www.ijarp.com ISSN 2456-9992 Page: 01-09

International Journal Advanced Research Publications

Review Article

Volume: 02
Issue: 07

BIOFILM FORMATION AND ANTIBIOTIC RESISTANCE IN STAPHYLOCOCCUS AUREUS: A REVIEW OF ORTHOPAEDIC AND CLINICAL PERSPECTIVES

Nutan Kumari*[1], Dr Rajender Kumar[2], Ramandeep Kaur[3], Dr Praveen Kumar[3]

[1]Faculty of Health and Allied Sciences, Guru Kashi University.

[2]Dean, Faculty of Allied and Healthcare, Guru Kashi University.

[3]Assistant Professor, Faculty of Allied and Healthcare, Guru Kashi University.

Article Received: 19 June 2026, Article Revised: 09 July 2026, Published on: 29 July 2026

***Corresponding Author: Nutan Kumari**

Faculty of Health and Allied Sciences, Guru Kashi University.

Doi: https://doi-doi.org/101555/ijarp.1415

ABSTRACT

Staphylococcus aureus is a leading cause of healthcare-associated infection, distinguished by its capacity to form biofilms, acquire multi-drug resistance, and generate small-colony variants (SCVs) that support persistent infection. This review synthesises the microbiological literature on S. aureus virulence, biofilm biology, and antibiotic resistance underlying a comparative thesis study of 189 clinical samples (wound, nasal, bed, and urine) from 49 orthopaedic in-patients at the National Orthopaedic Hospital Dala (NOHD), Kano, Nigeria, which confirmed 28 S. aureus isolates (14.3% isolation rate) and reported 96.4% biofilm production, 67.9% MRSA prevalence, and 85.7% multi-drug resistance (MDR), together with molecular detection of icaA, bbp, mecA, and vanA genes. The review traces S. aureus surface adhesins (MSCRAMMs) and toxin-mediated virulence, the staged process of biofilm formation via the icaADBC operon, the genetic basis of methicillin resistance (mecA/SCCmec) and vancomycin resistance (VISA/VRSA via cell-wall thickening and the vanA operon), and the clinical significance of small-colony variants, inducible clindamycin resistance, and mupirocin resistance for MRSA decolonisation. A comparative synthesis of nine published donor and patient studies from Nigeria, Ethiopia, and India shows wide regional variation in MRSA and MDR prevalence, generally attributable to local antibiotic-use pressure and infection-control practice. The review concludes that biofilm formation and multi-drug resistance should be treated as distinct, independently arising therapeutic challenges, and that antibiotic stewardship, routine D-testing, and vanA surveillance are priority measures for

orthopaedic infection control.

KEYWORDS: Staphylococcus aureus; biofilm; MRSA; VRSA; multi-drug resistance; small-colony variants; icaA; mecA; vanA; orthopaedic infection.

1. INTRODUCTION

Staphylococcus aureus is a Gram-positive, catalase-positive, facultative anaerobic coccus that colonises human skin and the anterior nares and causes a wide spectrum of disease, from minor skin abscesses to osteomyelitis, bacteraemia, and necrotising pneumonia (Bhattacharyya et al., 2012; Asadi and Jamali, 2017). Its clinical importance rests on three intersecting capabilities: an extensive repertoire of adhesins and toxins that mediate tissue invasion and immune evasion; the ability to form biofilms that are markedly more resistant to antibiotics than free-living (planktonic) cells; and a remarkable capacity to acquire antibiotic resistance genes via mobile genetic elements, exemplified by methicillin-resistant S. aureus (MRSA) and, more recently, vancomycin-resistant strains (VRSA).

These properties converge with particular severity in orthopaedic infection, where implanted hardware and exposed bone tissue provide surfaces for biofilm colonisation and where the bone-sialoprotein-binding adhesin (bbp) confers a specific tropism for bone. This review consolidates the literature on S. aureus biology, biofilm formation, and antibiotic resistance mechanisms, using as its empirical anchor a comparative thesis study of orthopaedic S. aureus isolates from a Nigerian tertiary hospital, and situates its findings — high biofilm production, high MRSA and MDR prevalence, and detection of the vanA resistance gene — within the wider regional and international evidence base.

