J.Mycopathol.Res. 64(2) : 435-442, 2026, ISSN : 0971-3719 (Print), 2583-6315 (Online) doi.10.57023/JMycR.64.2.2026.435
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Seasonal variation of fungal bioaerosol over lentic ecosystems in an urban atmosphere

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Received : 21.04.2026 Accepted : 26.05.2026 Published : 29.06.2026

The urban bioaerosol is an understudied topic in developing countries like India. The warm and moist tropical climate of India helps to harbour a wide variety of fungal genera in the ambient air. Previous studies have been carried out in and around Kolkata to capture the urban fungal population which has revealed that fungal diversity varies in different environmental niche in the urban environment. So, a long-term study was carried out to isolate and study the fungal diversity present in the ambient air of South Kolkata near freshwater lentic ecosystems. The study was carried out from October 2023 to September 2025. The fungal isolates were *Aspergillus, Penicillium, Curvularia* and *Colletotrichum*. *Aspergillus* was found to be the dominant fungal genera in the urban ambient air throughout the sampling period. *Penicillium* was abundant in the summer season. *Curvularia* was absent in the winter season and *Colletotrichum* was absent in the summer season. The fungal isolates tested positive for protease and DNase enzyme indicating their host colonization potential. The fungal study of an urban niche is important as it is associated with various respiratory illness like allergy, hypersensitivity, pneumonitis, lung infection and many more. So, a long-term monitoring study of these fungal population is the first step to solve the problem as we do not have bioaerosol repertoire. Monitoring of the aeromycoflora would help us to understand the seasonal variation in the fungal community.

Keywords : Ambient bioaerosol, extracellular enzyme characterisation, fungal characterisation,health impact, urban atmosphere

INTRODUCTION

The aerosol particle of biological origin, present in the ambient atmosphere, are collectively referred to as bioaerosol. The biological components primarily comprise of bacteria, fungus, algae, microalgae, cyanobacteria, viruses and their cellular components like pollens, cellular fragments, endotoxins and many more. These particles remain suspended in the air and might be transported across varying distances. The warm and humid weather condition across the Indian subcontinent leads to the predomination of fungal spores in the ambient air. These spores may survive harsh environmental conditions and dispersed across longer distances. The recent studies have emphasized on the growing importance of ambient bioaerosol in environmental monitoring and public health due to their association with respiratory diseases, lung infection and allergy (Jabeen *et al.* 2023; Kour *et al.*2025).

Bioaerosols sources across urban ambient air have a complex and assorted nature, regulated by anthropogenic activity and environmental conditions. The sources include soil cover, human activities, decaying organic matter and aquatic ecosystem. Urban bioaerosol around freshwater ecosystem like ponds and lakes in the tropical subcontinent have been an understudied topic as relevant from the available literature. Recent studies indicate airborne fungal communities present in both outdoor and indoor environment with reports emphasizing *Aspergillus, Penicillium, Alternaria* and *Cladosporium* as the dominant taxa. These fungal genera play a crucial role in maintaining ecological balance via nutrient cycling

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but their presence in ambient air also raises concern related to public health (Al-Shaarani *et al.* 2024).In recent years, inhalation of fungal spores has been closely associated with various respiratory illness like asthma, lung infection, hypersensitivity pneumonitis, allergic reactions and invasive infections especially in immuno compromised individuals (Memon *et al*. 2024). Some fungal genera produce mycotoxins and other extracellular enzymes like protease and lipase which exert toxic effect when inhaled. Current assessments have exhibited that ambient bioaerosol components contributes in respiratory diseases and their transmission in densely populated urban areas (Xinyuan *et al.* 2024).Regular monitoring of bioaerosol is required in various urban niche across India.

This gap in research is distinctly evident in developing countries like India where rapid urbanisation has led to enhanced population density and environmental degradation resulting in air quality dynamic alteration. Kolkata is a densely populated metropolitan city in eastern India. The tropical climate of India, consisting of a dominant hot and humid weather, helps in fungal proliferation and spore formation which result in increased fungal concentration in the ambient air. Previously, aeromycological studies have been conducted but these studies did not focus on a specific microenvironment or niche. Instead, they tried to capture the urban and suburb ambient air. The studies exhibited high concentration of fungal spores present in the air and almost 30 different taxa have been identified pertaining to allergenic species (Sengupta *et al.* 2023; Chakraborti *et al.* 2012).

