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EVALUATION OF INDOOR AIR BACTERIA LOAD AND THE ANTIBIOTIC SUSCEPTIBILITY PROFILE OF ISOLATES FROM LIBRARIES IN KADUNA STATE UNIVERSITY, NIGERIA

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ABSTRACT

Background: The quality of air inhaled by man has the tendency to impact his state of well-being. Since considerable number of hours are usually spent in the library by students and lecturers the microbiological quality of university libraries' indoor air is highly essential. The aim of this study was to assess the microbiological quality of indoor air of the libraries in Kaduna State University, and to determine the antibiotics susceptibility of the bacteria isolated.
Methods: A total number of 26 samples were collected from eight Libraries including the University main Library and Faculty Libraries morning and afternoon. Settle plate method, using open petri dishes containing nutrient agar was used, bacterial concentration of the indoor air in the various sampling site was determined. Biochemical tests were used to identify the bacterial isolates. Agar disc diffusion method was used for the susceptibility testing and multidrug resistant bacteria was detected.
Results: The concentration of bacteria in the university indoor air ranged from 1.26 x 10^3 to 6.13 x 10^3 CFU/m^3. The bacteria isolated were *Staphylococcus aureus* 7 (28%), *Bacillus subtilis* 7 (28%), *Klebsiella pneumoniae* 4 (16%), *Streptococcus pneumoniae* 3 (12%), *Micrococcus spp* 2 (8%) and *Pseudomonas aeruginosa* 2 (8%). Amoxicillin- clavulanate (84.2%) was the most active against Gram positive organisms while gentamicin (83.3%) was the most active against Gram negative. Five (83.3%) and 9 (47.4%) of Gram negative and Gram positive respectively were multidrug resistant.
Conclusion: The indoor air of Kaduna State University libraries was highly contaminated with bacteria. Proper and regular cleaning of the libraries at least twice a day, and proper ventilation are recommended.
Keywords: antibiotics, bacteria, contamination, concentration.

INTRODUCTION

Most day-to-day human activities take place indoor in places like offices, classrooms, laboratory, and the library, people spend 80 – 90% of their time in indoor environment [1]. Indoor air quality (IAQ) is the quality of air within and around a building. It is an important environmental health matter that needs to be assessed because people inhale 6–10 liters of air per minute, which amounts to 11,000 liters per day [2]. According to studies

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conducted in the last 20 years by the United States Environmental Protection Agency, indoor air is often more polluted than outdoor air, and indoor air pollution has been ranked among the top five risks to public health [3]. The health effects associated with poor air quality contribute to heart and lung diseases.

Bioaerosol particles are one of the pollutants that can cause a reduction in IAQ in library. They account for 5– 34% of indoor air pollution [4]. A bioaerosol is a colloidal suspension formed by liquid droplets and particles of solid matter in the air [5]. These particles suspended in the air may consist of bacteria, fungi, viruses, fragments of any or all of these or their metabolic products (e.g., mycotoxins), endotoxins (part of the outer membrane of the cell wall of Gram-negative bacteria), plant pollen, and fragments of plant tissues. Bioaerosols can become a serious risk to the health of the population, mainly because airborne bacteria and fungi can cause infectious diseases, as well as allergic and toxic effects. Microbes from the soil or from plants can also be vectored by students or can be carried on dust particles from the outdoor air [6]. In fact, an air-conditioning (AC) system is practically the only technical solution used both to improve the air quality and to provide students with proper reading conditions.

The library is an indispensable place for some students and most spend a lot of time in the library. Due to inadequate hygiene and improper ventilation system, many indoor environments have a low IAQ with the library inclusive. The sources of library airborne contamination could be traced to a variety of factors. These include the student's own normal flora, students' clothes, bags, shoes; as well as activity of students like sneezing, coughing, talking and yawning [7,8]. Materials such as shelves, books and files have been implicated as viable sources. House-keeping activity of the library such as sweeping or using dry dust mops can aerosolize particles that may contain microorganisms. Infectious nuclei in air currents and dust may be inhaled during normal breathing [9-11].

