

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

ABSTRACT

This study investigated the biodeterioration of classroom wall surfaces in the University of Port Harcourt, Nigeria. The microbiological and physicochemical parameters of the classroom wall surfaces were determined. The population of culturable bacterial and fungal biodeteriogens was determined by plating. Antibiotic susceptibility pattern of the bacterial isolates was determined using the disc diffusion method. 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. 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

Comment [GQ1]: More information in the introduction section can report weather conditions in that place if there are differences throughout the year.

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*

52 [1,5]. The major environmental parameters affecting biodeterioration are water availability,
53 humidity, temperature, UV light and inadequate ventilation [7].

54 Despite the widespread knowledge of building deterioration, research on biodeterioration is
55 lagging. It is curious though, as studies have pointed to the severe impact of paint components
56 and their degradation products on human health [8,9]. Spoilage of building components come
57 with proliferation of undesirable microorganisms and their degradation products. Consequently,
58 human health and the environment are threatened. This study aimed to assess the microbiological
59 and physicochemical properties of deteriorating painted building surfaces of University of Port
60 Harcourt Faculties and the health implication on students.

61 **2.0 MATERIALS AND METHOD**

62 **2.1 Collection of Samples**

63 Samples from visibly deteriorating classroom painted building surfaces were collected under
64 aseptic conditions from selected Faculties of University of Port Harcourt. Samples were gotten
65 by scraping off superficial material to a depth of 2-5 mm. Samples were moved to the laboratory
66 for immediate analyses.

67 **2.2 Isolation and Enumeration of Bacterial and Fungal Isolates**

68 One (1) g sample of superficial scrapings was transferred into 9 ml sterile normal saline to make
69 a stock solution. One (1) ml was pipette aseptically into a test tube containing 9ml of normal
70 saline to make 10^{-1} - 10^{-5} dilutions. Nutrient agar and potato dextrose agar were prepared used
71 for plating out the diluted samples. Triplicate plates were inoculated with 0.1ml aliquot of each
72 dilution and spread using a flame sterilized hockey stick. Bacterial plates were incubator at 37°C
73 for 24 hours while fungal plates were incubated at 27°C for 48-72 hours. The number of colonies

Comment [GQ2]: How many samples were taken? And how were they identified?

74 that developed from each plate ranging between 30 and 300 after incubation was counted and
75 recorded.

76 The bacterial isolates were identified based on their cultural and biochemical characteristics
77 with reference to Holt *et al.* [10]. Morphological characteristics such as shape, colour,
78 arrangement of spores, structure of the mycelium, and structure of hyphae and arrangement of
79 sporangiophores were used in identifying the fungal isolates as described in Ellis *et al.* [11].

80 2.3 Physicochemical Analyses

81 The pH of building surface was measured in situ using a pH meter JENWAY 3071, model pH 82
82 (degree of accuracy 0.01) equipped with a temperature probe. Determination nitrate, sulphate
83 phosphate and Total Organic Carbon were carried out according to Anyanwu *et al.* [12].

Comment [GQ3]: reference electrode?

84 2.4 Bacterial antibiotic susceptibility test

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

92 2.5 Statistical Analysis

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

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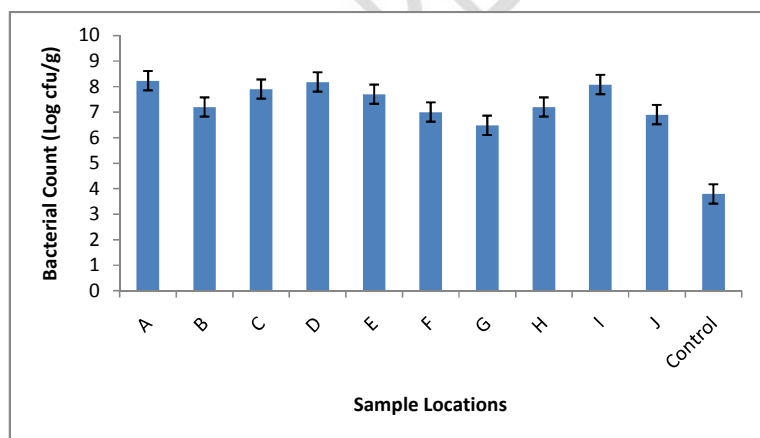
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97 3.0 RESULTS