On the basis of these considerations, there is an immediate need for localized, microenvironment specific airborne fungal study around urban ecosystems for better understanding of the ambient fungal dynamics. The present study thus focuses on the identification and characterisation of the airborne fungus obtained from the ambient air near freshwater ecosystems in Kolkata using the conventional macroscopic and microscopic techniques. The aim of the study is to explore the ambient niche near urban ponds and the fungal diversity present in these environments. This would help to bridge the gap in urban aeromycology in tropical environment like Kolkata, India.

MATERIAL AND METHODS

Sampling

Sampling was executed in south Kolkata, West Bengal, India (22º51'N and 88º35'E) near urban lentic ecosystems. The sampling site has a number of freshwater ponds within a radius of 500m. The sampling site is located in a densely populated urban area comprising of residential complex, market places, schools, hospitals and railway infrastructures all within a radius of 1km. The location of the sampling area is approximately 390m from Tollygunge metro station and Tollygunge metro crossing and 650m from the Prince Anwar Shah Road crossing (Fig.1). The sampling was carried out using a TISCH six stage Anderson Sampler. According to the Indian Meteorological Department, the city experiences four seasons throughout the year, namely: Winter Season (January and February), Pre-Monsoon Season (March to May), Monsoon Season (June to September) and Post Monsoon Season (October to December). The city primarily experiences a humid climate, dominant for most part of the year.

Isolation of fungal isolates and morphological characterization

Sampling was carried out for two years from October 2023 to September 2025 to study the impact of seasonal variation in the diversity of aeromycoflora over an urban environment located close to freshwater lentic ecosystems. The fungal isolates were sampled using potato dextrose agar media. The plates were incubated at 27ºC for 3-7 days. The fungal colonies were reinoculated on fresh potato dextrose agar plate and incubated under similar condition. The colonies were characterised on the basis of their morphological character and colony colour. The fungal isolates were observed under the microscope to study their spore shape and spore structure along with mycelial growth (Jabeen *et al*.2023).

Enzymatic characterisation

The fungal isolates were tested for protease and DNase production. The fungal isolates were

grown on 2% skim milk agar for protease assay **(**Krikstaponis,2001). The DNase assay was carried out by inoculating the fungal isolate on DNase agar media (Landry, 2014).

RESULTS AND DISCUSSION

Isolation and characterisation of fungal isolates

The sampling of aeromycological population revealed that the ambient air harbours cosmopolitan fungal population. The aeromycoflora exhibited seasonal variation in the urban environment. The fungal colonies were characterised on the basis of their morphological characteristics like colony colour, texture and margins. The colonies were observed under the microscope for morphological characterisation of the spores. The characterisation of the fungal spores indicates the presence of different species of *Aspergillus* in the ambient air. *Aspergillus* and *Penicillium* was found to be the dominant fungal genera in the summer season (Table 1).The samples contained different species of the genus *Curvularia* in the Monsoon Season (Table 2), Post Monsoon (Table 3) and Summer (Table 1) along with *Colletotrichum* which was found in the ambient air during Winter Monsoon Season and Post Monsoon. The post-monsoon and summer sample also exhibited the presence of *Bipolaris* in the aeromycoflora. *Talaromyces* was found in the ambient air during the winter season. The recent study revealed that *Penicillium* and *Aspergillus* have been reported as the most common fungal genera in the Indian subcontinent which are present in the ambient air in all seasons (Jiménez-Uribe *et al.*2026). A study carried out in the suburb of Kolkata exhibited similar result where *Penicillium* and *Aspergillus* was reported as the dominant mycoflora along with *Curvularia* (Das *et al*. 2012). Another study carried out in the Purulia district indicated *Aspergillus, Penicillium, Cladosporium, Fusarium* and *Alternaria* as the dominant fungal genera in the sampling site. In Assam, aeromycoflora study revealed *Aspergillus* as the dominant fungal genera. This indicates that *Aspergillus* is the dominant genera in the eastern side of India which might be as a result of the tropical humid climate experienced by these regions (Das, 2019).