The number of microorganisms present in library will depend on the number of students occupying the library, the amount of physical activity, the rate of air exchange, the ambient temperature, relative humidity, level of environmental sanitation, type of ventilation, numbers of windows available for cross ventilation amongst others [8]. World Health Organization in an epidemiological studies review showed that there is association between indoor dampness-related factors and a large effect on respiratory health. These include asthma development, asthma exacerbation, current asthma, respiratory infections, upper respiratory tract symptoms, wheeze, cough and dyspnoea [12]. Poor indoor air quality can be harmful to vulnerable groups such as children, young adults, the elderly, or those suffering from chronic respiratory and/or cardiovascular diseases [13].

Thus, microbiological air quality is highly important in the university libraries and other indoor workplaces in order to provide a safe environment. The aim of this research was to assess the bacterial contaminants in indoor air of the central and faculties libraries of Kaduna State University, Kaduna, Nigeria and to determine the antibiotics susceptibility profile of the isolates to commonly prescribed antibiotics.

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MATERIALS AND METHODS

Study area

The study area was the main library and faculty libraries of Kaduna State University (KASU), Tafawa Balewa way, Kaduna town, Kaduna State. Kaduna is about 162 km away from the Federal Capital Territory (FCT) Abuja and 234 km away from Kano, the capital of Kano state. Tafawa Balewa is located on latitude 10.44565N and longitude 7.4565N. The eight university libraries involved in this study were main KASU library, KASU e-library, Faculty of Pharmaceutical Sciences library, Faculty of Arts library, Faculty of Science library, Faculty of social science library, College of Medicine library and Faculty of Management science library.

Sample collection

Air sampling was made by passive air sampling technique: the settle plate method using 9 cm diameter Petri dishes containing nutrient agar. The sampling height which approximated to human breathing zone was 1 m above the floor and at the center of the room. The sampling time was 15minutes. Samples were collected twice a day at 8:00 a.m. and 4:00 p.m. After exposure the sample were taken to pharmaceutical microbiology laboratory, Faculty of pharmaceutical sciences, Kaduna state university for analysis. The plates were incubated at 37 °C for 24 h. Once colony forming units (CFU) were enumerated, CFU/ m^3 were determined, using the following equation [14].

$N = 5a \times 10^4 (bt)^{-1}$,

Where N = microbial CFU/m^3 of indoor air ; a = number of colonies per Petri dish ; b = dish surface (cm^2) ; t = exposure time (min).

Purification and identification of isolates

Each colony was isolated in a pure form by sub culturing in a fresh nutrient agar plate and pure cultures were preserved on nutrient agar slants at 4°C for further work. An overnight activated culture was then used to determine colony morphology, gram staining and biochemical tests. The procedures for the biochemical tests were carried out as described by Chesbrough [15]. These include indole, citrate utilization, catalase, oxidase, coagulase, methyl red, Voges-Proskauer, and glucose fermentation tests. The samples were also grown on selective media such as mannitol salt agar, cetrimide, and MacConkey agar.

Antibiotics Susceptibility Testing

This was carried out using Kirby Bauer agar disc diffusion test according to Clinical Laboratory Standard Institute [16] guidelines. The results were recorded as sensitive, intermediate and resistant. The following antibiotic discs were used: Gentamicin (10µg), Chloramphenicol (30 µg), Ceftriaxone (30 µg), Cefoxitin (30µg), Tetracycline (30µg), Ciprofloxacin (5µg), Erythromycin (15µg), Vancomycin (30µg), Cotrimoxazole (15µg), Amoxicillin/clavulanic acid (30 µg)

Detection of Methicillin-resistant *Staphylococcus aureus*

Clinical and Laboratory Standards Institute recommends Cefoxitin disc diffusion test as a laboratory test for MRSA. This was carried out according to CLSI guidelines [16].

Determination of multidrug resistance

Isolates that were non susceptible to at least one agent in three or more antimicrobial categories were defined as multidrug resistant (MDR) [17].

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STATISTICAL ANALYSIS

Descriptive analysis was carried out using Microsoft excel.

