African Journal of
Sciences and Traditional Medicine

ISSN : 1595-7942
Index: Harvard, Boston, Sydney University, Dimensions, Lens, ResearchGet Scilit, Semantic, Google Scholar, Base etc

https://doi.org/10.58578/AJSTM.v3i1.9090

Isolation and Identification of Bacteria Species Associated with the Deterioration of Painted Wall Surfaces in Wukari Metropolis, Taraba, Nigeria, North East

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Article Info:

Submitted:	**Revised:**	**Accepted:**	**Published:**
Dec 27, 2025	Jan 20, 2026	Feb 2, 2026	Feb 7, 2026

Abstract

Wall paintings are among the most expressive forms of human creativity, serving not only decorative purposes but also protecting wall surfaces; however, bacterial colonization of water-based paints can lead to biodeterioration that reduces both durability and aesthetic value. This study aimed to identify and characterize the bacterial species responsible for painted wall degradation using combined conventional microbiological and molecular approaches. Samples were collected by scraping portions of intact and deteriorated painted walls, serially diluted, and cultured on nutrient and MacConkey agars to estimate bacterial loads and isolate different species. Identification and characterization of isolates followed a four-step process comprising macroscopic and microscopic observations, biochemical tests, and sugar fermentation assays. Total bacterial counts ranged from 0.2×10^6 to 2.7×10^6 CFU/g, while coliform counts varied between 0.9×10^6 and 3.6×10^6 CFU/g. Both traditional and molecular analyses identified *Bacillus foraminis* (CV53), *Pseudomonas aeruginosa* (PA01), and *Staphylococcus aureus* as key species associated with paint deterioration, with *Bacillus foraminis* (69%) being dominant, followed by *Pseudomonas aeruginosa* (20%) and *Staphylococcus aureus*

(11%). Molecular data further revealed a broader bacterial diversity, including previously unreported taxa, with notable variation among sampling locations. Overall, this study advances the understanding of bacterial ecology on painted surfaces and provides a scientific basis for developing strategies to protect wall paintings and other painted substrates from microbial degradation.

Keywords: Painted Surfaces; Bacterial Biodeterioration; Water-Based Paints; Molecular Techniques; Microorganisms.

INTRODUCTION

Wall paintings represent one of the most prominent forms of figurative art created by humans since prehistoric times (Laura *et al.*, 2022). Their production involves a multi-layered technique comprising a support base, a ground layer, and a paint layer, each evolving through different historical and cultural contexts. Techniques such as secco and fresco were developed to achieve varying artistic effects (Caneva *et al.*, 2019). In the secco method, pigments are applied to dry plaster, remaining on the surface, while in fresco painting, the pigments are added to wet plaster, allowing deeper penetration and stronger adhesion. The pigments used are typically mineral-based, though some may be plant-derived, and organic compounds may be introduced later through restoration processes or other human interventions (Laura *et al.*, 2022). Paint a liquid substance used to protect and decorate surfaces by applying a layer of color. It consists of pigments (colors), binders (polymers or resins), solvents (water, oil, or organic liquids), and additives (thickeners, fillers, or UV stabilizers) (Christiansen *et al.,* 2020).

Paint serves several functions: it adds visual appeal, protects surfaces from corrosion, wear, and environmental degradation, creates a smooth and cleanable finish, and enhances resistance to fading and scratching. Additionally, paint is commonly used as a medium for artistic and creative purposes (Marchant, 2016). Different types of paints exist depending on their composition and application method. These include latex (water-based), oil-based (solvent-based), acrylic, enamel, epoxy, primer, and topcoat varieties (Mendelson, 2019).

Painted surfaces serve multiple purposes: they add aesthetic value through color and texture, provide protection from wear and environmental factors, facilitate cleaning, and can display murals, branding, or decorative designs (Rudgard, 2023). Paint finishes vary widely, including matte, satin, gloss, textured, or patterned types. The selection of paint and finish depends on factors such as the intended look, the nature of the surface, and the surrounding environmental conditions (Marchant, 2016).

Today, numerous paint types are available globally, primarily designed for surface protection and decoration. Paints protect substrates from corrosion and weathering while enhancing aesthetic appeal (Ashwini and Devi, 2018).

Although paints are engineered to protect surfaces from environmental and biological damage, microorganisms often undermine this purpose. These biological agents referred to as deteriogens can degrade the chemical components of paint, leading to reduced durability, weaker adhesion, and loss of decorative appeal (Ogbogu *et al.*, 2017). Microbial activity is influenced by various environmental factors such as humidity, temperature, and pH (Padmini & Kiran, 2020).

