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Prevalence of Culturable Airborne Fungi in Fruit Markets of Delhi and Noida, India

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Abstract

Fungi have been identified as the foremost agents responsible for deteriorating fruits kept in storage after harvesting, leading to significant economic losses. The present study involves the qualitative and quantitative assessment of various airborne fungi in the three fruit markets of Delhi and Noida. An aerobiological survey was conducted using gravity settling technique. The frequently isolated fungi were Aspergillus, Alternaria, Cladosporium, Penicillium, Fusarium and Heliminthosporium. The most dominant species of Aspergillus were A. niger, A. flavus and A. fumigatus. Aspergillus, Fusarium, Penicillium and Alternaria were also isolated from rotten fruit samples collected from these markets. The present study identified major airborne fungi associated with post harvest rot in fruit markets. The information may help in efficient control of post-harvest and storage diseases of fruits.

Keywords: Aerobiology, fungi, fruit markets, post harvest spoilage, percent abundance, *Aspergillus*, *Alternaria*.

Introduction

Fruit and vegetable markets are known to contain several species of fungi. Various pathogenic fungi are a dominant component of air which eventually settle on the surface of fruits. They are major spoiling agents responsible for causing post harvest fruit spoilage, leading to significant economic losses[1]. Fruits contain high levels of sugars and nutrients and their low pH values make them vulnerable to fungal decay[2]. Besides, mishandling during harvesting, grading, packaging, transportation and storage are the other contributing factors[3].

It has been estimated that about 20-25% of the harvested fruits are decayed by pathogens during post-harvest handling even in developed countries[4,5]. In developing countries, postharvest losses are often more severe due to inadequate storage and transportation facilities. India accounts for 8% of the total world fruit production (32 million metric tonnes). Over 30% of fruit and vegetable produce is wasted during harvest, grading, packing, transport, marketing and storage[6]. According to a recent newspaper report, annual post-harvest losses in India are over Rs 2000 billion[7]. There is a considerable gap in food production and net availability to consumers. Post harvest loss reduction is essential for increasing food availability and cost reduction[6].

Besides, spoiling fungi are potentially toxigenic or pathogenic. Molds, which are of importance in food because of potential mycotoxin production, include members of the genera *Aspergillus, Trichothecium, Fusarium*[8]. Some potent fungal toxins like aflatoxins, ochratoxinA, patulin have been detected in fruits during storage[9]. Most of these mycotoxins are neurotoxic and carcinogenic for humans. Airborne pathogenic fungi, on the other hand, could cause infections or allergies[10]. Thus, the presence of fungi is a serious health hazard for workers as well as consumers in markets.

Quantification of airborne fungi in these markets is essential to identify major pathogenic fungi causing post harvest diseases of fruits. It is crucial for the post-harvest quality management of a wide range of high value fruit crops. Systematic studies have been reported on the occurrence of airborne fungi in fruit markets and their possible role in post harvest spoilage from different parts of India[11-14]. However, no studies on the incidence of post-harvest disease and airborne fungal spores in fruit markets of Delhi and Noida region have been reported. Hence, the present study was undertaken in the major fruit markets of Delhi and Noida for the identification and quantification of common airborne fungi. Besides, qualitative assessment of fungi associated with post harvest fruit rots was also undertaken.

Material and Methods

The latitudinal and longitudinal locations of National Capital Region of Delhi are 23.38 degree north and 77.13 degree east and boundaries are depicted in figure-1. Air sampling was undertaken during March-June 2012. Three major fruit markets were selected for the present study: i) Shaheen Bagh, Delhi (Market 1), ii) Sector 37, Noida (Market 2) and iii) Sector 18, Noida (Market 3).