2. Staphylococcus aureus: Structure, Virulence, and Pathogenesis

S. aureus pathogenesis follows a staged sequence of colonisation, immune evasion, tissue invasion, and toxin-mediated damage (Asadi and Jamali, 2017). Adhesion to host tissue and extracellular matrix is mediated by Microbial Surface Components Recognising Adhesive Matrix Molecules (MSCRAMMs), including clumping factors ClfA/ClfB (fibrinogen-binding), fibronectin-binding proteins FnBPA/FnBPB (which also mediate biofilm accumulation in MRSA), serine-aspartate repeat proteins SdrC/D/E (associated with bone infection; Ajayi et al., 2018), and the bone-sialoprotein-binding protein bbp, which is of particular relevance in orthopaedic disease. Protein A contributes to immune evasion by binding the Fc region of IgG and blocking antibody-mediated opsonisation. Toxin-mediated virulence includes alpha-haemolysin (Hla), a haemolytic, cytotoxic, and dermonecrotic pore-

forming toxin; Panton-Valentine leukocidin (PVL), associated with severe necrotising pneumonia in community-associated MRSA (Kitur et al., 2015); and superantigens such as TSST-1, which cause toxic shock syndrome.

3. Biofilm Formation: Mechanism and Clinical Significance

A biofilm is a community of micro-organisms adhering to a surface within a self-produced matrix of extracellular polymeric substances comprising polysaccharides, proteins, and nucleic acids (Jamal et al., 2015; Fong and Yildiz, 2015). Biofilm formation proceeds through four recognised stages: reversible attachment mediated by weak physical forces and MSCRAMMs; irreversible attachment via specific adhesins including biofilm-associated protein (Bap); microcolony formation, driven by synthesis of polysaccharide intercellular adhesin (PIA) through the icaADBC operon; and biofilm maturation with formation of a three-dimensional structure and eventual dispersal of cells to colonise new sites. Bacteria within a mature biofilm are 10–1,000-fold more resistant to antibiotics and disinfectants than planktonic cells (Sharma et al., 2019), through reduced antibiotic penetration of the exopolysaccharide matrix, slowed growth in deeper layers, spatial nutrient/oxygen gradients, and expression of biofilm-specific resistance determinants (Gilbert et al., 2002). In the reviewed thesis, icaA was detected by PCR in 30% of tested isolates and bbp — the bone-adhesion gene of specific relevance to orthopaedic infection — in 37.5%, directly linking genotype to the orthopaedic clinical setting.

4. Antibiotic Resistance: MRSA, VISA/VRSA, and Multi-Drug Resistance

Methicillin resistance arises from the mecA gene, carried on the mobile Staphylococcal Cassette Chromosome mec (SCCmec, 21–67 kb), which encodes penicillin-binding protein 2a (PBP2a) — a transpeptidase with markedly reduced affinity for all beta-lactam antibiotics, conferring cross-resistance to the entire class (Loomba et al., 2010). Vancomycin resistance follows a graded pathway: vancomycin-intermediate S. aureus (VISA) arises through progressive cell-wall thickening that traps the antibiotic before it reaches its target (Cui et al., 2003), while full vancomycin-resistant S. aureus (VRSA) results from acquisition of the vanA operon, which redirects peptidoglycan precursor synthesis to a form (D-Ala-D-Lac) that vancomycin cannot bind (Chang et al., 2003; Courvalin, 2006). In the reviewed thesis, vanA was detected in 20% of tested MDR isolates — a finding of considerable clinical concern given that vancomycin remains, in most settings, the drug of last resort for MRSA.