RESULTS

A total of 26 air samples were collected from eight libraries in Kaduna State University, Kaduna State. The average number of bacterial colony forming unit CFU/m^3 in the libraries studied are presented in Table 1. Faculty of Management Science had the least bacterial concentration both at 8.00 am and 4.00 pm sampling, 1.26 x10^3 and 3.04 x10^3 respectively. While Faculty of Science had the highest bacterial concentration at both morning and evening sampling, 3.20 x 10^3 and 6.13 x 10^3 respectively. The biochemical characterization of the bacterial isolates indicated the presence of *Staphylococcus aureus*, *Bacillus subtilis*, *Streptococcus pneumoniae*, *Pseudomonas aeruginosa*, *Klebsiella pneumoniae* and *Micrococcus* spp. The frequency of occurrence of the bacterial isolates and their occurrence in each library is presented in Tables 2 and 3 respectively. *S. aureus* and *B. subtilis* had the highest frequency of 7 (28%) each while *P. aeruginosa* and *Micrococcus* spp had the least frequency 2(8%).

Table 1: Number of Bacterial Colony Count in CFU/m^3

S/No	Samples Sites and Abbreviation	Number of Samples (CFU/m^3)	
		8:00am (1)	4:00pm (2)
1.	Main Library ML	1.91 x 10^3	5.82 x 10^3
2.	E- Library EL	2.0 x 10^3	3.31 x 10^3
3.	Faculty of Arts, FA	2.54 × 10^3	3.25 × 10^3
4.	Faculty of Science, FS	3.20 × 10^3	6.13 × 10^3
5.	Faculty of Management Science, FMS	1.26 × 10^3	3.04 × 10^3
6.	Faculty of Pharmaceutical Sciences, FPS	2.20 × 10^3	5.60 × 10^3
7.	College of Medicine, CM	1.52 × 10^3	4.56 × 10^3
8.	Faculty of Social Sciences, FSS	1.73× 10^3	4.87 × 10^3

The total number and percentage of Gram negative isolates were 6 (24%) while that of Gram positive was 19 (76%). Table 4 showed the evaluation of air quality in the libraries according to Commission of European Community [18].

Table 2: Frequency of Occurrence of Bacterial Isolates and Percentage

Bacteria	**No. of Occurrence**	**Percentage (%)**
Staphylococcus aureus	7	28
Streptococcus pneumoniae	3	12
Pseudomonas aeruginosa	2	8
Klebsiella pneumoniae	4	16
Bacillus subtilis	7	28
Micrococcus spp	2	8
Total	**25**	**100**

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Table 3: Bacterial isolates from each Kaduna State University Libraries

Bacteria	ML	EL	FS	FMS	FPS	FA	FSS	CM
S. aureus	+	+	+	+	+	+	-	+
S. pneumoniae	+	+	-	-	-	+	-	-
B. subtilis	+	+	-	+	+	+	+	+
Micrococcus spp	-	-	+	-	-	-	+	-
Ps. aeruginosa	+	-	+	-	-	-	-	-
K. pneumoniae	-	+	+	-	+	-	+	-

Key: ML = Main library, EL = E-library, FS = Faculty of Science, FMS = Faculty of Management science, FPS = Faculty of Pharmaceutical Sciences, FA = Faculty of Art, FSS = Faculty of Social Sciences, CM = College of Medicine, + = present, (-) = absent

Table 4: Evaluation of AIR quality in the Designated Libraries at Kaduna State University according to the Sanitary Standards for Non-industrial premises

Range of values	Pollution degree	ML		EL		FS		FMS		FPS		FA		FSS		CM	
		A	B	A	B	A	B	A	B	A	B	A	B	A	B	A	B
< 50	Very low	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
50 – 100	Low	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
100 – 500	Intermediate	-	-	-	-	-	-	-	-	-	-	-	-	-	-	--	-
500-2000	High	+	-	+	-	-	-	-	-	+	-	-	-	+	-	+	-
> 2000	Very high	+		+		+	+	+	+	+		+	+	+		+	

Key: ML = Main library, EL = E-library, FS = Faculty of Science, FMS = Faculty of management science, FPS = Faculty of Pharmaceutical Sciences, FA = Faculty of Art, FSS = Faculty of Social Sciences, CM = College of Medicine, (+) in the range, (-) not in the range, A = 8am B = 4pm

