# INVESTIGATION OF MYCOBIOTA FROM BOVINE MILK SAMPLES VEND IN MALIR DISTRICT, KARACHI

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### Abstract

Fungi are spoilage organisms in food microbiology since some molds produce mycotoxins and aflatoxin that can cause some severe health concern in humans. In this perspective the present study was designed to determine the mycobiota in fresh bovine milk samples available for residents of Malir District, Karachi, employing culture plate technique. For this task Potato dextrose agar (PDA) and Sabouraud dextrose agar were synthesized for the purpose of the recovery of fungi from the samples. There were seven genera of fungi *Alterneria, Aspergillus, Fussarium, Mucor, Penicillium, Pythium* and *Rhizopus* was isolated from this District of Karachi while, *A. flavus* was recovered as most abundant fungus.

## Introduction

Fungi are considered spoilage organisms in food microbiology. When conditions are not favorable for the growth of bacteria, yeasts and molds will take over and spoil the food items. Fungi secrete digestive enzymes in order to break down complex food sources they can absorb (Srivastava, 2008). The real food-poisoning concern of mold is the possible production of mycotoxins and aflatoxin by some molds which cause hepatic cancer in humans (Hui, 1992; Johny *et al.*, 2012). There are more than hundred fungal spp. which be capable of cause infection in plants and contaminate through production of mycotoxins. Usually a large portion of ruminants diet is plant or fodder and contaminated cattle feed ingested by cows and buffaloes. These mycotoxins especially aflatoxin  $M_1$  and  $M_2$ , which are metabolite of aflatoxin  $B_1$  and  $B_2$ , respectively in milk of cattle feed on contaminated foods pass through animal's origin milk and meat as well and lastly become part of human food. Beside this, occurrence of fungal spp. in bovine milk greatly depends on the geographical locations as well as on the climate conditions. Such factors involvement clearly evident according to the study done by Mikulec *et al.*, (2005), genus *Fussarium* was isolated from raw milk collected from the locality of lowland and hilly-mountainous.

Malir Town is densely colonized near to Malir farms of fruits, vegetables and forage crops. The majority of inhabitants of the Town are lower earnings and pitiable ecology. Their biggest problems are hygiene, sanitation, and the hazards of eviction as well water and sewerage. Rhinitis a disease was recorded in the local people of Malir Extension Colony, Malir Town as the most prevailed epidemic (Rao *et al.*, 2011). However, Gulshan-e-Maymar, Gadap Town is an open area has relatively better human health and environmental situations. While the aerial mycobiota of this location consists of *Aspergillus niger, Aspergillus wentii, Alternaria solani, Drechslera dematioidea* and *Penicillium notatum* reported by Rao *et al.* (2009).

In current work identification of fungal species have been carried out in fresh milk vend in Malir District of Karachi city and estimate the hygienic level.

#### **Materials and Methods**

Random sampling was carried out from 13 different locations accordance with the AOAC (2000) method. Two separate sets of 250 ml of fresh milk in polyethene pouch were taken from shops existed in Malir District. For this task Potato dextrose agar (PDA) and Sabouraud dextrose agar were synthesized for the recovery of fungi from the samples. To attain precision, three replicates of 1ml samples were inoculated in each media plates and incubated at room temperature (30-40° C) for 5-12 days. The colonies growing on plates were identified on the basis of micro and macro morphological features following the standard manuals (Ellis, 1976; Nelson *et al.*, 1983; Domsch *et al.*, 1980; Singh *et al.*, 1991). Cluster analyses, Ward's linkage strategy applied with Euclidean distance organize data into groups based on their similarity. For this purpose, multivariate statistical technique performed employing the Minitab Version 11.