Microorganisms can cause several forms of damage to painted surfaces. This includes colour changes, increased porosity, weakening of structural integrity, and the facilitation of moisture penetration. Bacteria can also deteriorate water-based paints, shortening their shelf life and aesthetic value (Maduka, 2019).

The presence of bacteria on painted surfaces can lead to both visible and structural issues. Visually, this includes staining, discolouration, and biofilm formation. Structurally, it can result in cracking, blistering, detachment of paint layers, and the breakdown of binders and adhesives. Over time, structural damage can diminish the overall appearance and integrity of the artwork. Similarly, initial aesthetic damage can often indicate the beginning of more severe underlying deterioration (Fichera *et al.*, 2015). Hence this study is to determine bacteria load present on the deteriorated painted wall surfaces in wukari metropolis, and to isolate and characterize bacteria associated with the deterioration of painted wall surfaces in wukari metropolis using conventional techniques. Given these concerns, it is essential to examine the extent of bacterial degradation affecting painted wall surfaces, particularly in areas like Wukari metropolis.

MATERIALS AND METHODS

Study Area

This research was conducted in Wukari, one of the major towns in Taraba State, Nigeria. The area covers approximately 4,308 km² and, according to the 2006 national census, has a population of about 241,546 people. Wukari lies between latitude 7°51'N and longitude 9°47'E. The vegetation in the region represents the Sudan savannah zone, characterized by grassland interspersed with scattered trees. The area experiences two distinct climatic seasons: a rainy season from April to October and a dry season from November to March, during which rainfall is absent. Wukari is traditionally recognized as the headquarters of the Kwararafa Kingdom, with the Jukun people as the predominant ethnic group, alongside the Hausa and Fulani communities. The major economic activities include farming, involving both subsistence and commercial cultivation of crops such as cereals, tubers, legumes, fruits, and vegetables. Fishing and animal husbandry are also common occupations among the inhabitants (Adesoji & Alao, 2009).

Description of the Sampling Site

The study was carried out in three difference places within Wukari metropolis namely; Government Day Secondary School, Police Barrack and Shagari Quarter all are within Wukari metropolis. Government Day Secondary School Wukari is a public secondary school located along Federal University Teaching Hospita in Wukari, Taraba State, Nigeria. It's a day school that offers secondary education to students and also one of the oldest schools in the area, offering a wide range of academic programs. The Police Barracks in Wukari, Taraba State, Nigeria, is located eastern part of Wukari. Close to rice mill area. It's a police housing facility that provides accommodation for police officers and their families (Ayeni. 2020). Shagari Quarter also known as Shagari estate located along Wukari-Takum road is a residential area with a mix of old and new buildings. The area has seen significant development over the years, with many old houses being renovated or replaced with modern structures. The estates were initially built as low-cost housing schemes by the government (Umar *et al.,*2014).

Materials and Sterilization Procedures.

The materials and equipment used in this study included: a weighing balance, autoclave, conical flasks, microscope, Petri dishes, razor blades, syringes, test tubes, masking tape, glass slides, beakers, Bunsen burner, nutrient media, and antibiotic discs

(Ekeogu, 2024). All glassware (Petri dishes, conical flasks, beakers, and test tubes) were thoroughly washed, air-dried, wrapped in aluminum foil, and sterilized in an autoclave at 121°C for 15 minutes. The work surfaces were disinfected with 70% ethanol using sterile cotton wool before and after use. The inoculating wire loop was sterilized by flaming it to red-hot before and after each inoculation to prevent contamination (Ekeogu, 2024).

Media Preparation

The nutrient agar and MacConkey agar media were prepared following the manufacturer's specifications. Specifically, 28 g of nutrient agar and 56 g of MacConkey agar were each dissolved in 100 ml of distilled water. The mixtures were heated until fully dissolved and then sterilized in an autoclave at 121°C for 15 minutes. After sterilization, the media were cooled to approximately 50°C before being poured aseptically into sterile petri dishes in 20 ml aliquots. The plates were allowed to solidify at room temperature before being used for bacteriological analyses.

Sample Collection

Samples of paint films (scrapings) were collected from sixty (60) buildings comprising 30 normal and 30 deteriorated painted wall surfaces. These samples were obtained from three major locations in Wukari town: Government Day, Police Barracks, and Shagari Quarters.

