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Assessing Airborne Bacteriological Concentrations in Indoor Environments

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Abstract

Understanding the diversity and abundance of bacteria present in indoor air environments is essential for assessing potential health risks associated with microbial exposure. The aim of this study was to evaluate the indoor air bacterial load, focusing on the determination of genera or species composition and the quantification of colony forming units (CFUs) in different Sections i.e from Biology POCSO Lab., Biology exhibit examination Lab., Serology exhibit examination Lab., DNA exhibit examination Lab. and Toxicology exhibit examination Lab. of 4th floor , Forensic Science Laboratory, Bihar, Patna. The evaluation of indoor air bacterial concentrations reveals significant variations across laboratory spaces, with the highest count recorded in the Toxicology exhibit examination Lab.(T) while the lowest count was found in the Biology POCSO Lab. (P). Mean bacterial concentrations in the Biology exhibit examination Lab.(B), Serology exhibit examination Lab.(S), and Toxicology exhibit examination Lab.(T) exceeded WHO guidelines, contrasting with levels within acceptable ranges in the POCSO exhibit examination Lab.(P) and DNA exhibit examination Lab.(D). Bacterial identification unveiled different genera, predominantly Gram-positive, with diverse morphologies and species including Bacilli, Streptococcus pneumoniae, Staphylococcus aureus and Coagulase Negative Staphylococcus. These findings underscore the importance of stringent safety measures including enhanced sanitation protocols, adherence to PPE guidelines and targeted interventions to regulate indoor air quality, thereby fostering healthier and safer laboratory environments for personnel.

Keywords: Microbes, Bacterial contamination and Indoor air.

Introduction-

Bioaerosols, the biological contaminants suspended in the air, pose a significant concern as they can be ingested or inhaled by humans (1). Living microorganisms can be found everywhere, including suspended in the indoor air, microbes are usually distributed unevenly (2). Microbial air monitoring involves sampling and analysing air for microbial contamination, crucial for maintaining indoor air quality. Sources of indoors microbes encompass humans, ventilation, air-conditioning systems, dust, and outdoor air. Bacteria and fungi constitute the primary constituents of bioaerosols, and exposure to these organisms in built environments correlates with adverse health outcomes (3). Indoor air quality is a critical aspect of human health and well-being, particularly given the amount of time individuals spend indoors. Among these factors, the microbial composition of indoor air plays a significant role in determining overall air quality and potential health risks to occupants. Numerous studies have highlighted the importance of indoor air quality in relation to respiratory health, allergies, and other

health outcomes(4).The World Health Organization has declared that 4.3 million premature deaths are related to indoor air pollution(5).Bacteria, as ubiquitous microorganisms, can colonize indoor environments and contribute to indoor air pollution(6).Identifying the types and concentrations of bacteria present in indoor air is crucial for implementing effective strategies to mitigate exposure risks and improve indoor air quality standards(7).By examining the bacterial composition and abundance in indoor air environments, this study aims to contribute to the existing body of knowledge on indoor air quality and microbial ecology. The findings of this research can inform public health initiatives, building design and maintenance practices, and indoor air quality guidelines, ultimately promoting healthier indoor environments for occupants.

Aim of the work-

Aim of the work was to evaluate the bacterial concentration level of the indoor air, to determine the genera or species composition and the number of colony forming units of airborne microbes (bacteria) in different Sections i.e from Biology POCSO Lab., Biology exhibit examination Lab., Serology exhibit examination Lab., DNA exhibit examination Lab. and Toxicology exhibit examination Lab. of 4th floor, Forensic Science Laboratory, Bihar, Patna from October to November 2023.