Antibiotics Susceptibility Testing Results

Methicillin resistance was detected from only 2 out of the 7 *S. aureus* isolates meaning 28.6% were MRSA while 5 (71.4%) were methicillin sensitive *S. aureus.* The results of the susceptibility testing showing percentage sensitive, intermediate and resistant for *S. aureus*, Gram negative and Gram positive isolates are presented in Table 5 and Figures 1 and 2 respectively. All the *S. aureus* isolates showed 100% susceptibility to amoxicillin/clavulanic acid. Considering the antibiotics activities against Gram positive isolates, amoxicillin/clavulanic acid was the most active, 84.2%, followed by vancomycin (73.7%), ceftriaxone (63.2%), ciprofloxacin and gentamicin (57.9%). But gentamicin (83.3%) demonstrated the highest activity against the Gram negative isolates followed

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by ciprofloxacin and ceftriaxone (66.7%). Both the Gram negative and Gram positive isolates were resistant to erythromycin, 66.7% and 52.6% respectively.

Five out of the six Gram negative isolates were multidrug resistant (83.3%) while 9/19 (47.4%) were multidrug resistant among Gram positive isolates.

Table 5: Percentage of Antibiotic Susceptibility Pattern of the Isolated *Staphylococcus aureus*

S/n	Antibiotic	Sensitive	Intermediate	Resistant
1	Ciprofloxacin	4 (57.41)	2 (28.57)	1 (14.29)
2	Tetracycline	3 (42.86)	2 (28.57)	2 (28.57)
3	Amoxicillin/clavulanic acid	7 (100.00)	-	-
4	Vancomycin	6 (85.71)	1 (14.29)	-
5	Cotrimoxazole	2 (28.57)	3 (42.86)	2 (28.57)
6	Ceftriaxone	5 (71.43)	2 (28.57)	-
7	Gentamicin	4 (57.41)	0	3 (42.38)
8	Cefoxitin	5 (71.43)	0	2 (28.57)
9	Erythromycin	2 (28.57)	1 (14.29)	4 (51.41)
10	Chloramphenicol	5 (71.43)	2 (28.57)	-

