Original Research Article

Biodeterioration of classroom wall surfaces in the University of Port Harcourt, Nigeria

ABSTRACT

Aim: This study investigated the biodeterioration of classroom wall surfaces in the University of Port Harcourt, Nigeria.

Study design: Scrapings from selected classroom wall surfaces were analyzed for their microbiological and physicochemical parameters. Isolated bacteria were screened for their antibiotics susceptibility.

Place and Duration of Study: This study was carried out at the University of Port Harcourt between March - June 2018.

Methods: The population of culturable bacterial and fungal biodeteriogens was determined by plating. Physicochemical parameters were determined using standard methods. Antibiotic susceptibility pattern of the bacterial isolates was determined using the disc diffusion method.

Results: The total culturable heterotrophic bacterial counts ranged from 6.48 to 8.23 log CFU/g while the total fungal counts ranged from 5.00 to 7.28 log CFU/g. The bacterial isolates identified by biochemical characterization and their frequency of occurrence are *Micrococcus* spp. (7.3%), *Citrobacter* spp. (3.2%), *Bacillus* spp. (39.1%), *Serratia* spp. (3.2%), *Corynebacterium* spp. (10.9%), *Staphylococcus aureus* (20.1%), *Proteus* spp. (9.2%) and *Shigella* spp (7.0%). The fungal isolates and their frequency of occurrence are *Aspergillus flavus* (39.1%), *Penicillium* spp. (20.1%), *Microsporium canis* (14.3%), *Coccidioides* spp. (10.9%), *Aspergillus fumigates* (3.2%) and *Tricophyton* spp (3.2%). All antibiotics used showed activity against all bacterial isolates except *Proteus* spp. From the results of the physicochemical parameters, pH values ranged from 6.15 to 9.01, nitrate ranged from 5.30 to 14.83 mg/kg, phosphate ranged 2.19 to 5.94 mg/kg, sulphate ranged from 12.97 to 19.07 mg/kg and Total Organic Carbon ranged from 74.89 to 119.43 mg/kg.

Conclusions: This study has shown the potential public health risk associated with classroom building deterioration owing to the presence of pathogenic microorganisms. Therefore, measures towards prevention and mitigation of classroom building biodeterioration should be in place.

Keywords: Biodeterioration, buildings, public health risk, antibiotics, resistance

1.0 Introduction

Biodeterioration can be defined as undesirable changes to a product or substance or material, influenced by living organisms. Organisms are able to interact with nutrients and material environment to form specific communities. This interaction and association could bring about many physical and chemical destructive processes. Both biotic and abiotic activities contribute simultaneously during the deterioration of building materials. Hence, the level of biodeterioration is difficult to quantify due to the involvement of uncontrollable external (abiotic) factors. However, the involvement of microorganisms in biodeterioration of materials in the environment has been estimated to be up to 30% in the United States [1].

Buildings, just like every other material are subject to microbial colonization, deterioration and degradation or "weathering". Architectural structures including buildings and bridges in contact with water, soil, waste, sewage, plant materials or any organic matter, can undergo deterioration. The hard and firm nature of these structures only limits the biodeterioration process to a slow, eventual and inevitable process of corrosion after microbial colonization, under conducive conditions [2]. The presence of utilizable substrates as part of the building components makes some building more prone to microbial deterioration. For examples, pigment, thinner, binder and drier are the main components of paints used to coat walls, and the most prone to attack by microorganisms [3].

Microorganisms use parts of building components for energy generation [4]. Painted surfaces provide the nutrients and micro-environment for microbial colonization before access to the building proper is later gained. During this attack and colonization, microorganisms produce different forms of corrosive acids which can solubilize the lattice structure [5]. *Bacillus* spp. for example produce sulphuric acid from the oxidation of reduced sulphur compounds [6].

