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An Investigation on the Indoor Air Quality in 2 Nursery Schools

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Abstract

Indoor Air Quality (IAQ) has a significant impact on a student's ability to succeed. The purpose of this study was to conduct an investigation on the Indoor Air Quality in 2 Nursery Schools. The researcher utilized a quantitative design for this study. For each school, culture media bi-plates containing Nutrient Agar and Sabouraud Dextrose Agar were used to obtain bacterial and fungal levels for two days. Subsequently, the plates were incubated. Colony counts were conducted and compared to the maximum limits set by the WHO guidelines for bacterial and fungal count of 500 CFU/m³ and 1000 CFU/m³ respectively. Each bacterium isolated was gram stained to ascertain whether it was gram-positive or negative, and the genus of fungi was identified through microscopic examination. Results were analyzed using Excel 2016. The findings revealed that, in most cases, fungal levels were below the recommended standard. Eight genera of fungi were successfully identified. These were *Aspergillus*, *Penicillium* sp., *Chrysosporium* sp., *Paecilomyces* sp., *Cladosporium herbarum*, *Penicillium digitatum*, *Chrysonilia* sp., and *Cladosporium* sp. The IAQ level in both nursery schools was at an unacceptable level due to high bacterial levels. This finding could be due to high student activities, high occupancy number, lower temperature, and a higher relative humidity percentage. Three groups of culturable bacteria were identified, namely Gram-positive cocci, Gram-positive bacilli and Gram-negative bacilli. This finding may contribute to diseases in children such as upper respiratory tract ailments and allergic symptoms, such as headaches, coughing, and itchy skin. This study made a unique contribution as it is the first study in Guyana to determine the levels of airborne bacteria and fungi in schools. Compared to adults, children are more vulnerable to air pollution due to their underdeveloped respiratory and immune system.

Keywords: Indoor Air Quality, Health Implications, Children

1. Introduction

Clean air is a basic necessity for a healthy life.[1] Outside of the home, school is where children spend the second-highest percentage of their time.[2] Therefore, good Indoor Air Quality (IAQ) is vital because it contributes to a safe, healthy, comfortable, and productive environment for both students and staff.[2] Despite its importance, good IAQ seems to be a standard that is difficult to achieve. Approximately 1 million children under five years old are dying annually due to exposure to indoor air pollutants.[3] Meanwhile, millions of other children are suffering from respiratory diseases such as asthma that reduce their resilience and affect their cognitive and physical development.[4] For example, in the United Kingdom, approximately 1.1 million children suffer from asthma symptoms, and there is always an increase in hospital admissions in September, which corresponds closely to students return to the indoor school environment.[5]

Several factors compromise the IAQ in schools by contributing to elevated levels of pollutants such as airborne bacteria and fungi. Some of these factors are overcrowded classrooms, inadequate ventilation, location of schools in close proximity to major roads, and unsanitary conditions.[6] There have been news report about unsanitary conditions at educational institutions in Guyana. Two primary schools were reported to have overflowing sewage due to a flood event, and the schools were shut down to facilitate repairs.[7] This created a toxic school environment for students and teachers and disrupted the normal functioning of the schools.[7] Recently, the Ministry of Education also closed a secondary school because one of the main washroom facilities was flooded, which became breeding grounds for mosquitoes that can spread viruses.[8] Additionally, the excess moisture contributed to mold growth in the school.[8]

Currently, there are no standards on exposure levels for airborne bacteria and fungi in nursery schools;[3] however, the World Health Organization (WHO) has published guidelines on exposure levels. The guidelines stipulate that the IAQ should not exceed a total bacterial and fungal count of 500 CFU/m³ and 1000 CFU/m³, respectively.[10] These guidelines protect occupants' health from the adverse effects of poor IAQ as well as eliminate or decrease exposure to these dangerous pollutants.[10]