Enzymatic characterisation of the fungal isolates

The fungal isolates were tested for protease production.83.33%of the fungal samples from monsoon tested positive for protea seen zymeand only 16.67% of the fungal samples tested positive for protease in the post monsoon samples (Fig.2).The fungal isolates were also tested for DNase production. 50% of the fungal isolates from winter tested positive for DNase production and the pre-monsoon isolates had the least number of DNase producing fungal isolates (Fig. 3). The airborne fungal mycoflora has been associated with chronic respiratory illness. The presence of *Aspergillus* and *Penicillium* in the ambient air have been linked with asthma exacerbations, allergic bronchopulmonary mycosis, invasive aspergillosis and hypersensitivity pneumonitis. However, the extent of the infection depends on various factors. Immunocompromised individuals along with elderly people and children tends to be affected more than normal individuals. Medical history of the exposed individuals along with the time period of exposure also leads to variation in the impact of the airborne fungal spores (Kour *et al.* 2025). The presence of *Talaromyces* in warm and humid climate of South Asia led to inflammatory lung damage in immuno compromised people (Liam *et al*., 2025). Airborne mycoflora of *Curvularia* and *Colletotrichum* have been isolated as a part of the cosmopolitan fungal diversity in previous studies. They have been identified as potent plant pathogen. They are the causal organism of leaf spot and seedling rot (Lumyong, 2019). The presence of these plant pathogens in urban environment indicates the generation of bioaerosol from leaf and plant surface from the surrounding areas.

CONCLUSION

The aeromycoflora of the ambient air in the urban environment comprise of various fungal genera. They are responsible for animal and plant diseases. The air quality of the ambient air near urban lentic ecosystems is important as they have been associated with various skin, ocular and respiratory infections, to people exposed for a long time or immunocompromised people. The long-

