *Sardinella longiceps, Scomberoides commersonniaus, Otolithus ruber, Opisthopterus tardoore, Thryssa vitrirostris* and *Thryssa setirostris* were collected randomly from Ormara station of Pakistan.

Fishes were identified from the FAO published books (Fischer and Bianchi, 1984). Potato Dextrose Agar (PDA) media containing antibiotics was used for the isolation of fungus. After dissection small pieces of muscles were removed and 3 small pieces of muscles placed into culture media petri dishes. Then petri plates were incubated at  $25^{\circ}$ C for 5 days. After growth of fungus small portion of fungi was taken out with the help of needle and put on a slide containing 2 drops of lacto phenol then examined under a compound microscope. Isolated fungi on fish identified with the help of Barnett and Hunter (1998). Statistical analysis was performed using Microsoft excel software. Data are presented as means  $\pm$  standard deviation of means.

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Results and discussion

Only 4 genera of fungi viz., A. niger, Aspergillus flavus, Rhizopus stolonifer, Penicillium sp. and Fusarium sp. were isolated from 14 fishes viz., Acanthopagrus sp. P. niger, Nemipterus sp., P. argenteus, Ilisha sp., A. djedaba, Epinephelus sp., Terapon jarbua, T. puta, S. koreanus, E. coioides, Lutjanus sp., Pomadasys sp. and L. johnii from Kemari harbour which represent that freshly catch fishes less susceptible for fungal contamination. Aspergillus flavus was the most dominant fungus as compared to other species of fungi but it represents the common mycoflora of this region (Table I). Penicillium sp. and Fusarium sp. was recorded in very less percentage as compared to Aspergillus sp. and Rhizopus stolonifer. S. koreanus contain Aspergillus flavus, and Rhizopus stolonifer. Ilisha sp., A. djedaba, Epinephelus sp., E. coioides, Terapon jarbua, T. puta and Lutjanus sp. infected with Aspergillus flavus only. A. niger and Aspergillus flavus isolated from P. argenteus and Pomadasys sp. Fusarium sp. identified from Acanthopagrus sp. whereas Penicillium sp. isolated from L. johnii. Rhizopus stolonifer isolated from three genera of fishes S. koreanus, Nemipterus sp. and L. johnii.

Seven genera and 9 species of fungi were recorded from fishes of Ormara region. The most dominant isolated fungal sp. in the examined fishes was *A. niger*. *A. niger* and *Penicillium* sp. were isolated from *O. ruber*. *A. niger*, *A. fumigatus*, *Rhizopus* sp. and *Rhizoctonia solani* were examined from tissues of *S. commersonniaus*. *A. niger*  and *Fusarium* sp. were recorded from *O. tardoore. A. niger* was observed in *T. vitrirostris. A. niger, A. flavus and Penicillium* were identified from *T. setirostris.* 3 genera of fungi *A. niger, Cladosporium* sp. and *Mucor* sp. were associated with *S. longiceps* (Table II). Our results represent that very less number of fungi isolated from *T. vitrirostris.* 

We isolate 4 genera of fungi from different variety of fishes from Kemari harbour similar studies carried out by Refaiet al. (2010) reported healthy and diseased fish sample contains 2081 fungal species among which Saprolegnia, Scopulariopsis, Aspergillus, Rhizopus, Penicillium, Paecilomyces, Curvularia, Fusarium and Mucor from Oreochromis sp. and Clarias gariepinus. Shahbazian et al. (2010) reported Fusarium oxyparum, F. npoa, penicillium citrinium, Aspergillus treuse, Alternaria sp., Aspergillus clavatus, Cladosporium sp., Helmintosporium sp. and *Mucor* sp. and 5 fungi belongs to saprolegniaceae family from eggs of trout. Eight genera of fungi Aspergillus sp., Mucor sp., Penicillium sp., Acreomonium sp., Alternaria sp., Fusarium solani, Saprolegnia sp. and Cladosporium sp. were isolated from eggs and brood stock of trout fish (Fadaeifard et al., 2011).