Isolation of culturable fungi: Air samples were collected on Czapek Dox Agar (CDA) medium plates (supplemented with streptomycin sulphate- 0.06 g/L) by gravity settling technique. Samples were collected at an interval of 10 days during study

duration from each market. The sampling was done during morning hours (7 am- 11 am) when the market was most active. Six CDA plates were taken to the markets in sterilized containers. In each market, petri plates were exposed in air at 1 m height at multiple places for 10 minutes. After exposure, petri plates were brought back to the laboratory in pre-sterilized bags and kept in the incubator for 5 days at 25 ± 2°C. The fungal colonies observed after incubation were counted. The fungal isolates were sub-cultured on CDA slants and subsequently identified by preparing lactophenol cotton blue mounts. The identification was based on colony morphology and microscopic characteristics[15-19]. *Aspergillus* colonies were identified up to species level[15-19]. The data was reported as percent abundance of each fungi calculated as per the formula[11]:

$$\text{Percent abundance} = \frac{\text{Total number of colonies of any genus or species in all replicates}}{\text{Total number of colonies in all replicates}} \times 100$$

Figure-1
Location Map of Delhi and Noida

Meteorological data (temperature, rainfall and relative humidity) for the specific days of sample collection was obtained from Weather based agro-advisory services, Agrometeorological observatory, Division of Agricultural Physics, Indian Agricultural Research Institute, New Delhi.

Isolation of fungi from rotten fruits: Diseased fruits (mango, papaya, guava and banana) were identified on the basis of visual examination. They were collected in sterile polythene bags from the survey sites.

Approximately 1 g of rotten portion of the fruit was removed with an aseptic knife and suspended in sterile distilled water (10 ml). A suspension was prepared by gentle stirring for 30 minutes followed by centrifugation at 3,000 rpm for 20 minutes. An aliquot of 0.1 ml of the above suspension was cultured on CDA plates in triplicates. The plates were then incubated at 25 ± 2˚C in a BOD incubator for 5 days. The fungal colonies developed were identified up to species level based on their colony morphology and microscopic characteristics (at 400x magnification)[15-19]. The fungi isolated from diseased fruits were purified by sub-culturing on CDA slants and preserved till further use.

Results and Discussion

The quantitative and qualitative analysis revealed that the investigation site (fruit market environment) was contaminated with a variety of conidial fungi. The total viable airborne fungal counts of the three sampling sites are presented in figure-2.

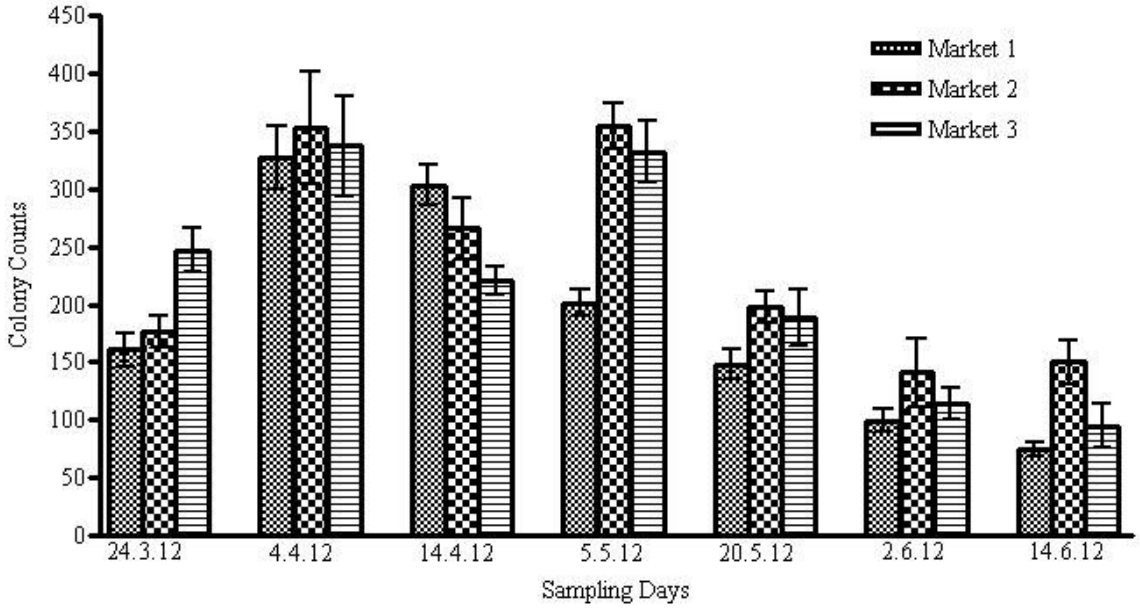

Figure-2
Distribution of total fungal colony counts in three markets between March-June 2012