Common building biodeteriogens include nitrifying bacteria, *Cyanobacteria,* and *Thiobacilli* and fungi of genus *Aspergillus, Fusarium Penicillium, Alternaria, Tricophyton* and *Cladosporium* [1,5]. The major environmental parameters affecting biodeterioration are water availability, humility, temperature, UV light and inadequate ventilation [7].

Despite the widespread knowledge of building deterioration, research on biodeterioration is lagging. It is curious though, as studies have pointed to the severe impact of paint components and their degradation products on human health [8,9]. Spoilage of building components come with proliferation of undesirable microorganisms and their degradation products. Consequently, human health and the environment are threatened.

Tropical climate not only impacts on the integrity of structural materials, but it is also critical to the colonization and survival of bioderiogens on these materials [10]. Port Harcourt has a tropical climate. Rainfall is significant most months of the year and the dry season short with little effect. The average annual temperature is 26.4^{o}C and the precipitation averages 2708mm. This study aimed to assess the microbiological and physicochemical properties of deteriorating painted building surfaces of University of Port Harcourt Faculties and the health implication on students.

2.0 MATERIAL AND METHODS

2.1 Collection of Samples

Samples from visibly deteriorating classroom painted building surfaces were collected under aseptic conditions from selected Faculties of University of Port Harcourt. Ten samples from deteriorating buildings and one non-deteriorated building which served as control were taken in triplicates. Samples were gotten by scraping off superficial material to a depth of 2-5 mm.

Samples were moved to the laboratory for immediate analyses. The samples were analyzed for their microbiological and physicochemical parameters.

2.2 Isolation and Enumeration of Bacterial and Fungal Isolates

One (1) g sample of superficial scrapings was transferred into 9 ml sterile normal saline to make a stock solution. One (1) ml was pipette aseptically into a test tube containing 9 ml of normal saline to make 10^{-1} - 10^{-5} dilutions. Nutrient agar (for bacteria) and potato dextrose agar (for fungi) were prepared used for plating out the diluted samples. Triplicate plates were inoculated with 0.1 ml aliquot of each dilution and spread using a flame sterilized hockey stick. Bacterial plates were incubator at 37^0C for 24 hours while fungal plates were incubated at 27^0C for 48-72 hours. The number of colonies that developed from each plate ranging between 30 and 300 after incubation was counted and recorded.

The bacterial isolates were identified based on their cultural and biochemical characteristics with reference to Holt *et al*. [11]. Morphological characteristics such as shape, colour, arrangement of spores, structure of the mycelium, and structure of hyphae and arrangement of sporangiophores were used in identifying the fungal isolates as described in Ellis *et al*. [12].

2.3 Physicochemical Analyses

The pH of building surface was measured *in situ* using a pH meter JENWAY 3071, model pH 82 (degree of accuracy 0.01) equipped with a temperature probe (924001). Determination nitrate, sulphate phosphate and Total Organic Carbon were carried out according to Anyanwu *et al*. [13].

2.4 Bacterial antibiotic susceptibility test

Isolated bacteria were subjected to antibiogram test. Susceptibility test was done using Muller Hinton agar with antibiotics discs effective against gram positive and gram negative bacteria. Following overnight incubation at 37°C, zones of inhibition (ZI) were determined and interpreted as sensitive, intermediate, or resistant for each of the assayed antimicrobial agent. Components of the antibacterial discs used include Erythromycin, Septrin, Ofloxacin, Gentamycin, Ampiclox, Pefloxacin, Amoxacillin, Rocephin, Cirpoflaxicin, Streptomycin and Zinnacef.

2.5 Statistical Analysis

The physicochemical parameters for the different samples were analyzed using one-way Analysis of Variance (ANOVA) with the SPSS vs 20 software.

3.0 RESULTS

3.1 Total Culturable Heterotrophic Bacterial Counts and Fungal Counts

The total culturable heterotrophic bacterial counts and total fungal counts are shown in Figure 1 and 2 respectively. Total culturable heterotrophic bacterial counts from the deteriorating buildings ranged from 6.48 to 8.23 log CFU/g while the control sample (non-deteriorated building) had 3.79 log CFU/g. Total spore counts from deteriorating buildings ranged from 5.00 to 7.28 log cfu/g. Control sample had the least count with 2.92 log CFU/g.