Multi-drug resistance (MDR) is internationally defined as non-susceptibility to at least one

agent in three or more antimicrobial categories, with extensively drug-resistant (XDR) and pan-drug-resistant (PDR) representing progressively narrower remaining treatment options (Magiorakos et al., 2012). Any MRSA isolate is, by definition, automatically classified as MDR. The Multiple Antibiotic Resistance (MAR) index — the proportion of antibiotics to which an isolate is resistant — provides a simple proxy for the antibiotic-use pressure of the environment from which an isolate was recovered; a MAR index above 0.2 is considered indicative of a high-antibiotic-use setting (Bauer et al., 1966).

5. Small-Colony Variants, Inducible Clindamycin Resistance, and Mupirocin Resistance

Small-colony variants (SCVs) are slow-growing S. aureus subpopulations with atypical colony morphology that favour intracellular persistence and are strongly associated with chronic, relapsing infection, particularly in cystic fibrosis and chronic osteomyelitis (Proctor et al., 2014; Tuchscherr et al., 2016). SCV formation is specifically linked to exposure to aminoglycosides and trimethoprim-sulfamethoxazole, which select for hemin/menadione- or thymidine-auxotrophic variants respectively (Wolter et al., 2013); their reported prevalence varies widely across settings, from 16.2% (Yagci et al., 2013) to 21.9% (Ode et al., 2015), and appears closely tied to local antibiotic prescribing patterns rather than being a fixed organism property.

Inducible clindamycin resistance (ICR), mediated by the erm gene family, causes an MLSBi phenotype that can switch to constitutive resistance (MLSBc) during treatment, leading to clinical failure if undetected; the D-zone disc test is the standard method for its detection (Ahmed et al., 2010). Because clindamycin has excellent bone penetration, it remains an attractive option for orthopaedic MRSA infection provided D-testing confirms susceptibility, potentially reducing reliance on vancomycin. Mupirocin resistance is of separate clinical concern because mupirocin-based nasal decolonisation is a standard pre-operative infection-control measure in orthopaedic surgery; high-level resistance (mediated by the mupA gene) renders decolonisation protocols ineffective and necessitates alternative strategies such as chlorhexidine bathing or topical fusidic acid/retapamulin (Park et al., 2015).

6. Comparative Evidence from Clinical Studies

Table 1 compares S. aureus isolation, MRSA, MDR, and biofilm findings across published studies from Nigeria, Ethiopia, and India alongside the reviewed thesis, illustrating substantial regional variability driven by antibiotic-use patterns, infection-control practice,

and specimen source.

Table 1. Comparative findings on S. aureus isolation, resistance, and biofilm formation across published clinical studies.

Study	Setting	n	S. aureus / MRSA (%)	MDR (%)	Biofilm (%)	Key note
Nwankwo et al., 2010	Kano, Nigeria (in-patients)	—	MRSA 62.0	—	—	Earlier Kano MRSA baseline
Udobi et al., 2013	Kaduna, Nigeria (orthopaedic wounds)	—	MRSA 75.0	—	—	Ampicillin resistance 100%
Taddesse, 2014	N. Ethiopia (clinical isolates)	—	—	—	—	Vancomycin resistance 0%
Kumurya, 2015	Kano, Nigeria	—	—	—	—	ICR 18.1%; E/DA resistance 46.9%
Dilnessa & Bitew, 2016	Addis Ababa, Ethiopia	—	Isolation 14.3	—	—	Nasal swabs most prevalent (33.3%)
Basak et al., 2016	India (hospital isolates)	—	—	37.1	—	XDR 13.8%; no PDR detected
Jayakumar et al., 2013	India (dermatology)	—	—	—	—	Mupirocin resistance: 2.0% high, 1.3% low
Ibrahim et al., 2018	Kano, Nigeria (AKTH)	—	—	—	—	Wound swabs highest prevalence (32.7%)
Present study (reviewed thesis), 2026	NOHD, Kano, Nigeria (orthopaedic in-patients)	189 samples / 28 isolates	Isolation 14.3 / MRSA 67.9	85.7	96.4	vanA in 20% of tested MDR isolates