Day-to-day variations were observed in airborne fungal counts. The high concentration of fungi in the fruit markets (survey sites) indicates the unhygienic conditions in the markets. Various things present in these markets including rotten fruits, vegetables, decaying paper bags, packing materials, straws, discarded leaves and stems etc. act as substrates for saprophytic fungal growth and further spore dispersal in air[8,11]. Total counts, range and percent abundance of various airborne fungi isolated from all the three markets are presented as table-1 and figure-3. *Aspergillus* species were the most dominant components of aeromycota constituting 46% of the total fungal colonies. Other frequently isolated fungi were *Alternaria*, *Cladosporium, Penicillium, Fusarium* and *Heliminthosporium.* However, the concentrations of these fungi varied from market to market. Sterile mycelia also formed a major component of isolated fungi in the three markets (11.9%). *Stemphylium*, *Curvularia* and *Mucor* were not isolated from all the three markets.

Deuteromycetes fungi have been found as most predominant component of aeromycota by various investigators throughout the world using diverse sampling techniques[8,11-14,20-24]. Our results also confirmed this observation. In our survey, *Aspergillus* species were the most common. Chenulu and Thakur (1968) conducted a survey of Delhi markets reported that *Aspergillus niger* and *Rhizopus oryzae* were considered to be responsible to cause major diseases in various fruits in Delhi market[20]. Besides, various other workers have also reported *Aspergillus* as most common airborne fungi of fruit markets from different parts of India[8, 11-14,20,21]. *Aspergillus* colonies were identified up to species level. Percent abundance of various *Aspergillus* species isolated from three markets is presented in figure-4. *A. niger* accounted for 30.58% of total fungal counts. The next dominant species of *Aspergillus* were *A. flavus* followed by *A. fumigatus*. These species were also recorded as common in other markets from different parts of India[8,11-14,21]. *A. nidulans* and *A. versicolor* were not found in the samples isolated from all the three markets. The predominance of *Aspergillus* can be explained by the existence of tropical climate, high temperature and relative humidity along with ample availability of plant debris[22]. Besides, *Aspergillus* conidia can withstand extreme environmental conditions and small spherical spores (3 μm) can be easily disseminated in the air.

Temperature, rainfall and relative humidity affect sporulation and subsequent dispersal of fungal spores in the air. However, no statistical correlation of temperature, precipitation and relative humidity levels with airborne fungi was observed during the sampling duration. It may be due to the fact that our sample size was very small for statistical analysis. Qualitative analysis of data indicated that maximum colonies were obtained in the samples collected in April and first week of May when high humidity (76-81%) and moderate temperatures (31-33˚C) were recorded. These environmental factors may have been significant contributors for abundant mycelial growth and sporulation of fungi. The low fungal counts in the month of June could have been due to high temperature (39.8-42.8˚C) and low humidity (56-65%) which are less favorable for fungal growth. These observations also emphasize that microclimatic conditions in storage godowns and even fruit/vegetable packing boxes (high heat and humidity) allow the growth of fungi causing post harvest decay[10].