Although the WHO guidelines for exposure exists, airborne bacteria and fungi are present in a school environment at significant levels.[3,9,11-14] For instance, Hasnah et al. revealed that the total level of bacteria exceeded the established guideline and three types of culturable bacteria were detected: Gram-negative bacteria, Gram-positive cocci, and Gram-positive bacilli.[3] The elevated levels of airborne bacteria and fungi could have been attributed to wall posters, high occupancy numbers, student activities, high relative humidity, and high temperature.[3]

Mainka et al. and Nabilah et al. found similar results with respect to the presence of bacteria in schools in excess of the WHO guideline.[9,11] Five reasons posited for this finding in the two studies were resuspension of road dust, high temperatures, high relative humidity, particulate matter, and high densities of students. These researchers suggested that children's saliva and respiratory tract have a variety of bacteria and fungi; therefore, high densities of students could cause bacteria to be released into the indoor air environment via sneezing, talking, coughing, and shedding of skin epidermis.[9,11] Gram-positive cocci and Gram-positive bacilli were dominant indoors and outdoors, respectively. Gram-positive cocci can be found on the skin and mucous membranes of humans or animals and can cause fever, rash, sore throat, diarrhea, conjunctivitis, and skin infections.[9,11] Whereas, Gram-positive bacilli are commonly found in the skin and mucosa flora of individuals and many animals, samples of food, and in dirty and dusty areas. This type of microbe can cause severe health implications such as inflammation of the eye, ear infections, peritonitis, meningitis, and respiratory and urinary tract infections.[9,11]

On the other hand, Crow et al., Viegas et al., and Mohan et al. carried out studies to examine schools for airborne fungi. The studies identified *Cladosporium* sp. and *Penicillium* sp. as being the dominant species present in the schools sampled. These genera pose a severe threat to humans, and their presence in schools is a great concern.[12-14] They produce mycotoxins, such as aflatoxin, ochratoxin and are commonly found in soils, food products, plants, painted surface, carpets, and wallpaper. Exposure to this pollutant over a prolonged period may lead to chronic and fatal illnesses such as lupus, chronic fatigue disorder, cancer, vomiting, sneezing, runny nose, burning eyes, nausea, and allergies.[17,18] Although *Cladosporium* sp. and *Penicillium* sp. were dominant in the indoor air, *Cladosporium herbarum* and *Chrysonilia* sp. were also present. *Cladosporium herbarum* lives on wallpaper, painted walls, fans, and carpet, whereas *Chrysonilia* sp. thrives in environments such as soil.[13,14] These fungi genera can cause respiratory diseases, eye, skin, and brain infection in students, teachers, and cleaners.[12-14] Mohan et al. suggested that schools located in coastal environments experienced high relative humidity and cooler conditions that are suitable for fungal growth.[14] However, while Crow et al. found that temperature and relative humidity were directly related to fungal concentration, Viegas et al. did not find this expected relationship.

According to Pommerville, most fungi thrive best at about 23°C while for others such as yeast and mold the optimum temperature is 36°C.[15] Fungi and bacteria grown under body temperature (36°C) are the ones that have severe health implications in building occupants such as headaches, tuberculosis, watery eyes, influenza, meningitis, whooping cough, skin diseases such as cellulitis, bronchitis, asthma, and allergic reactions in occupants.[15] Children are more vulnerable to air pollution due to their underdeveloped respiratory and immune system, and due to them being very active, they breathe a higher volume of air relative to their body weight.[9]

This current investigation sought to determine the levels of airborne bacteria and fungi in two nursery schools. The bacterial types and fungal genera identified were compared and contrasted with the findings of Crow et al., Viegas et al., Mohan et al., Mainka et al., Hasnah et al., and Nabilah et al. and to determine whether the rationale for low and elevated levels of airborne bacteria and fungi were comparable for schools in Guyana.

2. Methodology

A quantitative design was used to gather the data needed for the project during September 2019. The Nursery schools were purposively selected from schools within a five-mile radius of the University of Guyana based on three criteria. First, the schools consisted of one building in a compound. Second, the location of the schools, one school was located close to a roadway with moderate traffic activity while the other one was close to roads with low traffic activity. Moderate and low traffic activity was defined as those streets where the number of cars traveling is >100 cars/h and <100 cars/h, respectively.[16] Third, the design of the school buildings was one story flat buildings with natural ventilation (windows and doors) and mechanical ventilation (fans).