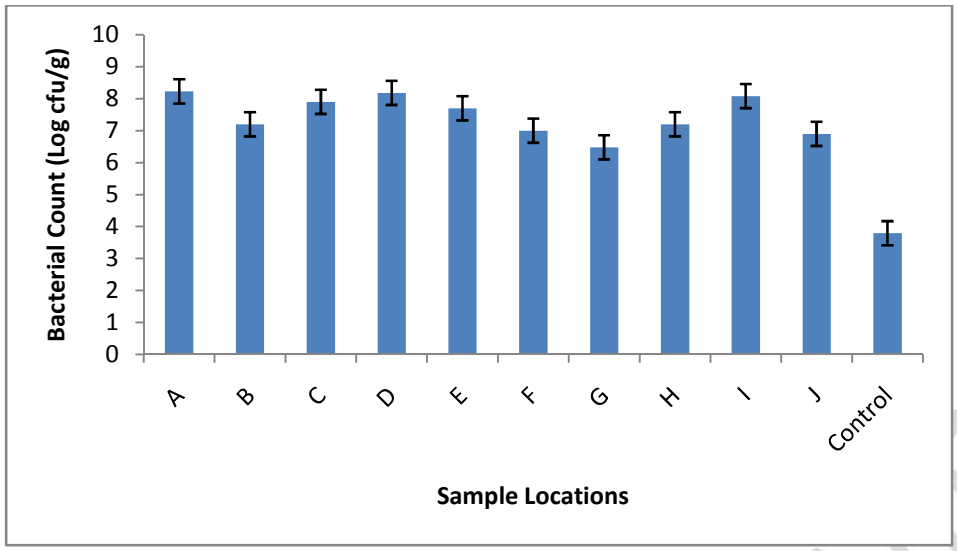

Figure 1: Bacterial counts obtained from classroom wall scrapings

Keys:

A= Dept of Marketing fin lecture Hall 1, B=Dept of crops &soil science, C=Faculty of Social Science, D=Dept of Human Physiology, E=Dept of Economics, F=Dept of Petroleum Engineering, G=Science MBS5, H=Dept of Educational Foundational, I=Dept of Fine Art &Design, J=Dept of Pharmaceutical