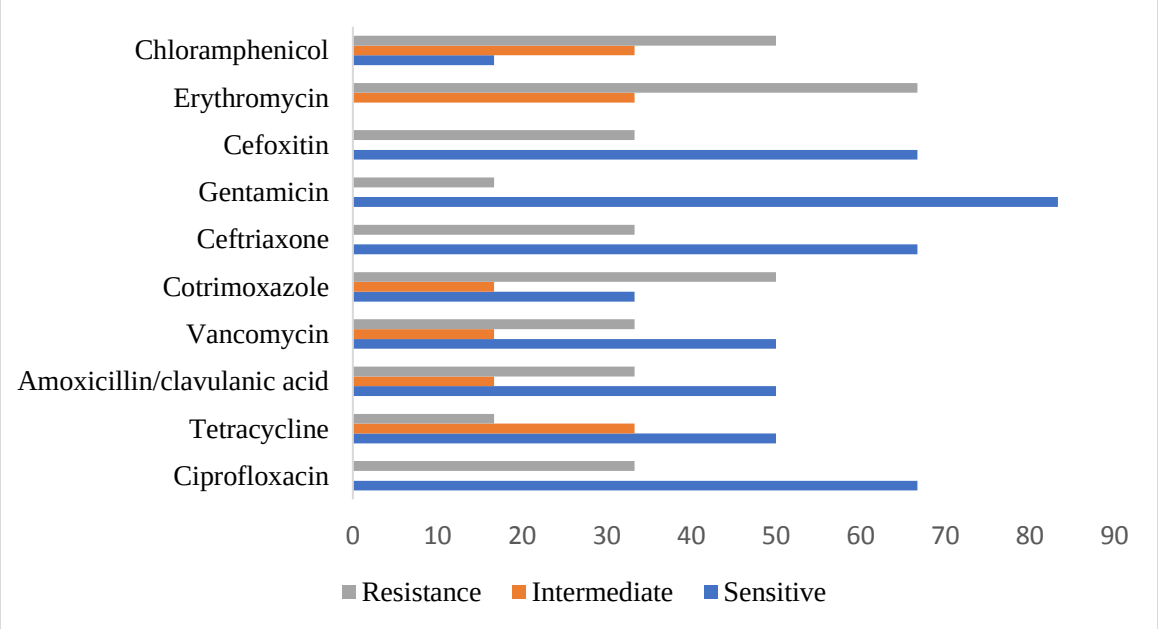

Figure 1: Percentage Antibiotics Susceptibility of Gram-Negative Isolates

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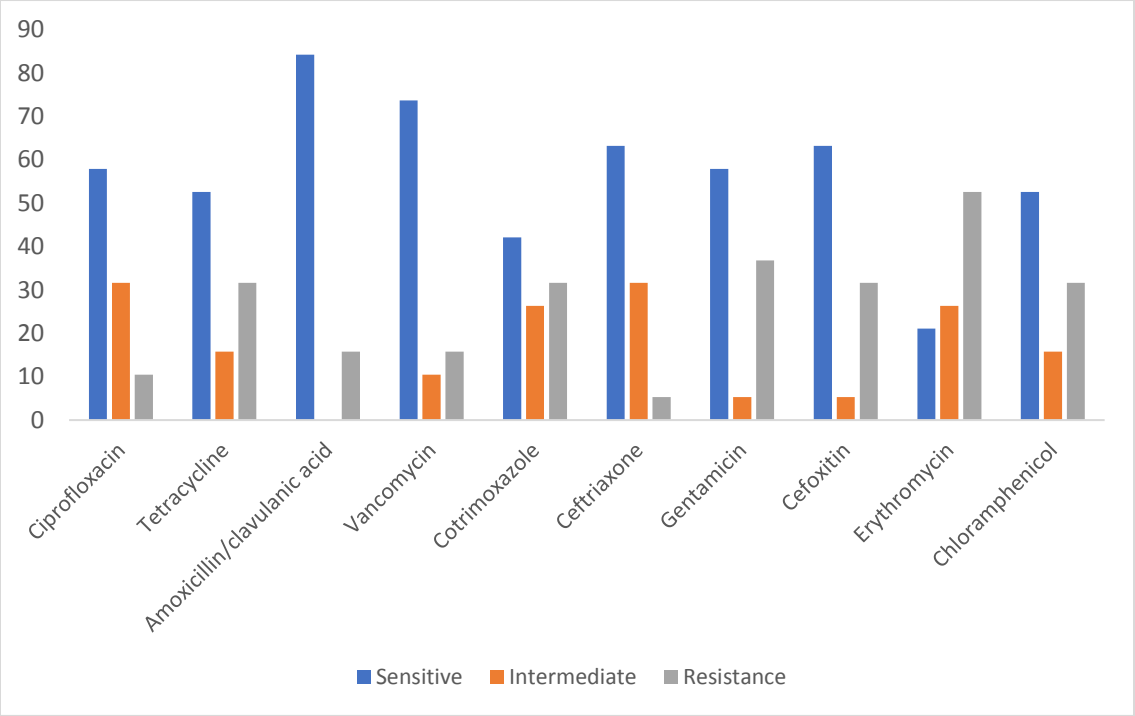

Figure 2: Percentage antibiotics susceptibility of Gram-Positive isolates

DISCUSSION

Microbiological assessment of library indoor air is highly essential considering the vast number of students, lecturers and other users and the implication of bacterial contaminants on their health. In this study, the concentration of bacteria in the various libraries under study ranged from 1.26 x 10^3 to 6.13 x 10^3 CFU/$m^{3.}$ According to Commission of European Community [18], standards for indoor air all the libraries in this study had indoor air bacterial concentration in the range of high and very high. Faculty of Science library had the highest bacterial concentration both in the morning and evening sampling time while Faculty of management Sciences had the least bacterial concentration. The difference in the bacterial concentration at the various libraries under study may be due to the difference in the population of the library users at the specific time. This implies that avoidance of overcrowding in university libraries is necessary to reduce the level of bacterial contamination in the library. The average bacterial concentration observed in this study was higher than that reported by Hayleeyesus and Manaye [19], on microbiological assay of indoor air of Jimma University libraries and that of Adeleye *et al.*,[20] in the study on indoor air quality assessment of Federal University Dutse Library, North West, Nigeria who reported bacterial concentration of 132-345 CFU/m^3.

The bacterial concentration in the afternoon sampling was found to be higher than that of the morning. The reason for this is that the libraries open at 8.00 am and at this time the

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number of users accessing the library at the opening time is small compared with those in the afternoon time. More so, the libraries are usually cleaned in the morning before opening compared with the afternoon after which several users would have accessed the libraries. It can also imply that more activities are carried out in the library in the afternoon compared with the morning time.