Passive air quality monitoring for bacteria and fungi was done in 10 classrooms, kitchen, headmistress office and on two windows in School A and 6 classrooms, kitchen, headmistress office and on one window in School B. Dual agar sedimentation bi-plates were used to collect samples from two Nursery schools for two days. At both schools, two bi-plates containing nutrient agar (NA) for bacteria and sabouraud dextrose agar (SDA) for fungi were placed in the same location and was exposed to the air for three hours (08:00 am to 11:00 am). From each sampling location, one of the bi-plates were incubated for twenty-four (24) hours at body temperature (36°C±1°C) and the other at room temperature (27°C±1°C) to isolate bacteria and fungi (yeasts and molds). Colony counts were conducted and compared to the maximum limits set by the WHO guidelines for bacterial and fungal count of 500 CFU/m³ and 1000 CFU/m³ respectively. Each bacterium isolated was gram stained to ascertain whether it was gram-positive or gram-negative and the genus of fungi was identified through microscopic examination. On each day of sampling, the total number of bi-plates used were twenty-eight (28) for School A and eighteen (18) for School B. A photograph of one of the bi-plates post incubation is shown in Figure 3.

The levels of airborne bacteria and fungi were organized and analyzed using Excel 2016. Descriptive statistics such as mean, standard deviation, minimum and maximum values were calculated. Tables were generated to represent the data visually.

Figure 1. Sterilization of Culture Medium

Figure 2. Media plates that were incubated at 36°C

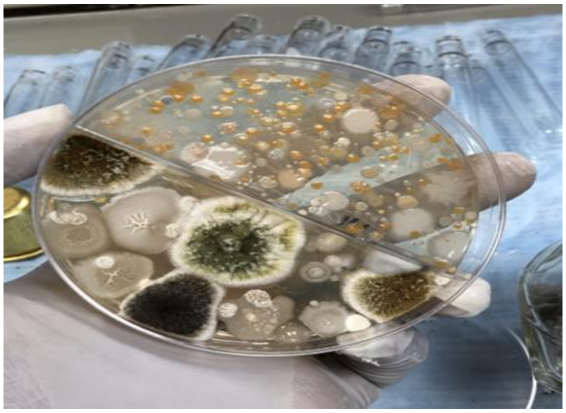

Figure 3. Airborne bacteria and fungi

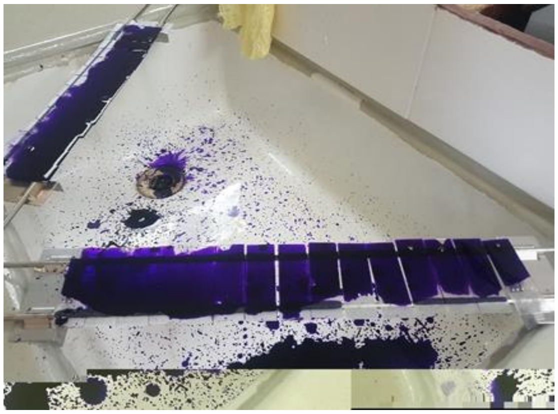

Figure 4. Gram-staining

2.1 Ethical Considerations

- Permission and confirmation- A letter was written to the Ministry of Education seeking permission to use two Nursery Schools. Permission was given.
- The researcher also sought consent from the headmistress of both schools prior to the study.
- This study did not involve any communication or collection of information from students.

2.2 Limitations

The limitations that affect the findings of this research were:

- The researcher was unable to identify airborne bacteria species because the lab did not have those capabilities.
- Sampling for airborne bacteria and fungi was only done for two days due to limited resources such as reagents and media plates.