Table 1: Characterisation of fungal isolates from Summer Season

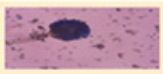
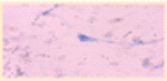
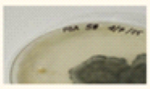
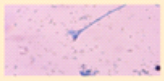
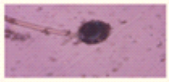
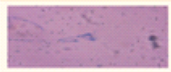
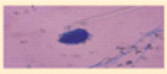
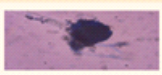
Season	Sample Number	Colony Picture	Colony Morphology	Staining Picture	Conidiophore Morphology	Probable Genus
SUMMER	S1		Colonies consist of a compact white basal felt covered by a dense layer of black conidial heads Colonies are velvety, flat and granular with radial grooves Curled margin		Conidiophores are globose to sub globose with roughened wall. Vesicles are spherical to sub-spherical Conidial heads are phialides borne on metulae.	Aspergillus spp.
	S4		Irregular, raised centrally and wrinkled colony. Feathery to cottony white to pale grey aerial mycelium surrounded by sparse superficial olivaceous dark mycelium, followed by white to pale grey mycelium at the margin		Conidia straight oblong, ellipsoidal, club shaped, rounded at the ends 3–5 distoseptate and transparent Light brown to dark brown walls with yellow ochre cells	Bipolaris spp.
	S5		Colonies consist of a compact brown basal felt covered by a dense layer of dark-brown to black conidial heads Colonies are velvety, flat and granular.		Conidiophores are globose to sub globose with roughened wall. Vesicles are spherical to sub-spherical, or pyriform to spatulate. Conidial heads are phialides borne on metulae.	Aspergillus spp.
	S6		Colony shaped circular and flat Fimbriated margin Black at the centre to grey olivaceous to white towards the edge		Conidia straight or curved 2-3 distoseptate, transparent apical cell Swollen median cell Umber to light brown in colour	Curvularia spp.
SUMMER	S7		Colony shaped circular and umbonated at centre Feathery to cottony white floccose at the centre, surrounded by dense dark green granular conidial heads White Fimbriated margin.		Conidiophores are biverticillate, containing 3-4 cylindrical phialides Conidia were globose to sub-globose and smooth-walled	*Penicillium* spp.
	S8		Colony shaped irregular, raised centrally and wrinkled Feathery to cottony white floccose at the centre, surrounded by sparse dark mycelium.		Conidia straight oblong, ellipsoidal, club shaped, rounded at the ends 3–5 distoseptate and transparent Light brown to dark brown walls with yellow ochre cells	*Bipolaris* spp.
	S9		Colony shaped circular and umbonated at centre Feathery to cottony white floccose at the centre, surrounded by dense dark green conidial heads White Fimbriated margin		Conidiophores are biverticillate, containing 3-4 cylindrical phialides Conidia were globose to sub-globose and smooth-walled stipes smooth walled	*Penicillium* spp.
	S10		Colonies consist of dark brown conidial heads Colonies are velvety, flat and granular		Conidiophores are globose to sub globose with roughened wall. Vesicles are spherical to sub-spherical, or pyriform to spatulate. Conidial heads are phialides borne on metulae	*Aspergillus* spp.
SUMMER	S11		Colonies are circular, velvety, flat and granular Colonies are cottony white at first and then matures into dense dark green conidial heads		Conidiophores are monoverticillate, containing 3-4 cylindrical phialides Conidia were globose to sub-globose and smooth-walled	*Penicillium* spp.
	S13		Colonies are circular, velvety, flat and granular Colonies are cottony white at first and then matures into dense dark green to olivaceous conidial heads White Fimbriated margin.		Conidiophores monoverticillate to sometimes biverticillate, containing 3-4 cylindrical phialides Conidia were globose to sub-globose and smooth-walled	*Penicillium* spp.
	S14		Colonies consist of a compact white basal felt covered by a dense layer of black conidial heads Colonies are flat and granular		Conidiophores are globose to sub globose with roughened wall. Vesicles are spherical to sub-spherical, or pyriform to spatulate. Conidial heads are phialides borne on metulae.	*Aspergillus* spp.
	S16		Colonies consist of a compact yellow basal felt covered by a dense layer of black conidial heads Colonies are flat and granular		Conidiophores are globose to sub globose with roughened wall. Vesicles are spherical to sub-spherical, or pyriform to spatulate. Conidial heads are phialides borne on metulae.	*Aspergillus* spp.

Table 2 : Characterisation of fungal isolates from Monsoon Season

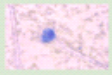
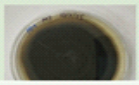
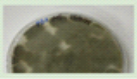
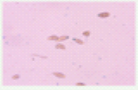
Season	Sample Number	Colony	Colony Morphology	Staining	Conidiophore Morphology	Probable identification
MONSOON	M1		Colonies are velvety, granular, flat Yellow at first but quickly becoming bright to dark yellow-green with age.		Conidiophores are globose to sub globose with roughened wall. Vesicles are spherical Conidial heads are phialides borne on metulae.	*Aspergillus* spp.
	M2		Colonies are fluffy and cottony Dark grey Aerial mycelium. White fimbriated margin		Aseptate hyphae with sporangiophore having oval vesicles surrounded which bear finger-like merosporangia directly over their entire surface (Daisy head appearance)	*Syncephalastrum* spp.
	M3		Colony shaped circular and flat Floccose at the centre Black at the centre to grey olivaceous towards the edge.		Conidia straight or curved 2-3 distoseptate, transparent apical cell Swollen median cell with distinctly Y-shaped conidia. Umber to rust coloured	*Curvularia* spp.
	M4		Circular and flat shaped colony Creamy yellow to dull yellow towards the centre, grey to dark grey towards the periphery		Most of the conidia shaped like a sickle, with a few straight ones, short length, blunt rounded ends, transparent, 1–3 septate	*Colletotrichum* spp.
	M5		Colonies are velvety, granular, flat, often with radial grooves. Yellow at first but quickly becoming bright to dark yellow-green with age.		Conidiophores are globose to sub globose Vesicles are spherical to sub-spherical, or pyriform to spatulate. Conidial heads are phialides borne on metulae.	*Aspergillus* spp.
	M6		Colony raised centrally and wrinkled Feathery to cottony white to pale grey aerial mycelium surrounded by sparse superficial olivaceous dark mycelium		Conidia straight or curved 2-3 distoseptate, transparent apical cell Swollen median cell with distinctly Y-shaped conidia Umber to rust coloured	*Curvularia* spp.