98 3.1 Total Culturable Heterotrophic Bacterial Counts and Fungal Counts

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

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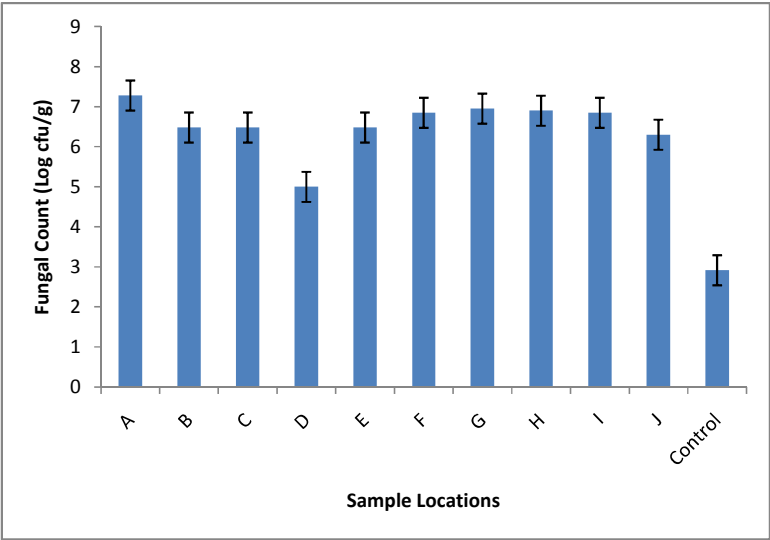


105

106 **Figure 1: Bacterial counts obtained from classroom wall scrapings**

107 **Keys:**

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



112
 113 **Figure 2: Fungal counts obtained from classroom wall scrapings**

114 **Keys:**
 115 Idem

116 **3.2 Bacterial and Fungal Biodeteriogens from wall scrapings**

117 The bacterial and fungal biodeteriogens isolated from wall scrapings and their percentage
 118 frequencies of occurrence are presented in Tables 1 and 2 respectively. The bacterial
 119 biodeteriogens include *Micrococcus* spp. (7.3%), *Citrobacter* spp. (3.2%), *Bacillus* spp. (39.1%),
 120 *Serratia* spp. (3.2%), *Corynebacterium* spp. (10.9%), *Staphylococcus aureus*. (20.1%), *Proteus* spp.
 121 (9.2%) and *Shigella* spp (7.0%). *Bacillus* spp were the highest occurring while *Serratia* spp. and
 122 *Citrobacter* spp were jointly the least predominant. The fungal biodeteriogens include *Aspergillus*
 123 *flavus* (39.1%), *Penicillium* spp. (20.1%), *Microsporium canis* (14.3%), *Aspergillus fumigates*
 124 (3.2%) *Coccidioides* spp. (10.9%) and *Tricophyton* spp. (3.2%). *Aspergillus flavus* was the

125 predominant fungi in the study while *Coccidioides* spp. and *Tricophyton* spp. were the least
126 occurring isolates.

127

128

129 **Table 1: Bacterial Biodeteriogens from wall scrapings**

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

130

131 **Table 2: Fungal Biodeteriogens from classroom wall scrapings**

Organism	% Frequency
<i>Aspergillus flavus</i>	39.1
<i>Penicillium</i> spp.	20.1
<i>Microsporium canis</i>	14.3
<i>Aspergillus fumigates</i>	3.2
<i>Coccidioides</i> spp.	10.9

132

133 **3.3 Antibiotic Susceptibility Pattern of Bacterial Isolates**

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

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

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

142

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

144

145 Keys:

146 E= Erythromycin, SXT= Septrin, PEF=pefloxacin, CN=Gentamycin, APX=Ampiclox, AM=Amoxacillin,

147 R=Rocephin, CPX=Cirpoflaxacin, S=Streptomycin, Z= Zinnacef.

148

149 3.4 Physicochemical Parameters of Deteriorating Buildings

150 Physicochemical parameters of deteriorating buildings are shown in Table 4. The pH ranged
 151 from 6.15 to 9.01, nitrate ranged from 5.30 to 14.83 mg/kg, phosphate ranged 2.19 to 5.94
 152 mg/kg, sulphate ranged from 12.97 to 19.07 mg/kg and Total Organic Carbon ranged from 74.89
 153 to 119.43 mg/kg. Results for Control sample (non-deteriorating building) were revealed to be pH
 154 6.69; Nitrate 14.62 mg/kg; Phosphate 6.31 mg/kg; Sulphate 18.05 mg/kg; TOC 125.08 mg/kg.
 155 Control sample had higher values for Nitrate, Phosphate, Sulphate and TOC.

156 **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

157

158 Discussion

159 This study investigated the biodeterioration of classroom wall surfaces in the University of Port
 160 Harcourt, Nigeria. The total culturable heterotrophic bacterial counts obtained from deteriorating
 161 painted walls ranged from 6.48 to 8.23 log CFU/g while the total fungal counts ranged from 5.00
 162 to 7.28 log CFU/g. The bacterial counts in this study exceeded those reported in a similar study

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163 carried out by Shinkafi and Haruna [13], with bacterial counts range of 1.1×10^4 CFU/g and 1.20
164 $\times 10^5$ CFU/g were recorded from buildings showing visibly signs of deterioration. The presence
165 bacteria on sampled walls might have been influenced by moisture, as seen in areas with visible
166 discoloration and peelings. The moisture was traced to walls outside which were exposed to
167 rainfalls.

168 Antimicrobial additives in paint formulation are intended to prevent biodeterioration. However,
169 microorganisms have been reported to breakdown preservatives such the biocides used in paints
170 and other paint components such as binders and resin [9]. The quality of biocides used in paints
171 could be affected by harsh environmental conditions. These environmental conditions could
172 diminish the quality of the paint thereby allowing microorganisms to thrive and colonize these
173 surfaces [14].