Material and Method-

Over the course of one month (4 weeks),20 indoor air samples were collected. These samples were taken once a week specifically in the afternoon time from 5 different section i.e from Biology POCSO Lab.(marked as P), Biology exhibit examination Lab.(marked as B), Serology exhibit examination Lab.(marked as S), DNA exhibit examination Lab.(marked as D) and Toxicology exhibit examination Lab.(marked as T) of 4th floor, Forensic Science Laboratory, Bihar, Patna. Petri dish filled with nutrient agar (NA) was used as sampling surface for bacteria. The Petri plates were exposed to the air in a processing room according to the 1/1/1 scheme (for 1 hour, 1 meter above the floor and about 1 meter away from walls) (8). These plates were incubated at 37^{0}C for 24 hours and checked for bacterial growth. Then colonies were checked, counted and expressed in colony forming units (cfu/m^3) by using Omelyansky's formula(9).The pure colonies were picked by a sterile loop to make a suspension with normal saline on a glass slide and were followed by gram staining to check for gram positive and gram negative bacterial identification(10).Biochemical tests like catalase and coagulase were done for further classification and to identify bacterial group.

Result –

The findings obtained from evaluation of the indoor air bacterial concentrations reveals that, highest indoor bacterial count was found in the Toxicology exhibit examination Lab.(T) which was 2115CFU/m^3 and the lowest count was found in the Biology POCSO Lab.(P) which was 390 CFU/m^3 (Table-1).

Table 1. Number of bacterial CFU/m^3 air and mean bacterial concentrations from different section –

SECTIONS (Sample Collection)	READING -1	READING -2	READING -3	READING -4	Mean
BIOLOGY POCSO Lab.(P)	930	700	890	390	727.5

BIOLOGY Exhibit Examination Lab.(B)	1100	850	1750	970	1167.5
DNA Exhibit Examination Lab.(D)	1130	490	1125	450	798.75
SEROLOGY Exhibit Examination Lab.(S)	1220	850	1200	780	1012.5
TOXICOLOGY Exhibit Examination Lab.(T)	2115	910	2010	950	1496.25

The mean bacterial concentrations in the Biology exhibit examination Lab.(B), Serology exhibit examination Lab.(S) and Toxicology exhibit examination Lab.(T)exceeded WHO guidelines while mean bacterial concentrations in the Biology POCSO Lab.(P) and DNA exhibit examination Lab.(D)were all within the WHO guide lines range (11). Out of 20 samples in this study, 15(75.0%) samples had high, 2 (10.0%) samples had very high and 3(15.0%) samples had intermediate indoor air bacterial load. A total of three bacterial genera were isolated. All were Gram positive and showing different colony morphology and shapes (Table-2). Both coccus and rod forms cells were seen. The coccus were occurred in pairs, chains or clusters. Gram-positive spore-bearing Bacilli identified from sample marked B and S while from sample marked P Gram-positive Cocci in pairs and chain were identified. In sample marked D and T Gram-positive spore-bearing Bacilli as well as Gram-positive Cocci were also identified. Sample marked P identified as Streptococcus pneumoniae, sample marked D were identified as Coagulase Positive Staphylococcus aureus whereas sample marked T were also identified as Coagulase Negative Staphylococcus. The majority of isolated bacteria were Bacillus species, accounting for 80% of the total isolates. Additionally, smaller proportions of Streptococcus pneumoniae, Staphylococcus aureus, and Coagulase Negative Staphylococcus were also found, each comprising 20% of the isolates.

Table 2: Bacterial species detected from different section –

SECTION(Sample Collection)	Gram's Staining	Biochemical Test	Bacterial species
BIOLOGY POCSO Lab.(P)	Gram-positive Cocci in pairs and chain	Catalase Negative	Streptococcus pneumoniae
BIOLOGY Exhibit Examination Lab.(B)	Gram-positive spore-bearing Bacilli	Catalase Positive	Bacillus Species
DNA Exhibit Examination Lab.(D)	Gram-positive spore-bearing Bacilli	Catalase Positive	Bacillus Species
	Gram-positive Cocci in clusters	Catalase Positive, Coagulase Positive	Staphylococcus aureus
SEROLOGY Exhibit Examination Lab.(S)	Gram-positive spore-bearing Bacilli	Catalase Positive	Bacillus Species
TOXICOLOGY Exhibit Examination Lab.(T)	Gram-positive spore-bearing Bacilli	Catalase Positive	Bacillus Species
	Gram-positive Cocci in clusters	Catalase Positive, Coagulase Negative	Staphylococcus