Buildings were selected based on visible signs of deterioration such as discoloration, loss of gloss, unpleasant odor, and surface degradation. Using new sterile razor blades, paint scrapings were aseptically collected and placed into sterile containers. The samples were immediately transported to the microbiology laboratory for analysis under aseptic conditions.

Determination of Bacterial Load

The bacterial load of each sample was determined using the method described by Ogodo *et al.* (2022). Exactly 1 g of each sample was dissolved in 9 ml of sterile distilled water in a conical flask to form the stock suspension. Serial dilutions were prepared using the ten-fold dilution technique. From the 10^{-5} and 10^{-6} dilutions, 1 ml aliquots were transferred aseptically using a sterile syringe and inoculated onto nutrient agar plates by the spread plate method. The inoculated plates were incubated at 37°C for 24 hours, after which distinct bacterial colonies were counted. The colony-forming units per gram (CFU/g) of sample were calculated using the formula:

$$CFU/g = \frac{Number\ of\ colonies\ counted}{Volume\ of\ inoculum \times dilution\ factor}$$

Isolation of Bacteria

The bacterial colonies that developed from the culture described above, were carefully examined, and distinct colonies were selected for isolation. Each selected colony was subcultured onto freshly prepared nutrient agar plates and incubated at 37°C for 24 hours. To ensure pure cultures, successive streaking was carried out on new nutrient agar plates, following the method described by Agwaranze *et al.* (2018). Each subculture was subsequently incubated at 37°C for 24 hours to obtain pure bacterial isolates.

Identification and Characterization of Isolates

The bacterial isolates were identified and characterized based on their morphological and biochemical properties, following standard microbiological methods (Akinnibosun & Ayejuyoni, 2015).

Morphological characteristics such as colony color, shape, elevation, margin, consistency, and surface appearance were recorded. Biochemical tests conducted included Gram staining, motility test, indole production, urease activity, citrate utilization, glucose/sucrose/lactose fermentation, and catalase test (Akinnibosun & Ayejuyoni, 2015; Williams & Hakam, 2016). The identification of each bacterial isolate was confirmed by comparing the observed characteristics with descriptions provided in Bergey's Manual of Determinative Bacteriology (Williams & Hakam, 2016).

Gram Staining of Isolates

A loopful of the bacterial suspension was transferred onto a clean microscope slide, and a small amount of sterile water was added to make a thin smear approximately 15 mm in diameter. The smear was air-dried and then heat-fixed by gently passing it over a flame.

The fixed smear was covered with crystal violet stain for 60 seconds, then rinsed gently with water. Afterward, Gram's iodine solution was applied for another 60 seconds and washed off with running water. The slide was then decolorized using ethanol (or acetone alcohol) for a few seconds and immediately rinsed again with water.

Subsequently, the smear was counterstained with basic fuchsin for 60 seconds, rinsed, blotted dry with bibulous paper, and air-dried. The slide was examined under a compound microscope using oil immersion (×100 objective lens). Gram-positive bacteria appeared purple, while Gram-negative bacteria appeared pink (Cheesbrough, 2006).

Biochemical Tests

A series of biochemical tests were performed to detect specific enzymatic activities that aid in the identification of bacterial species. The tests included catalase, coagulase, oxidase, indole, citrate utilization, and triple sugar iron (TSI) tests were carried out on the isolates (Ikrimah *et al*., 2025).

RESULTS

Six (60) samples were collected from three different study areas, twenty (20) samples from each study area and the samples were bacteriologically analyzed

Table 1: showing total viable count of Government Day Wukari

Sample area	Samples	Microbial count	cfu/g
GDW	Point A	12	1.2×10^6
	Point B	07	0.7×10^6
	Point C	10	1.0×10^6
	Point D	0	0
	Point E	06	0.6×10^6
	Point F	09	0.9×10^6
	point G	0	0
	Point H	11	1.1×10^6
	Point I	04	0.4×10^6
	Point J	14	1.4×10^6
	Point K	05	0.7×10^6
	Point L	0	0
	Point M	07	0.7×10^6
	Point N	13	1.3×10^6
	Point O	03	0.3×10^6

Sample area	Samples	Microbial count	cfu/g
	Point P	0	0
	Point Q	28	2.8×10^6
	Point R	14	1.4×10^6
	Point S	0	0
	Point T	20	2.1×10^6
	Control	03	0.3×10^6