3. Results and Discussion

Table 1. Statistical summary on the levels of airborne bacteria in School A and School B

Day 1 Levels of Bacteria (WHO Limit- 500 CFU/m³)					
Schools	**Incubation Temperatures**	**Mean (CFU/m³)**	**Standard Deviation (CFU/m³)**	**Minimum (CFU/m³)**	**Maximum (CFU/m³)**
School A	27°C ±1°C	3269	1579	580	5885
	36°C ±1°C	2279	701	1200	3995
School B	27°C ±1°C	1326	476	514	2155
	36°C ±1°C	2271	967	1160	3581
Day 2 Levels of Bacteria (WHO Limit- 500 CFU/m³)					
Schools	**Incubation Temperatures**	**Mean (CFU/m³)**	**Standard Deviation (CFU/m³)**	**Minimum (CFU/m³)**	**Maximum (CFU/m³)**
School A	27°C ±1°C	1803	908	600	3995
	36°C ±1°C	2275	1670	1000	7261
School B	27°C ±1°C	1000	283	580	1500
	36°C ±1°C	1397	674	796	2901

After examining the plates that were incubated at 27°C±1°C and 36°C±1°C for days 1 and 2, School A and School B exceeded the WHO exposure limit for bacteria, shown in Table 1. All 28 and 18 bi-plates for School A and School B respectively had counts that exceeded the WHO limit. This meant every room was deemed to be unsatisfactory with respect to the WHO guideline.

The participating schools were closed during the period July – August. In preparation for the reopening of the schools, teachers and cleaners periodically conducted extensive cleaning. Both schools reopened on September 02, 2019 and data were collected on September 10 and 17 of 2019. Noteworthy, although extensive cleaning took place before the schools reopened, airborne bacteria were still present in the indoor air environment at unacceptable levels. The high levels of airborne bacteria could be due to environmental factors such as high temperature. Throughout the data collection process, the indoor temperature of both schools ranged from 23°C to 34°C. Additionally, the relative humidity on both days ranged from 70% to 90%. Therefore, elevated levels of airborne bacteria could be associated with high relative humidity. A possible reason might be that if the school's relative humidity decreases, the bacterial load may decrease because it hinders their ability to survive. A dry indoor environment reduces the physiological activities and metabolism of microorganisms. Other researchers are of the view that low temperatures and low relative humidity are suitable conditions for bacteria growth.[3,11]

In this study, bacteria grown under body temperature were 7 to 11 times higher than the established guideline. Exposure to high levels of bacteria grown under body temperature can cause headaches, tuberculosis, watery eyes, influenza, meningitis, whooping cough, skin diseases such as cellulitis. Moreover, it can cause bronchitis, asthma, and allergic reactions in occupants.[11]

Table 2. Most prevalent types of bacteria identified in the selected schools.

Room temperature				**Body Temperature**			
Schools	**Types**	**Frequency (*n*)**	**Percentage (%)**	**Schools**	**Types**	**Frequency (*n*)**	**Percentage (%)**
School A	Gram-Negative Bacilli	8	14	School A	Gram-Negative Bacilli	8	12
	Gram-Positive Bacilli	12	21		Gram-Positive Bacilli	31	47
	Gram-Positive Cocci	24	43		Gram-Positive Cocci	20	30
	No Bacteria	12	21		No Bacteria	7	11
School B	Gram-Negative Bacilli	3	10	School B	Gram-Negative Bacilli	5	12
	Gram-Positive Bacilli	3	10		Gram-Positive Bacilli	17	40
	Gram-Positive Cocci	16	55		Gram-Positive Cocci	16	37
	No Bacteria	7	24		No Bacteria	5	12

From the 28 samples collected from School A, various types of bacteria were identified from the plates incubated at room and body temperature. However, since bi-plates were used, after incubation, some of the fungi started growing on the nutrient agar. Therefore "no bacteria" were detected. At room temperature, Gram-positive cocci were the most frequently isolated bacteria at School A (43%) and School B (55%), shown in Table 2. A similar finding was made by Mainka et al. where high student activity caused Gram-positive cocci to be spread to the air from their bodies and airways.[9]

At body temperature, Gram-positive bacilli were the most prevalent bacteria at School A (47%) and School B (40%). One possible reason for this finding could have been attributed to the dirty fans and cupboards, which consisted of a vast number of dusty books in both schools. A different result was seen in the literature reviewed.[9,11]

Similar to day 1, after examining the plates that were incubated at room temperature for day 2, Gram-positive cocci were the most dominant culturable bacteria found in School A (60%) and School B (55%). Whereas at body temperature, Gram-positive bacilli were the most frequently identified type isolated on culture plates from School A (53%) and School B (48%), shown in Table 3.