Table-1
Prevalence of airborne fungi in three fruit markets of Delhi and Noida, India

Fungi	Total Counts	Range	% Abundance	Present in Markets
A niger	1382	10-133	30.58	All
A flavus	288	6-22	6.37	All
A fumigatus	205	3-54	4.53	All
A nidulans	6	0-3	0.13	1
A versicolor	19	0-4	0.42	1, 2
Aspergillus spp.	204	2-42	4.5	All
Penicillum	153	3-21	3.4	All
Fusarium	129	1-17	2.8	All
Alternaria	864	11-95	19.1	All
Helminthosporium	156	1-18	3.45	All
Epicoccum	41	0-7	0.91	All
Cladosporium	259	10-37	5.73	All
Ulocladium	107	0-17	2.36	All
Sterile Mycelia	537	1-28	11.9	All
Mucor	12	0-4	0.26	2
Stemphylium	71	0-18	15.7	2, 3
Curvularia	70	1-11	1.54	2, 3

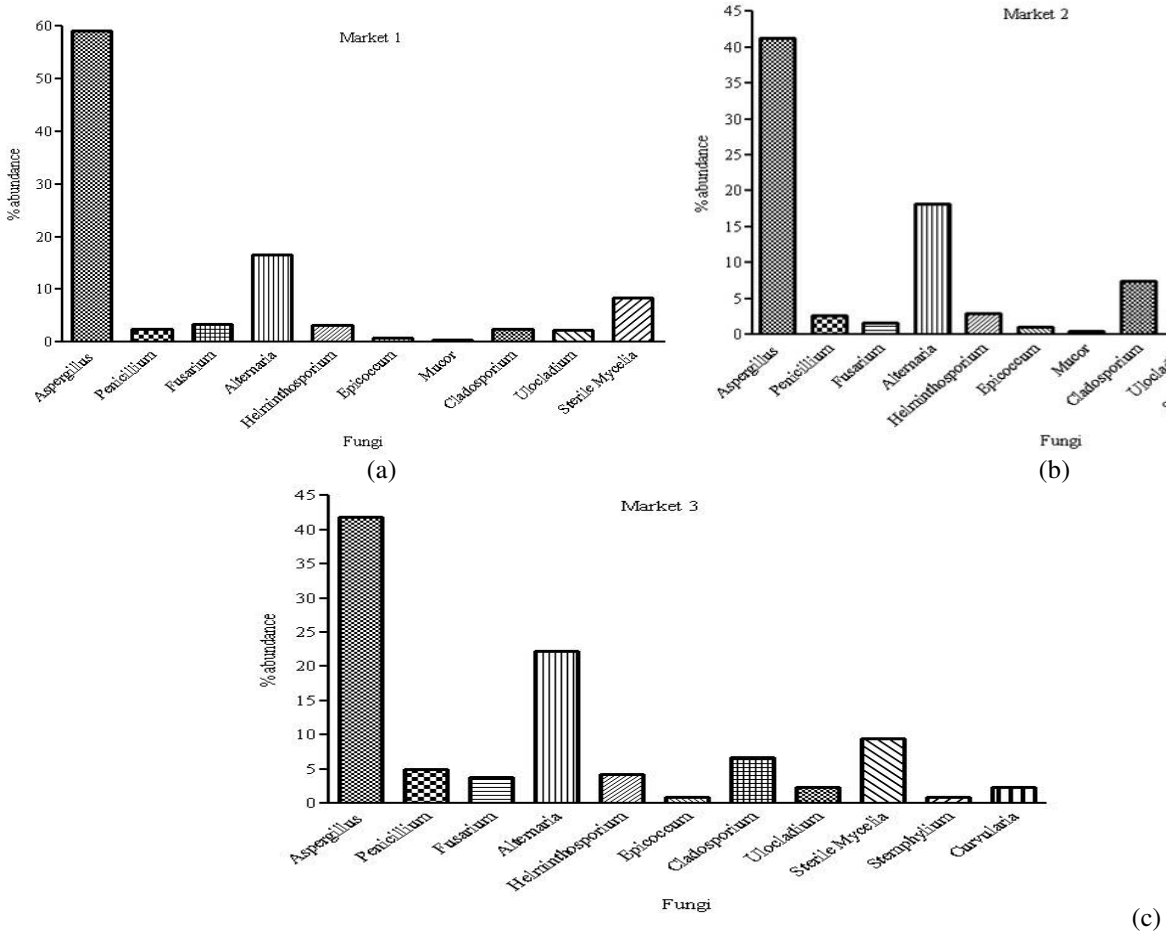

Figure-3
Percent abundance of different fungal genera isolated from three markets