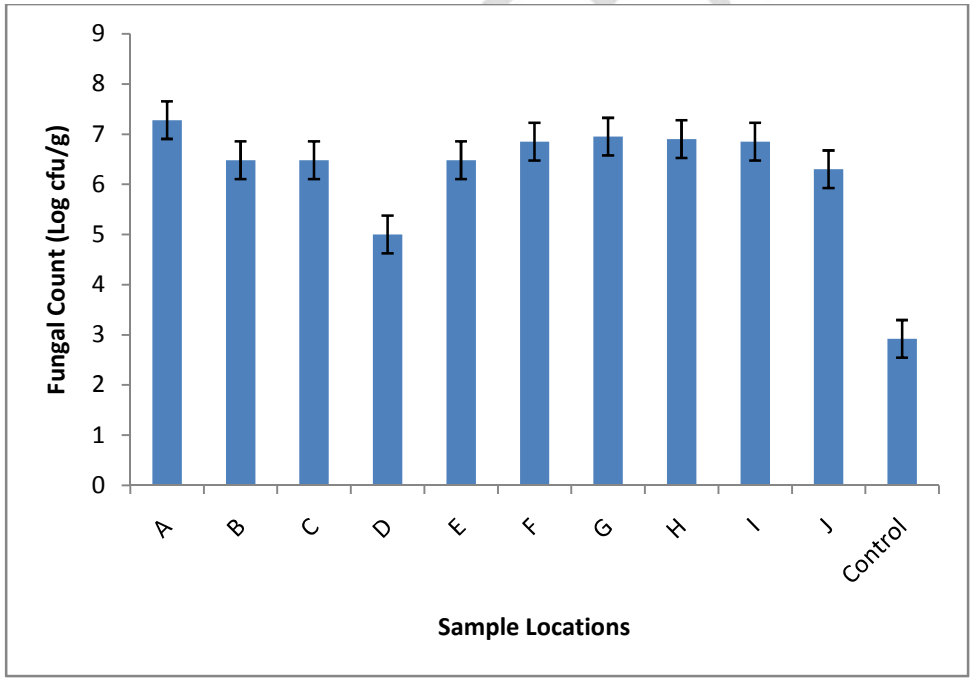

Figure 2: Fungal counts obtained from classroom wall scrapings

Keys:

Idem

3.2 Bacterial and Fungal Biodeteriogens from wall scrapings

The bacterial and fungal biodeteriogens isolated from wall scrapings and their percentage frequencies of occurrence are presented in Tables 1 and 2 respectively. The bacterial biodeteriogens include *Micrococcus* spp. (7.3%), *Citrobacter* spp. (3.2%), *Bacillus* spp. (39.1%), *Serratia* spp. (3.2%), *Corynebacterium* spp. (10.9%), *Staphylococcus aureus*. (20.1%), *Proteus* spp. (9.2%) and *Shigella* spp (7.0%). *Bacillus* spp. were the highest occurring while *Serratia* spp. and *Citrobacter* spp were jointly the least predominant. The fungal biodeteriogens include *Aspergillus flavus* (39.1%), *Penicillium* spp. (20.1%), *Microsporium canis* (14.3%), *Aspergillus fumigates* (3.2%) *Coccidioides* spp. (10.9%) and *Tricophyton* spp. (3.2%). *Aspergillus flavus* was the predominant fungi in the study while *Coccidioides* spp. and *Tricophyton* spp. were the least occurring isolates.

Table 1: Bacterial Biodeteriogens from wall scrapings

Organism	% Frequency
Micrococcus spp.	7.3
Citrobacter spp.	3.2
Bacillus spp.	39.1
Serratia spp.	3.2
Corynebacterium spp.	10.9
Staphylococcus aureus	20.1
Proteus spp.	9.2
Shigella spp.	7.0

Table 2: Fungal Biodeteriogens from classroom wall scrapings

Organism	% Frequency
Aspergillus flavus	39.1
Penicillium spp.	20.1
Microsporium canis	14.3
Aspergillus fumigates	3.2
Coccidioides spp.	10.9
Tricophyton spp.	3.2

3.3 Antibiotic Susceptibility Pattern of Bacterial Isolates

Results of the antibiotic susceptibility pattern of bacterial isolates are shown in Table 3. The antibiotics used in the study include Erythromycin, Septrin, Ofloxacin, Gentamycin, Ampiclox, Pefloxacin, Amoxacillin, Rocephin, Cirpoflaxicin, Streptomycin and Zinnacef. Results of the antibiotic susceptibility pattern revealed susceptibility to the antibiotics by all the test organisms except *Proteus* spp. The antibiotics showed more activity against *Bacillus* spp. and *Citrobacter* spp.

Table 3: Antibiotic Sensitivity Pattern of Bacterial Biodeteriogens of classroom wall scrapings

	Antibiotic / Zone of inhibition (mm)									
Organism	**E**	**SXT**	**PEF**	**CN**	**APX**	**AM**	**R**	**CPX**	**S**	**Z**
Staphylococcus aureus	0	10	0	0	0	0	0	15	10	0
Micrococcus spp.	10	9	4	12	5	0	0	20	15	8
Citrobacter spp.	20	20	20	20	0	0	20	20	20	0
Proteus spp	0	0	0	0	0	0	0	0	0	0
Shigella spp	20	15	24	20	0	0	10	21	20	0
Bacillus spp.	20	20	20	20	20	24	20	20	22	19
Serratia spp	17	17	21	20	0	0	0	20	20	0
Corynebacterium spp.	0	16	0	0	0	0	0	15	18	0

Resistance range 0-13mm, Sensitive range 15mm and above

Keys:
E= Erythromycin, SXT= Septrin, PEF=pefloxacin, CN=Gentamycin, APX=Ampiciox, AM=Amoxacillin, R=Rocephin, CPX=Cirpoflaxicin, S=Streptomycin, Z= Zinnacef.