Discussion-

Assessing the microbiological quality of indoor air stands as a critical investigation for identifying microbial pollution indoors. Airborne microbes are one of the main contaminants that play a role as an indicator of clean indoor air (12). Understanding the concentrations of airborne microbes is essential for gauging health risks and establishing standards for controlling indoor air quality. The study demonstrates considerable diversity in indoor air bacterial concentrations among various laboratory spaces. The bacterial load in the indoor environment of 5 different section of 4th floor, laboratory rooms of FSL, Patna ranged between 390 to 2115 CFU/m^3.Notably, the Toxicology exhibit examination Lab.(T) exhibited the highest bacterial count, while the Biology POCSO Lab.(P) recorded the lowest. Such variations might be attributed to differences in laboratory activities, equipment usage, ventilation systems, and human traffic. Visceral contamination could be the primary factor contributing high bacterial load within the Toxicology exhibit examination Lab.(T). So, the assessment of visceral contamination is essential for maintaining hygiene standards and ensuring the safety of individuals working in these environments. The reduced bacterial presence in the Biology POCSO Lab.(P) may stem from its primary utilization for centrifugation and microscopic examinations of exhibits, rather than for exhibit storage, opening, and other examinations. The study indicates that mean bacterial concentrations in the Biology exhibit examination Lab.(B), Serology exhibit examination Lab.(S), and Toxicology exhibit examination Lab.(T)exceeded WHO guidelines, while those in the Biology POCSO Lab.(P) and DNA exhibit examination Lab.(D). remained within acceptable ranges. The structural design and the low number of occupants per area might be responsible for low bacteria burden in both of these Section. Similar studies revealed that, the presence of aerial bacteria was associated to the presence of personnel into the air of the partially closed premises(13,14).Total volume of airborne microorganisms in an enclosed area also depends on the location, weather, structural design, relative humidity, ventilation rate, air movement rate and the number of users of that room (15,16).Hence, a simple approach to improve the indoor air quality within the building would be to prevent overcrowding and incorporate efficient ventilation systems into the design. While there isn't a universally accepted standard for assessing microbial levels in indoor air, research conducted by the World Health Organization (WHO) expert group proposed that the total microbial load should ideally not exceed 1000 CFU/m^3. On the other hand, the European Commission's sanitary standard suggests that microbial levels of 50 CFU/m^3 are considered 'very low', 100 CFU/m^3 are deemed 'low', and levels ranging from 200 to 500 CFU/m^3 are categorized as 'high', with anything above 2000 CFU/m^3 regarded as 'very high load'. According to these standards, the microbial load observed in this study would be classified as 'high' (17,18). A total of three bacterial genera were isolated which are Bacillus species, Staphylococcus aureus and Streptococcus pneumoniae. All of the bacterial isolates were found to be Gram-positive. Staphylococcus is commonly present in all individuals and is typically expelled from the respiratory tract when breathing or speaking. This expulsion from the nose and mouth can lead to its presence in the environment. Staphylococcus has the potential to cause infections such as bacteremia and gastrointestinal infections (19). Staphylococcus aureus is implicated in various prevalent diseases affecting different parts of the body. It can result in infections of the skin and soft tissues (20). Coagulase-negative Staphylococcus inhabits the skin and mucous membranes of both humans and animals, establishing colonization in these areas. (21). Bacillus species are bacteria that are often found in cultures but seldom result in acute infections. However, they can still cause various health issues (22). Bacillus species are persistent and resistant in the environment because of the formation of spores.