Key: GDW = Government Day Wukari, cfu = colony forming unit

Table 2: Showing total viable count of police barrack Wukari

Sample area	Samples	Microbial count	cfu/g
PBW	Point A	07	0.7×10^6
	Point B	0	0
	Point C	05	0.5×10^6
	Point D	14	1.4×10^6
	Point E	24	2.4×10^6
	Point F	19	1.9×10^6
	Point G	0	0
	Point H	08	0.8×10^6
	Point I	07	0.7×10^6
	Point J	05	0.5×10^6
	Point k	02	0.2×10^6
	Point L	11	1.1×10^6
	Point M	0	0
	Point N	11	1.1×10^6
	Point O	06	0.6×10^6
	Point P	03	0.3×10^6
	Point Q	13	1.3×10^6
	Point R	0	0

Sample area	Samples	Microbial count	cfu/g
	Point S	02	0.2×10^6
	Point T	12	1.2×10^6
	Control	02	0.2×10^6

Key: PBW= Police Barrack Wukari, cfu = colony forming unit

Table 3: Showing total viable count of shagari quarters Wukari

SQW	Samples	Microbial count	cfu/g
	Point A	06	0.6×10^6
	Point B	0	0
	Point C	10	1.0×10^6
	Point D	09	0.9×10^6
	Point E	07	0.7×10^6
	Point F	12	1.2×10^6
	Point G	0	0
	Point H	27	2.7×10^6
	Point I	07	0.8×10^6
	Point J	02	0.2×10^6
	Point k	0	0
	Point L	08	0.8×10^6
	Point M	11	1.1×10^6
	Point N	0	0
	Point O	14	1.0×10^6
	Point P	12	1.2×10^6
	Point Q	08	0.8×10^6
	Point R	09	0.9×10^6
	Point S	0	0
	Point T	TNTC	0
	Control	02	0.2×10^6

Key: SQW= Shagari quarters Wukari, cfu = colony forming unit, TNTC = too numerous to count

Table 4: Showing total coliform count of government day Wukari

Sample area	Samples	Coliform count	cfu/g
GDW	Point A	17	1.7×10^{6}.
	Point B	27	2.7×10^{6}
	Point C	13	1.3×10^{6}
	Point D	NG	0
	Point E	22	2.2×10^{6}
	Point F	17	1.7×10^{6}
	Point G	NG	0
	Point H	23	2.3×10^{6}
	Point I	26	2.6×10^{6}
	Point J	NG	0
	Point k	27	2.7×10^{6}
	Point L	NG	0
	Point M	12	1.2×10^{6}
	Point N	33	3.3×10^{6}
	Point O	23	2.3×10^{6}
	Point P	NG	0
	Point Q	36	3.6×10^{6}
	Point R	29	2.9×10^{6}
	Point S	NG	0
	Point T	23	2.3×10^{6}
	Control	04	0.4×10^{6}

Key: GDW = Government Day Wukari, cfu = colony forming unit, NG = no growth

Table 5: Showing total coliform count police barracks

Sample area	Sample A	Coliform count	cfu/g
PBW	Point A	29	2.9×10^{6}
	Point B	NG	0
	Point C	12	2.2×10^{6}
	Point D	36	3.6×10^{6}
	Point E	32	3.2×10^{6}

Sample area	Sample A	Coliform count	cfu/g
	Point F	29	2.9×10^6
	Point G	NG	0
	Point H	31	3.1×10^6
	Point I	21	2.1×10^6
	Point J	19	1.9×10^6
	Point k	15	1.5×10^6
	Point L	23	2.3×10^6
	Point M	NG	0
	Point N	32	3.2×10^6
	Point O	11	1.1×10^6
	Point P	09	0.9×10^6
	Point Q	32	3.2×10^6
	Point R	NG	0
	Point S	10	1.0×10^6
	Point T	12	1.2×10^6
	Control	04	0.410^6

Key: PBW = police barracks Wukari, cfu = colony forming unit, NG = no growth

Table 6: Showing total coliform count shagari quarters Wukari

Sample area	Sample A	Coliform count	cfu/g
SQW	Point A	14	1.4×10^6
	Point B	NG	0
	Point C	27	2.7×10^6
	Point D	29	2.9×10^6
	Point E	21	2.1×10^6
	Point F	23	2.3×10^6
	Point G	NG	0
	Point H	13	1.3×10^6
	Point I	28	2.8×10^6