Table 3 : Characterisation of fungal isolates from Post Monsoon Season

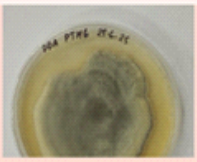
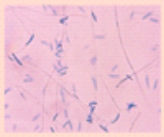
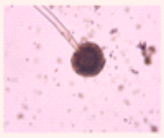
Season	Sample Number	Colony Picture	Colony Morphology	Staining Picture	Conidiophore Morphology	Probable Genus
Post-Monsoon	PTM1		Colony shape irregular, raised centrally and wrinkled Feathery to cottony white floccose at the centre, surrounded by sparse dark mycelium.		Conidia straight oblong, ellipsoidal, club shaped, rounded at the ends 3–5 distoseptate and transparent Light brown to dark brown walls with yellow ochre cells	*Bipolaris* spp.
	PTM5		Colony shaped circular and flat Funiculose surface Fimbriated margin Floccose at the centre Black at the centre to grey olivaceous towards the edge		Conidia straight or curved 2-3 distoseptate, transparent apical cell Swollen median cell with distinctly Y-shaped conidia Umber to rust coloured	*Curvularia* spp.
	PTM6		Colony shaped irregular and flat.Feathery to cottony white to pale grey aerial mycelium at the centre surrounded by dull yellow mycelium, followed by white to dark grey superficial mycelium at the margin. Dark concentric zonation		Most of the conidia shaped like a sickle, with a few straight ones, short length, blunt rounded ends, smooth walled to slightly rough, transparent, 1–3 septate	*Colletotrichum* spp.
	PTM7		Colonies consist of black conidial heads Colonies are velvety, flat and granular		Conidiophores are globose to sub globose with roughened wall. Vesicles are spherical to sub-spherical, or pyriform to spatulate. Conidial heads are phialides borne on metulae.	*Aspergillus* spp.

Table 4 : Characterisation of fungal isolates from Winter Season

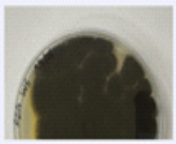
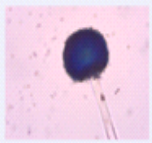
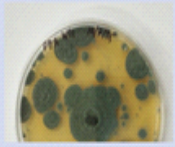
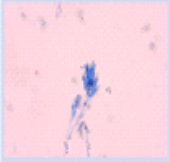
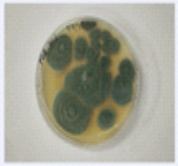
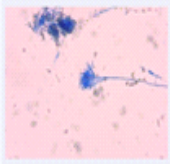
Season	Sample Number	Colony Picture	Colony Morphology	Staining Picture	Conidiophore Morphology	Probable genus
WINTER	W1		Cottony white dense mycelia. The reverse colour is creamy yellow to dull yellow towards the centre		Most of the conidia shaped like a sickle, with a few straight ones, longer in length, tapering towards both ends, smooth walled, transparent, 1–5 septate	*Colletotrichum* spp.
	W5		Colonies covered by a dense layer of dark-brown to black conidial heads Colonies are granular Fimbriated margin		Conidiophores are globose to sub globose with roughened wall. Vesicles are spherical to sub-spherical, or pyriform to spatulate.	*Aspergillus* spp.
	W11		Circular, velvety, flat and wrinkled colony covered by a dense layer of green conidial heads The colony grows as concentric circles from the centre		Conidiophores triverticillate to quaterverticillate Conidia were globose to sub-globose Phialides acerose, i.e., needle shaped	*Talaromyces* spp.
	W12		Circular, velvety, flat and granular colony. Feathery white floccose at the centre, surrounded by dense dark green conidial heads White Fimbriated margin		Conidiophores triverticillate to quaterverticillate Conidia were globose to sub-globose Phialides are cylindrical to acerose, i.e., needle shaped	*Talaromyces* spp.