174 From the results of the physicochemical parameter, pH ranged from 6.15 to 9.01, nitrate ranged
175 from 5.30 to 14.83 mg/kg, phosphate ranged 2.19 to 5.94 mg/kg, sulphate ranged from 12.97 to
176 19.07 mg/kg and TOC ranged from 74.89 to 119.43 mg/kg. The presence of phosphate, sulphate,
177 nitrate and carbon, with pH within the neutral range suggests an appropriate environment for
178 growth. Results of Control sample (non-deteriorating building) were revealed to be pH 6.69;
179 Nitrate 14.62 mg/kg; Phosphate 6.31 mg/kg; Sulphate 18.05 mg/kg; TOC 125.08 mg/kg. While
180 the pH was within the pH of the deteriorating surfaces, nitrate phosphate, sulphate and TOC were
181 found to be generally higher but not statistically significant. This further suggests that these
182 nutrients were present in higher concentrations until colonization and biodegradation began
183 where the nutrients were utilized. These physicochemical parameters have effect on microbial
184 growth. Warscheid and Braams [15] reported that pH, climatic factors, nutrient sources among

185 others influence microbial colonization of building. The pH range in this study (6.15 to 9.01) was
186 higher than the 3-6 range reported by Ogu *et al.* [14] from deteriorating painted buildings.

187 The bacterial biodeteriogens were *Micrococcus* spp., *Citrobacter* spp. (3.2%), *Bacillus* spp.
188 (39.1%), *Serratia* spp. (3.2%), *Corynebacterium* spp., *Staphylococcus aureus*, *Proteus* spp., and
189 *Shigella* spp. Similar bacteria were also isolated from painted surfaces in the study of
190 Okpokwasili and Iteun, [16]. In a similar study by Ogu *et al.* [14] *Micrococcus*, *Bacillus* were
191 isolated from deteriorating walls. Shinkafi and Haruna [13] isolated species of *Bacillus* and
192 *Staphylococcus* from deteriorating wall surfaces.

193 In the present study, *Bacillus* was the highest occurring bacteria with 39.1%. *Bacillus* spp. are
194 among the most abundant bacteria in the atmosphere [17] as they are spore formers and therefore
195 can withstand adverse environmental conditions. These organisms might have gained their
196 entrances onto painted surfaces through dust, dirt, soot and contaminants accumulating on the
197 painted surfaces, which may also represent another significant source of nutrients to the
198 microorganisms as alluded to by Ogu *et al.* [14].

199 The fungal biodeteriogens include *Aspergillus flavus*, *Penicillium* spp., *Microsporium canis*,
200 *Aspergillus fumigates*, *Coccidioides* spp. and *Tricophyton* spp. [13,14,18-20] also reported
201 similar fungal genera in their respective studies. Previous studies have largely attributed the
202 colonization of buildings by fungi and subsequent deterioration to moisture [13,20]. Hence, it
203 can be said that fungal development on painted surfaces could imply that moisture is absorbed
204 within the room walls and there is sufficient organic material on the walls to support fungal
205 growth and by extension poses health risk to humans through possible inhalation of those spores.

206 Fungi just like every other living organism require some sets of conditions to thrive. Some of
207 these conditions are optimal temperature, nutrient availability, oxygen and relative humidity. For

208 fungi to conveniently colonize a painted surface, these conditions would have either been
209 provided by the paint or the environment. Their ability to form spores makes them highly
210 resistant to high environmental temperature. According to Milica and Jelena [21] fungi are
211 ideally suited as biodeteriogens of buildings due to their morphology and physiology. This
212 further explains their presence on the sampled walls. Elumalai *et al.* [22] attributed visible
213 discoloration of painted surfaces as signs to possible fungal effect.

214 Results of the antibiotic susceptibility pattern revealed susceptibility to the antibiotics by all the
215 test organisms except *Proteus* spp. The antibiotics showed more activity against *Bacillus* spp.
216 and *Citrobacter* spp. It is imperative to add antimicrobial additives to paints to mitigate
217 biodeterioration. It is worrisome however that some of the bacterial isolates exhibited resistance
218 to the antibiotics used. Microorganisms are known to cause sick building illnesses [5] and
219 antibiotic resistant genes can be transferred within this environment to further worsen the problem
220 of antibiotics resistance.

221 **Conclusion**

222 This study has shown that bacteria are prevalent in deteriorating buildings suggesting they play a
223 critical role as deteriorating agents. The study also showed the diversity and abundance of
224 microorganisms in the affected buildings. Furthermore, the study revealed the influence of some
225 physicochemical parameters (pH, nitrate, sulphate, phosphate and organic carbon) on the
226 microbial bioburden of painted surfaces. The need to control the colonization and proliferation of
227 microorganisms on building surfaces is emphasized.

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Comment [GQ6]: write the conclusion showing some possible solutions

Comment [GQ7]: Check the references

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