In our studies we found that very less number of fungi was isolated from marine fishes whereas similar studies carried out by Siddique *et al.* (2009) but they found that fresh water culturable fishes more susceptible with fungal infection as compared to non culturable fresh water fishes.

Table I: Fungi isolated (%) from fishes of Karachi (Pakistan) fish harbor.

Name of fishes	Name of fungi							
	Aspergillus niger	Aspergillus flavus	Rhizopus stolonifer	Penicillium sp.	<i>Fusarium</i> sp.			
<i>Ilisha</i> sp.	0±0	55.55±1.0	0±0	0±0	0±0			
Alepes djedaba	$0\pm0$	$44.44 \pm 0.57$	$0\pm 0$	$0\pm0$	$0\pm0$			
Epinephelus sp.	$0\pm0$	22.22±1.15	$0\pm0$	$0\pm0$	$0\pm0$			
Scomberomorus oreanus	0±0	33.33±1.0	33.33±1.0	0±0	0±0			
Epinephelus coioides	$0\pm0$	11.11±0.57	$0\pm0$	$0\pm0$	$0\pm0$			
Terapon jarbua	$0\pm0$	44.44±1.15	$0\pm0$	$0\pm0$	$0\pm0$			
Terapon puta	$0\pm0$	44.44±1.52	$0\pm0$	$0\pm0$	$0\pm0$			
Pampus argenteus	11.11±0.57	11.11±0.57	$0\pm0$	$0\pm0$	$0\pm0$			
Parastromateus niger	22.22±1.15	0±0	$0\pm0$	$0\pm0$	$0\pm0$			
Nemipterus sp.	$0\pm0$	0±0	22.22±0.57	$0\pm0$	$0\pm0$			
Acanthopagrus sp.	$0\pm0$	0±0	$0\pm0$	$0\pm0$	11.11±0.57			
Pomadasys sp.	22.22±0.57	11.11±0.57	$0\pm0$	$0\pm0$	$0\pm0$			
Lutjanus johnii	$0\pm0$	0±0	11.11±0.57	11.11±0.57	$0\pm0$			
Lutjanus sp.	0±0	11.11±0.57	0±0	0±0	$0\pm0$			

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Name of fungi	Name of fishes							
	Otolithus ruber	Scomberoides commersonniaus	Opisthopterus tardoore	Thryssa vitrirostris	Thryssa setirostris	Sardinella longiceps		
Aspergillus niger	$55.55 \pm 2.08$	44.44±0.577	$44.44 \pm 0.577$	22.22±1.154	33.33±0.0	11.11±0.577		
A. flavus	0±0	$0\pm0$	0±0	$0\pm0$	11.11±0.577	$0\pm0$		
A. fumigates	$0\pm0$	22.22±1.154	$0\pm0$	$0\pm0$	$0\pm0$	$0\pm0$		
Penicillium sp.	44.44±1.154	$0\pm0$	0±0	0±0	33.33±1.00	$0\pm0$		
Rhizopus stolonifera	$0\pm0$	11.11±0.577	0±0	0±0	$0\pm0$	$0\pm0$		
Fusarium solani	$0\pm0$	$0\pm0$	11.11±0.577	0±0	$0\pm0$	$0\pm0$		
Cladosporium sp.	$0\pm0$	$0\pm0$	0±0	0±0	$0\pm0$	11.11±0.577		
<i>Mucor</i> sp.	0±0	$0\pm0$	0±0	$0\pm0$	$0\pm0$	11.11±0.577		
Rhizoctonia solani	0±0	22.22±0.577	0±0	0±0	0±0	0±0		

 Table II: Fungi isolated from economically important fishes of Ormara region.