Fig 1: The sampling site located near urban lentic ecosystem in south Kolkata.

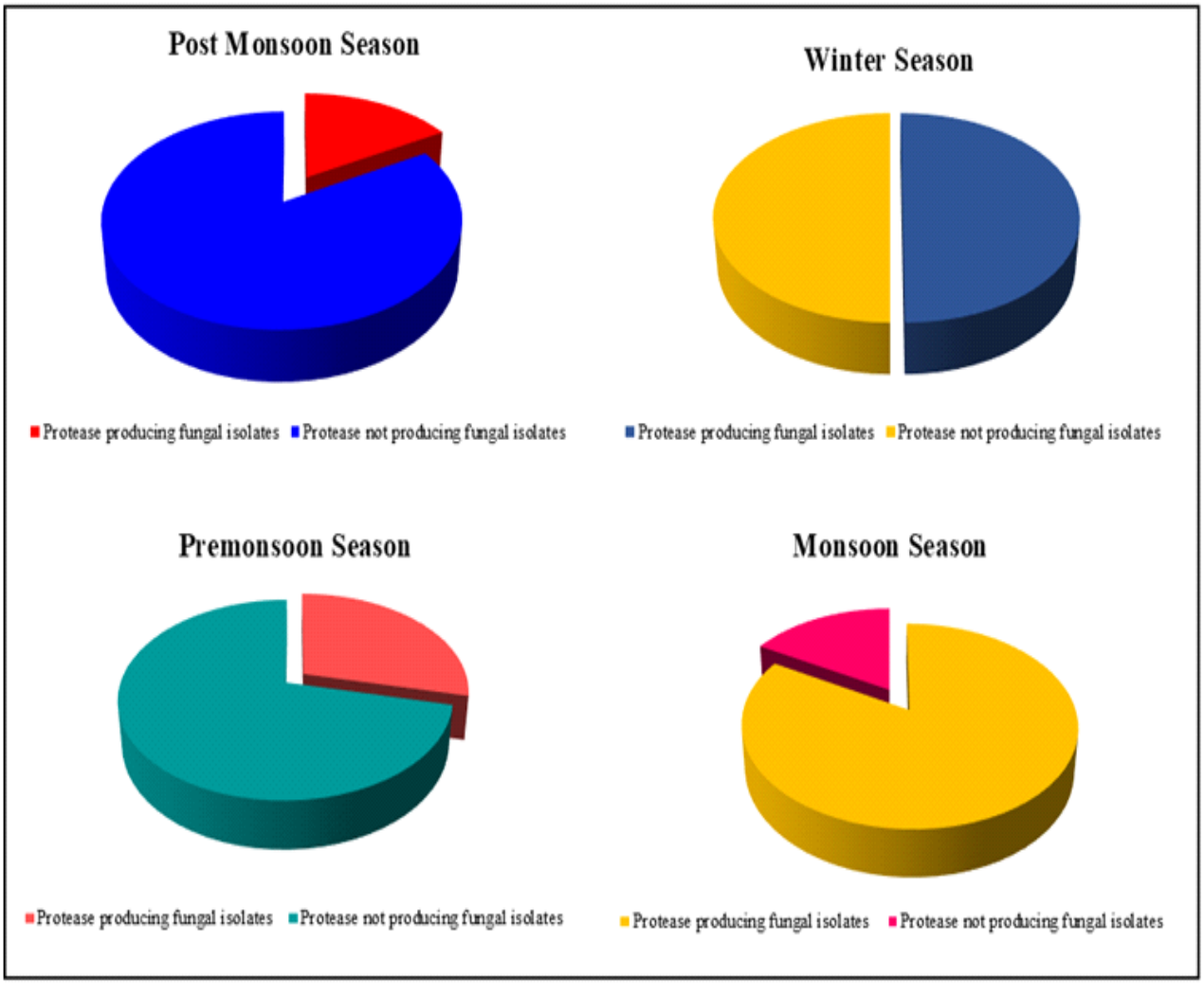


Fig 2: Seasonal variation of percentage of fungal isolates, from ambient bioaerosol ecosystem, producing protease.