Sample area	Sample A	Coliform count	cfu/g
	Point J	07	0.7×10^6
	Point k	NG	0
	Point L	20	2.0×10^6
	Point M	28	2.8×10^6
	Point N	NG	0
	Point O	30	3.0×10^6
	Point P	29	2.9×10^6
	Point Q	13	1.3×10^6
	Point R	17	1.7×10^6
	Point S	NG	0
	Point T	TNTC	0
	Control	03	0.3×10^6

Key: SQW= Shagari quarters Wukari, cfu = colony forming unit, NG = No growth

Table 7: Showing phenotypic identification of the bacterial isolates

S/N	Morphology	Gram Stain	Catalase Test	Oxidase Test	Indole Test	Citrate Test	Triple Sugar Iron			Coagulase Test	Isolate
							Glu	Lac	Suc		
1.	Large, spherical, moist, raised, and green colour on nutrient agar.	-ve Rod	+	+	-	+	-	-	-	-	*Pseudomonas spp*
2.	Medium, spherical, golden yellow, dried and flat on nutrient agar.	+ve Cocci	+	-	-	+	+	+	+	+	*Staphylococcus aureus*
3.	Creamy, medium, Spherical, raised surface on Mac Conke agar.	+ve Rod	+	-	-	+	+	-	+	-	*Bacillus* spp.

S/N	Morphology	Gram Stain	Catalase Test	Oxidase Test	Indole Test	Citrate Test	Triple Sugar Iron			Coagulase Test	Isolate
							Glu	Lac	Suc		
4.	Creamy, medium, Spherical, raised surface on nutrient agar.	+ve Rod	+	-	-	+	+	-	+	-	*Bacillus* spp.
5.	Large, spherical, moist and raised	-ve Rod	+	-	-	-	+	-	-	-	*Pseudomonas spp*
6.	Medium, spherical, golden yellow, dried and flat.	+ve Cocci	+	-	-	+	+	+	+	+	*Staphylococcus aureus*
7.	Creamy, medium, Spherical, raised surface.	+ve Rod	+	-	-	+	+	-	+	-	*Bacillus* spp.

Key: +ve = Gram positive, -ve = Gram negative, + = Positve, - = negative, Glu = Glucose, Lac = Lactose, Suc = Sucrose

Table 8: Showing the bacteria isolated from samples collected from different sample sites

Bacterial Isolates	Sample Sites
Bacillus foraminis	GDW= Points: B, C, E, F, I, J, K, L, Q, R, T PBW= Points: A, C, D, F, I, J, K, N, O, Q, SQW= Points; C, E, F, H, I, L, M, O, P, Q, R
Pseudomonas aeruginosa	GDW=Points; H, O, M PBW=Points: E, H, P, T SQW=Points: D, J
Staphylococcus aureus	GDW=Point: A PBW= Points L, S SQW= Point A

Key: GDW= Government Day Wukari, PBW= Police Barrack wukari, SQW= Shagari Quarter Wukari.

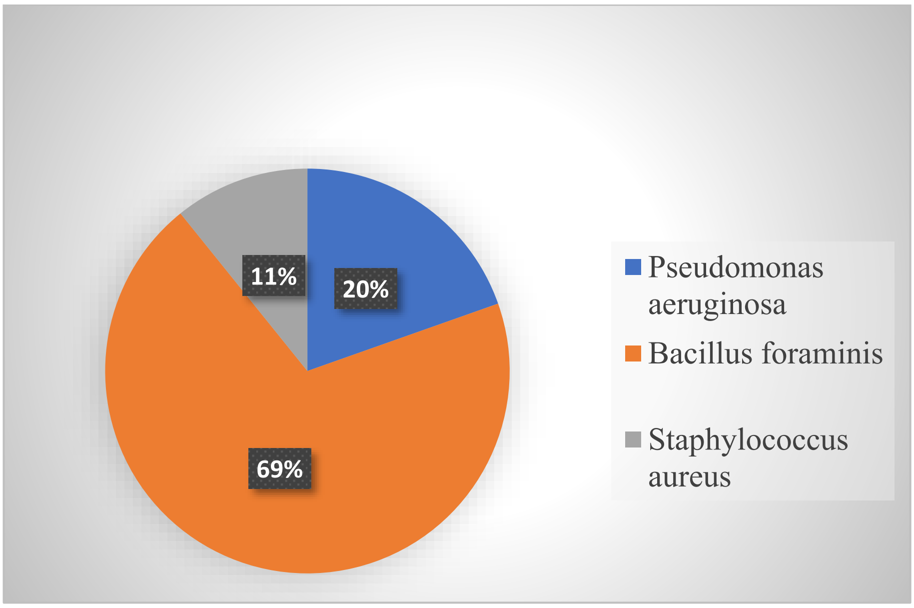

Figure 1: presents the percentages of occurrences of isolate from different samples