#### **Results and Discussion**

The averages of colony forming units (cfu mL<sup>-1</sup>) of fungal spp. in fresh milk samples gathered from Malir District were clustered as shown in Fig.1. Two distinct groups (A and B) were recognized. Group A found as most populated, included eight samples (1, 2, 4, 5, 6, 7, 8 and 12), characterized by the lowest ranges of *Alterneria solani, Aspergillus flavus, A. niger* and *Fussarium moniliforme* and the higher ranges of *A. terreus, Mucor recemosus*. Except these, *A. candidus, A. fumigatus, M. mucedo* and *Pythium insidosium* found individually. Lastly, group B included five samples (3, 9, 10, 11 and 13) characterized by the lowest ranges of *A. terreus* and *Mucor recemosus*, and the highest ranges of *Alterneria solani, Aspergillus flavus, A. niger* and *Fussarium moniliforme*. In addition, *Fussarium oxysporum, Penicillium italicum, P. notatum* and *Rhizopus stolonifer* showed individual presence in Table 1.



Fig. 1 Dendrogram derived from Ward's cluster analysis of cfu ml<sup>-1</sup> of microbes in fresh milk samples from Malir District, Karachi.

Group	Fungal species	Total fungal counts
А	Alterneria solani	1.0
	A. candidus	0.5-2.5
	A. flavus	0.5-2.5
	A. fumigatus	0.5-1.5
	A. niger	0.5 - 1.0
	A. terreus	0.5-1.5
	Fussarium moniliforme	0.5
	M. mucedo	1.0
	M. recemosus	1.0
	Pythium insidosium	1.5
В	Alterneria solani	0.5-3.5
	A. flavus	1.0-6.5
	A. niger	0.5-2.0
	A. terreus	1.0
	Fussarium moniliforme	1.0-4.5
	Fussarium oxysporum	0.5-1.0
	M. recemosus	0.5
	Penicillium italicum	1.5-2.5
	Penicillium notatum	1.0-3.5
	Rhizopus stolonifer	0.5

 Table 1. Ranges of cfu mL<sup>-1</sup> of fungal species in groups derived from Dendrogram of fresh milk samples from Malir District, Karachi.

The least frequency of occurrence about 07-08 % samples exhibited M. mucedo, Pythium insidosium and Rhizopus stolonifer, 15 % samples showed the existence of Fussarium oxysporum, M. recemosus and Penicillium italicum. 23% samples showed occurrence of Alterneria solani and Fussarium moniliforme. 31 % and 38 % samples showed the presence of P. notatum and A. candidus; A. fumigatus respectively. The presence of A. terreus found in 46% samples whereas A. niger recorded in  $\sim$  62% samples as second most frequent fungi. Finally, A. flavus recorded in 69% samples as top most abundant fungus. Iram et al. (2013) reported that these species possess extraordinary tolerance tendency against municipal and industrial sludge irrigated soil. A. flavus and A. niger strains produce toxins called aflatoxin B2 and Ochratoxin A respectively which are highly cancerous to humans due to its mutagenic ability (Samson et al., 2001; Schuster et al., 2002; Palma et al., 2007). According to the ranges as exposed in Table 1, Alterneria solani and A. candidus found in sample No.11 and 8 respectively; A. flavus, A. fumigatus, A.niger and A. terreus isolated in sample no.3, 6, 11 and 12 respectively; Fussarium moniliforme, Fussarium oxysporum, M. mucedo and M. recemosus showed maximum presence in sample no. 9, 11, 6 and 5 respectively and the other fungal species, Penicillium italicum, P. notatum, Pythium insidosium and Rhizopus stolonifer isolated in sample no. 13, 11, 10 and 3 respectively at the highest level. Our previous investigation recorded seven fungal species (Aspergillus candidus, A. flavus, A. fumigatus, A. niger, A. terreus, Mucor mucedo and Rhizopus stolonifer) in unprocessed or fresh bovine milk while two fungal species (A. flavus and A. niger) in UHT processed milk samples gathered from Gulshan-e-Iqbal Town, Karachi (Perween et al., 2013). Similar study was done by Jordal et al. (1993) and reported the mycobiota of bovine milk in Spain included genera Aspergillus, Fussarium and Geotrichum.

#### Conclusion

The present study revealed that geographical and local unhygienic conditions especially airborne and aquatic fungi contaminate the bovine milk and causes health consequences to consumers. This investigation also indicates the necessity of evaluation of mycotoxins levels in milk and dairy products.

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