3.4 Physicochemical Parameters of Deteriorating Buildings

Physicochemical parameters of deteriorating buildings are shown in Table 4. The pH ranged from 6.15 to 9.01, nitrate ranged from 5.30 to 14.83 mg/kg, phosphate ranged 2.19 to 5.94 mg/kg, sulphate ranged from 12.97 to 19.07 mg/kg and Total Organic Carbon ranged from 74.89 to 119.43 mg/kg. Results for control sample (non-deteriorating building) were revealed to be pH 6.69; Nitrate 14.62 mg/kg; Phosphate 6.31 mg/kg; Sulphate 18.05 mg/kg; TOC 125.08 mg/kg. Control sample had higher values for Nitrate, Phosphate, Sulphate and TOC.

Table 4: Physicochemical Parameters of Classroom Wall Scrapings

PARAMETER	A	B	C	D	E	F	G	H	I	J	control
pH	8.47	8.59	8.61	7.94	8.43	7.52	9.01	6.15	8.30	7.55	6.69
Nitrate (mg/kg)	5.94	14.83	10.21	9.86	6.47	11.04	9.08	5.64	5.30	7.01	14.62
Phosphate (mg/kg)	5.89	3.88	2.19	4.62	5.85	5.07	5.94	3.41	3.74	3.88	6.31
Sulphate (mg/kg)	17.32	13.37	15.21	17.82	16.93	13.55	19.07	15.61	12.97	16.40	18.05
TOC (mg/kg)	119.43	74.89	93.60	92.71	103.53	87.65	91.70	109.06	89.51	95.75	125.08

Discussion

The total culturable heterotrophic bacterial counts obtained from deteriorating painted walls ranged from 6.48 to 8.23 log CFU/g while the total fungal counts ranged from 5.00 to 7.28 log CFU/g. The bacterial and fungal populations in the deteriorating buildings were significantly higher than in the non-deteriorated building. The bacterial counts in this study exceeded those reported in a similar study carried out by Shinkafi and Haruna [14], with bacterial counts range of 1.1 x 10^4 CFU/g and 1.20 x 10^5 CFU/g were recorded from buildings showing visibly signs of deterioration. The presence of bacteria on sampled walls might have been influenced by moisture, as seen in areas with visible discoloration and peelings. The moisture was traced to walls outside which were exposed to rainfalls.

Antimicrobial additives in paint formulation are intended to prevent biodeterioration. However, microorganisms have been reported to breakdown preservatives such the biocides used in paints and other paint components such as binders and resin [9]. The quality of biocides used in paints

could be affected by harsh environmental conditions. These environmental conditions could diminish the quality of the paint thereby allowing microorganisms to thrive and colonize these surfaces [15].

From the results of the physicochemical parameter, pH ranged from 6.15 to 9.01, nitrate ranged from 5.30 to 14.83 mg/kg, phosphate ranged 2.19 to 5.94 mg/kg, sulphate ranged from 12.97 to 19.07 mg/kg and TOC ranged from 74.89 to 119.43 mg/kg. The presence of phosphate, sulphate, nitrate and carbon, with pH within the neutral range suggests an appropriate environment for growth. Results of control sample (non-deteriorating building) were revealed to be pH 6.69; Nitrate 14.62 mg/kg; Phosphate 6.31 mg/kg; Sulphate 18.05 mg/kg; TOC 125.08 mg/kg. While the pH was within the pH of the deteriorating surfaces, nitrate phosphate, suphate and TOC were found to be generally higher but not statistically significant. This further suggests that these nutrients were present in higher concentrations until colonization and biodegradation began where the nutrients were utilized. These physicochemical parameters have effect on microbial growth. Warscheid and Braams [16] reported that pH, climatic factors, nutrient sources among others influence microbial colonization of building. The pH range in this study (6.15 to 9.01) was higher than the 3-6 range reported by Ogu *et al*. [15] from deteriorating painted buildings.