DISCUSSION

In this study, the total bacterial count obtained in this study ranged from 0.2×10^6 to 3.6×10^6 CFU/g, which aligns with the results reported by Olayide and Faluke (2017), who recorded microbial densities between 0.02×10^2 and 3.3×10^3 CFU/g on painted walls. The high microbial population density and the heavy pigmentation observed in this research explain the visible discoloration and aesthetic degradation of painted surfaces. Ekeogu (2024) similarly observed that high bacterial loads on painted surfaces could be influenced by environmental factors such as dust accumulation, moisture infiltration, and rainfall conditions. These observations also support the findings of Elumalai *et al.* (2020).

In this study, three major bacterial species; *Bacillus foraminis, Pseudomonas aeruginosa*, and *Staphylococcus spp*. were consistently isolated from deteriorated painted walls in various locations within Wukari metropolis. Among these, *Bacillus foraminis* was the most frequently detected. The isolation of these organisms confirms that microbial deterioration of painted walls is a significant environmental issue affecting both the aesthetics and durability of buildings.

Bacillus foraminis has been widely reported for its ability to degrade organic materials through enzymatic activity, breaking down pigments and binders within paints (Lee *et al.,* 2021). These bacteria can survive in nutrient-poor and humid environments, making them highly adaptable to painted surfaces exposed to moisture. Similarly, *Pseudomonas aeruginosa* is known for its metabolic versatility and biofilm formation capability, which enables it to trap moisture and create favorable conditions for further microbial growth (Rodrigues *et al.,* 2020). The production of secondary metabolites by *Pseudomonas* species can also accelerate paint degradation. Staphylococcus species, though more commonly associated with human and environmental surfaces, contribute to deterioration by forming biofilms and interacting synergistically with other bacterial species (Wang *et al.,* 2022).

Molecular characterization of the bacterial isolates confirmed their genetic identities, offering insights into their roles in the biodeterioration process. The genetic analysis revealed that the Bacillus foraminis isolates showed high similarity with known *Bacillus* strains, suggesting a close phylogenetic relationship. Similarly, *Pseudomonas aeruginosa* isolates displayed significant genetic correspondence with previously characterized sequences, indicating reliability and accuracy in molecular identification (Bhutia *et al.,* 2021). These findings are consistent with Griffiths *et al.* (2000), who linked the presence of *Pseudomonas* species on painted surfaces to moisture damage and high humidity conditions.

The percentage similarity of the sequences, ranging from 98.10% to 99.04%, further validated the precision of the molecular approach used. These results demonstrate that the isolated bacterial strains share close genetic relationships with known reference organisms, supporting the reliability of 16S rRNA sequencing as a diagnostic tool. Molecular techniques not only confirmed bacterial presence but also provided deeper insights into their genetic diversity, evolutionary relationships, and potential environmental adaptations (Shinkafi and Haruna, 2013).

The molecular data obtained from this study serve as a baseline for future research on microbial communities associated with painted surfaces. Understanding their genetic diversity and metabolic potential could inform the development of preventive and control strategies against biodeterioration. As highlighted by Pérez *et al.* (2019), mixed bacterial communities tend to degrade materials more rapidly due to their synergistic enzyme production. This observation aligns with the findings of the present study, suggesting that the coexistence of Bacillus, Pseudomonas, and Staphylococcus species likely contributes to accelerated paint degradation. Environmental factors such as temperature and humidity further influence bacterial colonization, as demonstrated by Zhang *et al.* (2020), underscoring the importance of environmental management in mitigating microbial damage.

CONCLUSION

The study demonstrates the significant role of bacterial communities particularly *Bacillus foraminis, Pseudomonas aeruginosa*, and *Staphylococcus* spp. in the deterioration of painted wall surfaces in Wukari, Taraba State. The combined conventional and molecular approaches provided a comprehensive understanding of the organisms involved and their potential impact on paint integrity. The findings highlight that microbial-induced deterioration results from both environmental and biological factors. Effective control therefore requires integrated strategies that include improved paint formulations, environmental management, and routine maintenance. This research contributes valuable information to the growing body of knowledge on microbial biodeterioration and its implications for building preservation.

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