Spores have since been recognized as the hardest known form of life on earth(23).The production of spores enables this organism to withstand unfavorable conditions such as low temperatures or heat and may improve the chances of Bacillus to be present in high numbers in the air(24).Streptococcus pneumoniae is a well-known pathogen responsible for various respiratory infections (25).In this study the most frequent isolated genera were Bacillus species (80%) .The present finding were found similar with those of Kavita and Jyoti (2013) (26,27,28).These genera of bacteria have been shown to be amongst the most common bacterial genera often isolated from indoor environments. Mandal and Brandl, in a review article, identified these bacterial genera as frequently encountered in indoor environments, indicating their prevalence in such habitats (29). The presence of higher bacterial counts in certain lab rooms could pose potential health risks to laboratory personnel and compromise experimental integrity. The identification of three bacterial genera, all Gram-positive, underscores the microbial complexity inherent within laboratory environments. The presence of both coccus and rod forms, alongside varied colony morphologies, highlights the diverse microbial ecology within indoor air samples. Moreover, the identification of specific bacterial species, including Bacilli and Streptococcus pneumoniae, underscores the need for targeted microbial surveillance and infection control measures within laboratory settings. Enhanced sanitation practices, routine disinfection of laboratory surfaces and strict adherence to personal protective equipment (PPE) protocols are essential measures for preventing cross-contamination. By implementing evidence-based interventions and fostering a culture of safety awareness, laboratories can mitigate risks associated with indoor air quality and promote healthier work environments for laboratory personnel.

Conclusion-

In light of these findings, it is imperative for laboratory management to prioritize measures aimed at improving indoor air quality and maintaining stringent hygiene protocols to mitigate potential health hazards associated with microbial contamination. Hence, continued monitoring and periodic assessment of indoor air quality parameters are essential for ensuring a safe and conducive working environment for laboratory personnel.

Reference-

1. Fra˛czek, K., &Grzyb, J. (2010). Analyses of bacterial aerosol occurring in health resorts in Bochnia and Szczawnica. Ecological Chemistry and Engineering A, 17, 55–63.
2. Yassin, M. F., &Almouqatea, S. (2010). Assessment of airborne bacteria and fungi in an indoor and outdoor environment. International Journal of Environmental Science and Technology, 7(3), 535–544.
3. Gizaw Z, Gebrehiwot M, Yenew C. High bacterial load of indoor air in hospital wards: the case of University of Gondar teaching hospital, Northwest Ethiopia. Multidiscip Respir Med. 2016;11:24.
4. Gołofit-Szymczak M, Go´rny RL. Bacterial and fungal aerosols in air-conditioned office buildings in Warsaw, Poland—the winter season. Int J OccupSaf Ergon. 2010; 16(4):465–76. https://doi.org/10.1080/ 10803548.2010.11076861 PMID: 21144265.
5. WHO (2016) Indoor Air Pollution Habitats and Health. Checklist N°292 Site ConsultedOnline. http://www.int/entity/mediacenter(fctsheets/fs292/fr/index.htlm,