The bacterial biodeteriogens were *Micrococcus* spp., *Citrobacter* spp. (3.2%), *Bacillus* spp. (39.1%), *Serratia* spp. (3.2%), *Corynebacterium* spp., *Staphylococcus aureus*, *Proteus* spp., and *Shigella* spp. Similar bacteria were also isolated from painted surfaces in the study of Okpokwasili and Iteun, [17]. In a similar study by Ogu *et al*. [15] *Micrococcus, Bacillus* were isolated from deteriorating walls. Shinkafi and Haruna [14] isolated species of *Bacillus* and *Staphylococcus* from deteriorating wall surfaces.

In the present study, *Bacillus* was the highest occurring bacteria with 39.1%. *Bacillus* spp. are among the most abundant bacteria in the atmosphere [18] as they are spore formers and therefore can withstand adverse environmental conditions. These organisms might have gained their entrances onto painted surfaces through dust, dirt, soot and contaminants accumulating on the painted surfaces, which may also represent another significant source of nutrients to the microorganisms as alluded to by Ogu *et al*. [15].

The fungal biodeteriogens include *Aspergillus flavus*, *Penicillium* spp., *Microsporium canis*, *Aspergillus fumigates*, *Coccidioides* spp. and *Tricophyton* spp. [14,15,19-21] also reported similar fungal genera in their respective studies. Previous studies have largely attributed the colonization of buildings by fungi and subsequent deterioration to moisture [14,21]. Hence, it can be said that fungal development on painted surfaces could imply that moisture is absorbed within the room walls and there is sufficient organic material on the walls to support fungal growth and by extension poses health risk to humans through possible inhalation of those spores.

Fungi just like every other living organism require some sets of conditions to strive. Some of these conditions are optimal temperature, nutrient availability, oxygen and relative humidity. For fungi to conveniently colonize a painted surface, these conditions would have either been provided by the paint or the environment. Their ability to form spores makes them highly resistant to high environmental temperature. According to Milica and Jelena [22] fungi are ideally suited as biodeteriogens of buildings due to their morphology and physiology. This further explains their presence on the sampled walls. Elumalai *et al*. [23] attributed visible discoloration of painted surfaces as signs to possible fungal effect.

Results of the antibiotic susceptibility pattern revealed susceptibility to the antibiotics by all the test organisms except *Proteus* spp. The antibiotics showed more activity against *Bacillus* spp.

and *Citrobacter* spp. It is imperative to add antimicrobial additives to paints to mitigate biodeterioration. It is worrisome however that some of the bacterial isolates exhibited resistance to the antibiotics used. Microorganisms are known to cause sick building illnesses [5] and antibiotic resistant genes can be transferred within this environment to further worse the problem of antibiotics resistance.

Conclusion

This study has shown that bacteria are prevalent in deteriorating buildings suggesting they play a critical role as deteriorating agents. The study also showed the diversity and abundance of microorganisms in the affected buildings. Furthermore, the study revealed the influence of some physicochemical parameters (pH, nitrate, sulphate, phosphate and organic carbon) on the microbial bioburden of painted surfaces. The need to control the colonization and proliferation of microorganisms on building surfaces is emphasized. The university should carryout regular maintenance such as painting of buildings showing signs of deterioration such as discoloration and de-surfacing, so as to prevent possible exposure to toxic biodeterioration products and inhalation of airborne spores.

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