At School A, 26½ out of the 28 bi-plates recorded total fungal levels below the acceptable limit. Although the levels of fungi did not exceed the recommended threshold in most cases, there were still high in the indoor air of both schools. Whereas, for school B, all 18 bi-plates were below the acceptable limit. School A had a higher fungal count than

School B. It is likely that location may play a significant role in influencing these observed differences. Since School A is in closer proximity (0.89 km) to the Atlantic Ocean, the relative humidity and cooler environmental conditions

Table 3. Statistical summary on the levels of airborne fungi in School A and School B

Day 1 Levels of Fungi (WHO Limit-1000 CFU/m³)					
Schools	**Incubation Temperatures**	**Mean (CFU/m³)**	**Standard Deviation (CFU/m³)**	**Minimum (CFU/m³)**	**Maximum (CFU/m³)**
School A	27°C ±1°C	703	253	332	1210
	36°C ±1°C	716	763	83	3000
School B	27°C ±1°C	392	206	133	663
	36°C ±1°C	448	125	249	630
Day 2 Levels of Fungi (WHO Limit-1000 CFU/m³)					
Schools	**Incubation Temperatures**	**Mean (CFU/m³)**	**Standard Deviation (CFU/m³)**	**Minimum (CFU/m³)**	**Maximum (CFU/m³)**
School A	27°C ±1°C	671	234	332	1144
	36°C ±1°C	593	265	332	944
School B	27°C ±1°C	330	152	99	613
	36°C ±1°C	538	245	50	812

at this location likely prove more suitable for fungal growth. Similarly, Mohan et al. found that schools located in the coastal environment had more fungal growth due to high relative humidity and cooler conditions.[14]

Moreover, both schools had educational wall posters that teachers used to create a colorful, intriguing environment for students. However, these posters may promote the growth of fungi. Toxins from fungi growing on posters can become airborne, thus creating an indoor health risk such as asthma attacks, allergic reactions, inflammation, and irritation to the mucous membrane and eye infection, which can further lead to blindness.[3]

Most fungi thrive best at about 27°C.[15] However, in this study, there was a higher concentration of fungi at body temperature (36°C ±1°C) when compared to room temperature (27°C ±1°C). This finding may be due to those fungi found in schools in a high quantity such as yeast and mold. Fungi grown at body temperatures can cause adverse health effects such as Allergic Bronchopulmonary Mycosis, Hypersensitivity Pneumonitis and Allergic Fungal Sinusitis.[15]

Table 4. Most prevalent fungi genera identified in the selected schools

Room Temperature Fungi				**Body Temperature Fungi**			
Schools	**Genera**	**Frequency (*n*)**	**Percentage (%)**	**Schools**	**Genera**	**Frequency (*n*)**	**Percentage (%)**
School A	*Penicillium* sp.	9	15	**School A**	*Penicillium* sp.	9	15
	Chrysosporium sp.	3	5		*Chrysosporium* sp.	3	5
	Paecilomyces sp.	12	20		*Paecilomyces* sp.	12	20
	Cladosporium herbarum	16	27		*Cladosporium herbarum*	16	27
	Aspergillus	4	7		*Aspergillus*	4	7
	Pencillium digitatum	4	7		*Pencillium digitatum*	4	7
	Chrysonilia sp.	3	5		*Chrysonilia* sp.	3	5
	Cladosporium sp.	9	15		*Cladosporium* sp.	9	15
School B	*Penicillium* sp.	5	27	**School B**	*Penicillium* sp.	5	27
	Cladosporium herbarum	1	6		*Cladosporium herbarum*	1	6