Staphylococcus aureus and *Bacillus subtilis* had the highest frequency of occurrence. *S. aureus* is a normal microbial flora while *B. subtilis* is most commonly found in soil environments and on plant undergrowth. *B. subtilis* had also been reported to be a normal gut commensal in humans [21]. Other bacteria isolated were *Streptococcus spp, Micrococcus spp, Pseudomonas aeruginosa* and *Klebsiella pneumoniae*. Only two organisms, *S. aureus* and *B. subtilis* were isolated from Faculty of Management Science and College of Medicine libraries. Previous studies reported isolation of similar bacteria from indoor air of university libraries [19,22,23].

The presence of these bacteria in the indoor air maybe due to some activities in the library which include coughing, sneezing, talking, yawning and movements in the library. The dusts from the atmospheric air, and shoes can also be possible sources. These isolated bacteria are pathogenic and they pose a great health hazard to susceptible humans when inhaled or gained access through the mouth, eyes or nose through touching especially those people whose immune systems are already compromised. *Klebsiella spp* can cause severe infections of the lungs, bladder, brain, liver, eyes, blood and wounds [24].

The various activities observed by the antibiotics in the susceptibility testing was due to their diverse mechanisms of action. Gentamicin was the most active against Gram negative isolates while amoxicillin/clavulanic acid demonstrated the highest activity against Gram positive isolates. Gentamicin is an aminoglycoside which acts by inhibiting protein synthesis by binding to 30S ribosomes [25]. While amoxicillin/clavulanic acid is a combination of amoxicillin and clavulanic acid. Amoxicillin binds to penicillin binding proteins within the bacterial cell wall and inhibits bacterial cell wall synthesis while clavulanic acid inactivates certain beta lactamase enzymes [26].

The Gram negative isolates were more resistant to antibiotics compared with the Gram positive. This was reflected in the fact that 83.3% of Gram negative isolates were multidrug resistant while 47.4% of Gram positive were multidrug resistant. The presence of multidrug resistant strains in the library indoor air is risky. Inhaling these poses a lot of health dangers. This can lead to hard-to treat infections which become resistant to antibiotics, sometimes this resistance can be spread to other bacteria. The presence of outer membrane containing lipopolysaccharide, the peptidoglycan cell wall with peptide chains partially cross-linked serve as a permeability barrier, excluding certain antibiotics from penetrating the cells [27]. The isolation of two MRSA strains phenotypically is also a great concern. MRSA is a cause of Staphylococci infections that is difficult to treat because of resistance to some antibiotics. Such infections include bloodstream infection, pneumonia, surgical site infections [28].

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CONCLUSION/RECOMMENDATIONS

The indoor air of Kaduna State University libraries studied showed high and very high level of bacterial concentration. The following bacteria were isolated from the libraries: *Staphylococcus aureus, Bacillus subtilis, Klebsiella pneumoniae, Streptococcus pneumoniae, Micrococcus* spp and *Pseudomonas aeruginosa*. These are pathogenic organisms. The gram positive isolates were highly susceptible to amoxicillin/clavulanic acid while gentamicin was the most active antibiotic against the gram negative organisms. However, a high percentage of the gram negative organisms were multidrug resistant. Proper and regular cleaning of university libraries is therefore recommended at least twice daily. Proper ventilation and control of population in the libraries is also recommended.

Conflicts of interest: The authors declare no conflicts of interest.

Authors' Contribution: Author 'A' designed the study, performed the statistical analysis, wrote the protocol, and wrote the first draft of the manuscript. Author 'B' managed the literature searches. Author 'C' managed the analysis of the study. All authors read and approved the final manuscript.