Iqbal et al. (2012) isolated Saprolegnia and Achyla from Ctenopharyngodon idella, and Catla catla. Refai et al. (2010) reported that Aspergillus spp., Penicillium spp., Alternaria spp., Blastomyces spp. and Rhizopus spp. are normal mycoflora. Many pathogenic fungi cause diseases such as Saprolegnia sp. cause Saprolegniasis, Aspergillus sp. cause Aspergillosis and Paecilomyces cause Paecilomycosis. Surrounding environment mostly effect the population of marine fungi (Fadaeifard et al., 2011).

We found that Aspergillus sp. was a most dominant fungus in both Kemari harbour and Ormara region as compared to other fungi different scientist also observed that as compared to Penicillium spp., Candida spp., Rhodotorula spp., these fungi Aspergillus sp. was a most dominant fungi (Ammar, 2001; El-Ahl, 2010). Aspergillus flavus was a most dominant fungus isolated from fish feed (Ibrahim, 2000). From different samples of shellfishes, Penicillum corylophilum was isolated from them (Nofiani et al., 2010). Hassan (2003) observed that fishes exposed with fungal contamination during handling, storage and transportation process. Marine environment contain large amount of salt so marine fishes less susceptible for fungal contamination but in culturable fishes contaminated feeds by worker hands and poor water supply play an important role in fungal infection (Hassan and Abdel Dayem, 2004; Hassan et al., 2007).

Isolation of 4 genera of fungi viz., *A. niger, Aspergillus flavus, Rhizopus stolonifer, Penicillium* sp. and *Fusarium* sp. from Kemari harbour comparable to the study of Iqbal *et al.* (2014) isolated four genera of fungi *Aspergillus, Penicillium, Mucor, and Rhizopus* from head, eyes, buccal cavity, operculum, gills, skin of fresh water silver carpfish, *Hypophthalmichthyes molitrix.* Jalilpoor (2006) isolated 4 genera of fungi *Fusarium* sp., *Saprolegnia* sp., *Penicillium* 

sp. and *Mucor* sp. from eggs of *Aspencer percicus*. Infection by *Fusarium* sp. in fresh water fishes is reported to increase in recent years (Shahbazain, 2010). In Iran 16 species of fungi were isolated from infected eggs of rainbow trout *Oncorhynchus mykiss* (Das *et al.*, 2012). Haroon *et al.* (2014) isolated 5 genera of *Aspergillus*, *Rhizopus*, *Mucor*, *Penicillium* and *Alternaria* from freshwater ornamental fishes *Carassius auratus* (L.), *Xiphophorus maculates* and *Poecilia reticulate*.

In the present study we isolate nine species of fungi from six different variety of fishes from Ormara region, similar studies carried out by Hassan et al. (2011) isolated seven genera of fungi and two genera of yeast from different types of fish whereas Alternaria spp., and Penicillium spp. were the most dominant genera as compared to other fungi. Siddique et al. (2009) found that 15 fish species were infected with three genera of fungi viz., Branchiomyces sp., Saprolegnia sp. and Aphanomyces sp. Eli et al., (2011) reported 3 diseases of fungi viz., Saprolegniasis, Branchiomyces and Dermocystidium from culturist fish of Africa. Hashem (2011) isolated 15 species of fungi from African catfish whereas 6 number of fungal species isolated from bony bream. Iqbal and Saleemi (2013) isolated three genera of fungi such as Aspergillus sp., Blastomyces sp., Penicillium sp. and unidentified mycelium from eyes, head, gills, buccal cavity and operculum of Catla catla fish.

In the present study we found that very less percentage of common mycoflora isolated from fish meat of Ormara region and Kemari harbour that cannot disturb the nutritional value of fish.

# Conclusion

The foregoing study has indicated that Kemari harbour and Ormara region is less polluted and very less

percentage of fungi was isolated from this region. The present investigation is a significant work to study the mycological quality of marine fishes of Pakistan coast. It is suggested that further study is require to study the mycological quality of marine fishes of Pakistan from different regions.

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### Statement of conflict of interest

The authors in the paper showed no potential conflicts of interest.

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