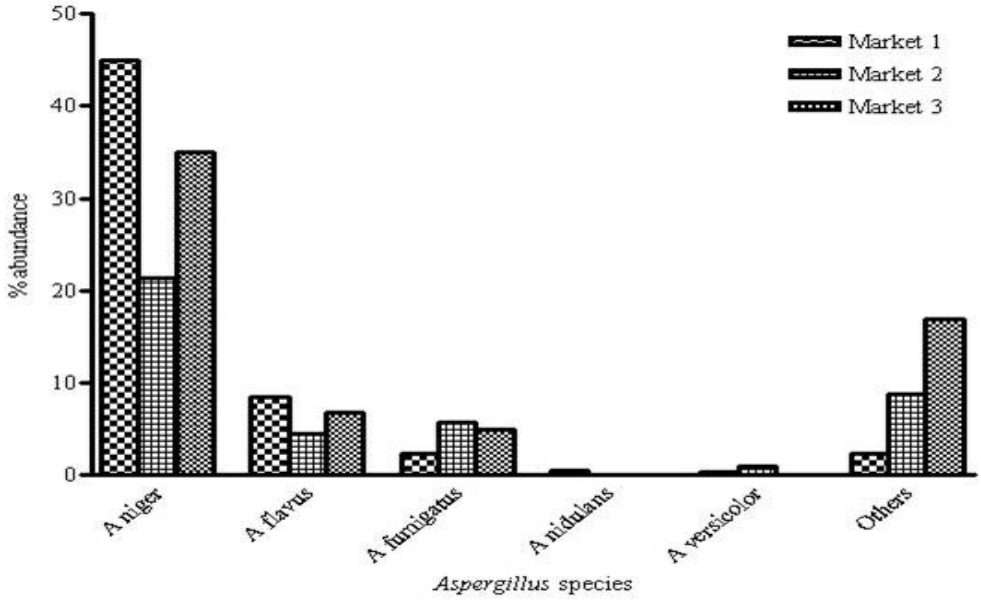

Figure-4
Relative distribution of *Aspergillus* species isolated from three markets

Airborne fungal spores present in these markets may settle on fruit surfaces passively. These spores may germinate due to the influence of favorable weather conditions and cause post harvest diseases before reaching the consumers. The fungi isolated from rotten fruits and vegetables collected from these markets have been listed in table-2. Banana rots showed the presence of *Aspergillus* species. *Penicillium* and *Aspergillus* rots were observed in papaya. *Aspergillus* and *Fusarium* were associated with mango rots. Guava had infection due to *A niger* and *A flavus*. All these spoilage fungi were recovered from air samples collected from the three markets of Delhi and Noida. Hence, there is probably a direct correlation between the prevalence of fungal propagules and spoilage diseases in market environments. It is worth mentioning here that *Aspergillus* species (*A flavus*) are major sources of mycotoxins (e.g. aflatoxins) which are carcinogenic for humans[23].

Table-2
Fungi isolated from rotten fruits and vegetables collected from three markets

S. No.	Fruit/Vegetable	Fungi Isolated
1	Guava (*Psidium guajava*)	*Aspergillus flavus*, *Aspergillus niger*
2	Mango (*Mangifera indica*)	*A niger*, *Fusarium*
3	Papaya (*Carica papaya*)	*A niger*, *Penicillium*
4	Banana (*Musa paradisiaca*)	*A niger*, *Aspergillus* spp.

Conclusion

Our results indicated that *Aspergillus, Penicillium* and *Alternaria* species were the most common components of aeromycota of fruit markets of Delhi and Noida. Besides, these fungi were also associated with postharvest fruit decay. The information of dominant fungi in fruit market environments may help in evolving forecasting system for efficient control of post-harvest and storage diseases of fruits.

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