REFERENCES

1. Awad AH, Farag SA. An indoor bio-contaminants air quality. Int J Environ Health Res 1999; 9: 313-319.
2. Wood RA, Burchett MD, Orwell RA, Tarran J, Torpy F. Plant/soil capacities to remove harmful substances from polluted indoor air. Plants and environment quality group, Centre for Ecotoxicology, UTS, Australia 2002. (Bezugsquelle: www.plants-for-people.de)
3. Gawronska H. and Bakera B. Phytoremediation of particulate matter from indoor air by *Chlorophytum comosum* L. plants. Air qual Atmos Health 2015; 8: 265 – 272. DOI 10.1007/s11869-014-0285-4
4. Wanner, HU, Verhoff A, Colombi A. Biological particles in indoor environments. European Collaborative Action "Indoor Air quality and its Impact on Man". Commission of the European Communities Report No. 12. 1994; Luxembourg.
5. Srikanth P, Sudharsanam S, Steinberg R. Bioaerosols in indoor environment: composition, health effects and analysis. Indian J. Med. Microbiol. 2008; 26, 302 – 312.
6. Rintala H, Pitkaranta M, Taubel M. Microbial communities associated with house dust. Adv Appl Microbiol. 2102; 78: 75- 120. doi:10.1016/B978-0-12-394805-2.00004-X. PMID: 22305094.
7. Foarde KK, Dulaney P, Cole E, Van Osdell D, Ensor D, Chang J. Assessment of fungal growth on ceiling tiles under environmentally characterized conditions. Indoor Air 1993; 4:357-362.
8. Kalogerakis N, Paschali D, Lekaditis V, Pantidou A, Eleftheriadis K, Lazaridis M. Indoor air quality-bioaerosol measurements in domestic and office premises. J Aerosol Sci 2005; 36: 751-761.
9. Graudenz GS, Oliveira CH, Tribess A, Mendes C Jr, Latorre MR, Kalil J.

Association of air conditioning with respiratory symptoms in office workers in tropical climate. Indoor Air 2005; 15: 62-66.

10. Wamedo SA, Ede PN, Chuku A. Interaction between building design and indoor airborne microbial load in Nigeria. Asian J Biol Sci 2012; 5: 183-191.
11. 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
12. World Health Organization. WHO guidelines for indoor air quality: dampness and mould. Copenhagen, Denmark: World Health Organization; 2009. [Online] Available from: http://www.euro.who.int/__data/assets/pdf_file/0017/43325/E92645.pdf.
13. Cincinelli A and Martellini T. Indoor air quality and health. Int J Environ Res Public Health. 2017; 14(11):1286 doi: 10.3390/ijerph14111286
14. Omeliansky VL. Manual in Microbiology. USSR academy of sciences, Moscow, Leningrad, 1940.
15. Cheesbrough M. Medical laboratory manual for tropical countries. 2nd ed. Cambridge, UK: University Press Cambridge; 2002, p. 508-511.
16. Clinical and Laboratory Standards Institute (2018). Performance standards for antimicrobial susceptibility testing approved standard M100, M02, M07, and M11. 28th Edition. Clinical and Laboratory Standards Institute, Wayne, PA.
17. Magiorakos AP, Srinivasan A, Carey RB, Carmeli Y, Falagas ME, Giske C, Harbarth S. et al. *Bacillus subtilis* isolated from human gastrointestinal tract. Research in Microbiology 2009; 160 (2): 134 – 143.
18. Stryjakowska-Sekulska M., Piotraszewska-Pajuk A., Szyszka A., Nowicki M., Filipak M. Microbiological quality of indoor air in university room. Polish J. of Environ. Stud. 2007;16 (4): 623 – 632
19. Ilusanya OAF, Banjo OA, Orjinta AJ, Oluwole OM, Garuba HM. Microbiological assessment of indoor air quality of science laboratories in Olabisi Onabanjo University, Ago Iwoye, Ogun State, Nigeria. Equity Journal of Science and Technology. 2020; 7 (2): 66-70.
20. Podschun R and Ullmann U. (1998). *Klebsiella* spp as nosocomial pathogen: epidemiology, taxonomy, typing method and pathogenicity factors. Clinical Microbiology Reviews. Oct 589 – 603
21. Kevin T. Fitzgerald (2013). Poisonings in the captive reptile, In Small animal toxicology (3rd Edition).
22. Evans J., Hannoodee M., Wittler M. Amoxicillin clavulanate. In: StatPearls (Internet). Treasure Island: Stat Pearls Publishing, Jan 2021
23. Silhavy TJ, Kahne D., Walker S. The bacteria cell envelope. Cold Spring Harb Perspect. Biol. 2010;2(5): a000414. Doi: 10.1101/cshperspect.a000414
24. Centers for Disease Control and Prevention, Methicillin Resistant Staphylococcus aureus. https://www.cdc.gov/mrsa/index.html