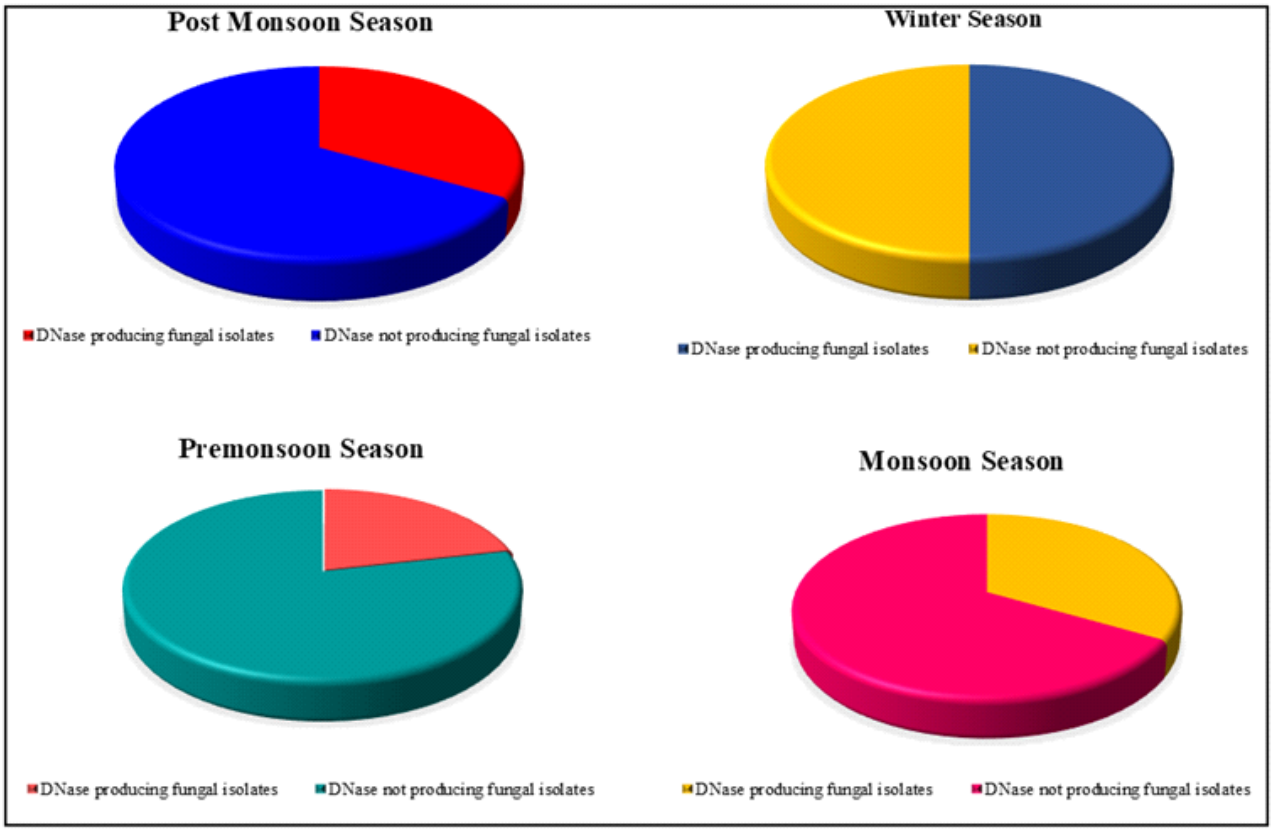


Fig 3: Seasonal variation of percentage of fungal isolates, from ambient bioaerosol ecosystem, producing DNase.

term monitoring of the airborne fungus in urban environment is necessary for the understanding of the impact of the urban fungal diversity on the environment and human health. It would help us to control the pathogens in the long run. A further in-depth study of the fungal isolates is important for identifying the pathogenic and non-pathogenic strains and their health hazards.

ACKNOWLEDGEMENT

The authors would like to acknowledge the Principal of St Xavier's College (Autonomous), Kolkata for providing the opportunity to carry out our research work. Authors also acknowledge Intramural Research Grant of St Xavier's College (Autonomous), Kolkata for supporting this research work.

DECLARATION

Authors declare no conflict of interest.

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