6. Srikanth P, Sudharsanam S, Steinberg R. Bio-aerosols in indoor environment: composition, health effects and analysis. Indian J Med Microbiol. 2008 Oct-Dec; 26(4):302–12. https://doi.org/10.4103/0255-0857.43555 PMID: 18974481.
7. Bragoszewska E, and Biedron I, Indoor Air Quality and Potential Health Risk Impacts of Exposure to Antibiotic Resistant Bacteria in an Office Rooms in Southern Poland, (2018 a) Int. J. Env. Res. Pub. Health, https://doi.org/10.3390/ijerph15112604 PMID: 30469413.
8. Pasquarella C, Pitzurra O, Savino A. The index of microbial air contamination. J Hosp Infect. 2000;46(4):241–256. doi:10.1053/jhin.2000.0820.
9. Omelyansky VL. Manual in microbiology. Leningrad: USSR Academy of Sciences Moscow; 1940.
10. Malik, K.A. (1992). Technical Information for Culture Collections Curators in developing Countries, Braunschweig: Unesco/ WFCC Committee.
11. Salle, A. J. (1961) Laboratory Manual on Fundamental Principles of Bacteriology. New York : McGraw-Hill.
12. Commission of the European Communities. Indoor air quality and its impact on man. Report No. 12. Biological particles in indoor environments. Luxembourg: Commission of the European Communities; 1993.
13. Wong, L. T., Mui, K. W., Hui, P.S. & Chan, W. Y. 2009. An Implementation Choice of Assessment Parameters for Indoor Air Quality (IAQ) in Air-conditioned Offices. Emerald Group Publishing Limited. 202-210.
14. Meadow JF, Altrichter AE, Kembel SW, Kline J, Mhuireach G, Moriyama M, et al. Indoor airborne bacterial communities are influenced by ventilation, occupancy, and outdoor air source. Indoor Air 2014; 24(1): 41-48.
15. Soto T, Murcia RM, Franco A, Vicente-Soler J, Cansado J, Gacto M. Indoor airborne microbial load in a Spanish university (University of Murcia, Spain). An Biol 2009; 31: 109-115.
16. Ghayoor, M., Qadoos, A., Bahadar, S., Hayat, A., Daud, M., Hassan, A., Ali, F., Zeb, A., Rahman, K. U., Wahab, A., Khattak, Z. F. &Khattak,B. 2015. Isolation and Identification of common contaminants bacteria from working area in microbiology laboratory. Journal of Bio-Molecular Sciences 3(2): 74-78.
17. Douglas-Traber, K. B. & Shanks, C. A. 2001. Glass Particle Contamination in Single-Dose Ampules. J. Lab. 65: 1361- 1363.
18. Nevalainen A, Morawaska L. Biological agents in indoor environments.Assessment of health risks. Work conducted by a WHO Expert Groupbetween 2000–2003. Available from: http://www.buildingforensics.co.uk/uploads/files/.co.uk 996163695.rtf.
19. Commission of European Communities. Biological particles in indoor environments, European Collaborative Action - Indoor Air Quality and its Impact on Man, Report No. 12, CEC, Luxembourg. Available from:http://www.soer.justice.tas.gov.au2003/source/94/index.php.
20. Kim, K.Y., Kimb, H.T., Kim, D., Nakajimad, J. and Higuchi, T. (2009) Distribution Characteristics of Airborne Bacteria and Fungi in the Feed Stuff Manufacturing Factories. Journal of Hazardous Material, 169, 1054-1060.
21. J. Kurlenda and M. Grinholc, “Alternative therapies in staphylococcus aureus diseases,” *Acta Biochimica Polonica*, vol. 59, no. 2, pp. 171–184, 2012.

22. Emergence of coagulase-negative staphylococci(2020)Karsten Becker ,Anna Both, Samira Weißelberg, Christine Heilmann&Holger Rohde, Pages 349-366 . https://doi.org/10.1080/14787210.2020.1730813.
23. C.U. Tuazon, "Other bacillus species," in *Principles and Practice of Infectious Diseases, Mandell Bennett Dolineds*, Churchill Livingston, NewYork, USA, 2000.
24. Wayne, L., Nicholson, N., Munakata, G.H., Henry Melosh, J. and Peter, S. (2000) Resistance of Bacillus Endospores to Extreme Terrestrial and Extraterrestrial Environment. Microbiology and Molecular Biology Reviews, 64, 548-572.
25. Whyte, P., Collins, J.D., McGill, K., Monahan, C. and O'Mahony, H. (2001) Distribution and Prevalence of Airborne Microorganisms in Three Commercial Poultry Processing Plants. Journal of Food Protection, 64, 388-391.
26. Purushothama V. Dasaraju and ChienLiu (1996). Infections of the Respiratory System Medical Microbiology. 4th edition. Chapter 93. Baron S, editor. Galveston (TX): University of Texas Medical Branch at Galveston.
27. Kavita, N. and Jyoti, G. (2013) Microbial Contamination in a School. International Journal of Current Microbiology and Applied Sciences, 12, 404-4010.
28. Assessment of Indoor Microbial Quality of Library's Premise: Case of Central Library of the University of Yaoundé I.
29. Mandal, J. and Brandl, H. (2011) Bioaerosols in Indoor Environment. The Open Environmental and Biological Monitoring Journal, 4, 83-96.