	Chrysonilia sp.	12	67		*Chrysonilia* sp.	12	67

From the samples collected from School A and School B, various genera of fungi were identified from the plates incubated at room and body temperature as shown in Table 4. At room temperature, *Cladosporium herbarum* (27%) was the most prevalent culturable fungal type found indoors at School A. Whereas, at School B, *Chrysonilia* sp. (67%) was the most dominant culturable fungal type. The results were similar for those fungi grown at body temperature. However, a different result was seen in a study done by Viegas et al. and Crow et al. Their findings indicated that the dominant species in the investigated schools were *Cladosporium* sp. and *Penicillium* sp., respectively. Similar to day 1, after examining the plates that were incubated at room and body temperature for day 2, *Cladosporium herbarum* (35%) and *Chrysonilia* (55%) were the most frequently identified type isolated on culture plates from School A and School B respectively. Although *Aspergillus* sp., *Penicillium* sp., and *Cladosporium* sp. were not the predominant fungi genera found in schools, these genera are the greatest threats to humans.[13]

All plates that were placed in the kitchen of both schools had the lowest fungal and bacterial counts. However, the highest bacterial and fungal colony counts were detected on plates that were located in the headmistress office and classrooms for both schools. During the data collection process for airborne bacteria and fungi, the researcher observed high densities of students in a limited space. For instance, at School B; 24-31 students occupied classrooms that were 129.6 sq. ft. in size. Meanwhile, School A 130 sq. ft. classrooms had approximately 15-21 students.

Furthermore, roads surrounding participating schools are access roads especially for students and staff attending the University of Guyana, residents in the community and persons traversing along the East Coast of Demerara. As a result, traffic activity close to nursery schools (particularly School A) generated road dust. Prevailing winds could have carried road dust into schools through open doors and windows. Road dust can cause deposition in the respiratory tract, chronic obstructive pulmonary disease, asthma, and allergies.[9,17]

Infectious diseases may spread more quickly among children in this age group since there are now in the process of learning and practicing good personal hygiene. Direct contact during games, inadequate hand-washing, and being in constant motion may aid the spread of fungal and bacterial infections. Children have underdeveloped immune and respiratory systems,[9] hence their vulnerability to high bacterial levels and even below the guideline level for fungal exposure. Because they are very active, they breathe a higher volume of air than their body weight.[9]

4. Conclusion

The IAQ level in both nursery schools was at an unacceptable level due to high bacteria levels (>500 CFU/m³). Although fungi levels did not exceed the recommended WHO threshold (<1000 CFU/m³) in most cases, there was still a considerable number of fungi in the indoor air of both schools. Gram-positive cocci and Gram-positive bacilli were the most frequently identified bacterial type isolated on culture plates from School A and School B, respectively. Meanwhile, *Cladosporium herbarum* and *Chrysonilia* sp. were the most prevalent fungal genera found indoors at School A and School B respectively. Although *Aspergillus* sp., *Penicillium* sp., and *Cladosporium* sp. were not the predominant fungi genera found in the participating schools, these genera are a great concern. Exposure to these pathogenic microorganisms can cause health implications in building occupants such as bronchitis, asthma, allergies, headaches, watery eyes, and skin diseases such as cellulitis.

5. Recommendations

1. There are lack of data on IAQ in schools in Guyana. Therefore, more research needs to be done to investigate the IAQ in all schools, especially nursery schools, over a prolonged period.
2. The Guyana Environmental Protection Agency in collaboration with the relevant stakeholders should develop standards for airborne bacteria and fungi in nursery schools.
3. An IAQ Management Plan should be developed and implemented to monitor and enhance the quality of air in school buildings. The objectives of this plan should be to:
   - Reduce indoor air pollutant levels through preventative measures such as regular building inspections and evaluations, maintenance activities, and IAQ policies.
   - Respond to IAQ-related problems and concerns thoroughly and promptly, through effective communication and documentation.

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7. Endnotes

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