The reviewed thesis's MRSA prevalence (67.9%) sits between the 62% reported earlier in Kano (Nwankwo et al., 2010) and the 75% reported in orthopaedic wards in Kaduna (Udobi et al., 2013), while its MDR rate (85.7%) substantially exceeds the 37.1% reported from India (Basak et al., 2016), though the XDR rate (14.3%) is comparable (13.8%). Isolation-source patterns also vary regionally: wound swabs yielded the highest S. aureus prevalence in both the reviewed thesis and in Ibrahim et al. (2018) from the same city, whereas nasal swabs were the leading source in Addis Ababa (Dilnessa and Bitew, 2016) — a divergence that may reflect differing local colonisation ecology rather than methodological differences.

7. Discussion: Clinical and Public Health Implications

Two structural findings recur across the reviewed literature and the thesis data. First, biofilm formation and multi-drug resistance, while both prevalent, behave as statistically independent

traits (as confirmed by chi-square testing in the reviewed thesis, $p > 0.05$), meaning effective orthopaedic infection management must address them as separate problems: antibiotic stewardship to curb MDR selection, and biofilm-targeted strategies (mechanical debridement, biofilm-disrupting adjuncts such as DNase or dispersin B) to address biofilm-associated persistence, since conventional antibiotic dosing alone under-treats biofilm-embedded organisms by one to three orders of magnitude (Sharma et al., 2019).

Second, the mismatch between phenotypic and genotypic resistance testing warrants caution in molecular-only surveillance: in the reviewed thesis, several phenotypically MRSA isolates were mecA-negative, suggesting beta-lactamase hyper-production or other resistance mechanisms, while 20% of tested MDR isolates carried vanA — a resistance determinant that, if it converts to phenotypic resistance, would remove vancomycin as a therapeutic option. This underscores the importance of pairing phenotypic susceptibility testing (disc diffusion, D-test, dual mupirocin testing) with targeted molecular surveillance rather than relying on either method alone.

8. CONCLUSION AND RECOMMENDATIONS

The evidence reviewed here, read alongside the reviewed thesis's findings (96.4% biofilm formation, 67.9% MRSA, 85.7% MDR, and vanA detection in 20% of tested MDR isolates among orthopaedic S. aureus in Kano, Nigeria), confirms that biofilm-forming, multi-drug-resistant S. aureus represents a substantial and likely under-addressed threat in orthopaedic hospital settings, with considerable regional variation in severity linked to local antibiotic-use pressure and infection-control practice.

Priority measures supported by the literature include: (1) implementing formal antibiotic stewardship programmes to reduce the selective pressure driving MDR and MAR index elevation; (2) routine D-testing for inducible clindamycin resistance before prescribing clindamycin; (3) evaluating alternative MRSA decolonisation strategies where mupirocin resistance is prevalent; (4) instituting molecular vanA surveillance in MDR isolates, with immediate infection-control response on detection; (5) strengthening environmental decontamination given the documented role of contaminated surfaces (bed swabs) as MRSA reservoirs; and (6) incorporating biofilm-targeted adjunctive therapy into treatment protocols for implant-associated and chronic orthopaedic infections.

REFERENCES

1. Ahmed, M.O. et al. (2010). Detection of inducible clindamycin resistance (MLSBi) among MRSA from Libya. The Libyan Journal of Medicine, 5.
2. Ajayi, C. et al. (2018). Genetic variability in the sdrD gene in Staphylococcus aureus from healthy nasal carriers. BMC Microbiology, 18, 34.
3. Asadi, S. and Jamali, M. (2017). Assessment the Frequency of MRSA and VRSA using E-Test. Immunological Disorders and Immunotherapy, 2(1), 1000104.
4. Basak, S. et al. (2016). Multidrug resistant and extensively drug resistant bacteria: A study. Journal of Pathogens.
5. Bauer, A.W. et al. (1966). Antibiotic susceptibility testing by standardized single disk method. American Journal of Clinical Pathology, 45, 493–496.
6. Bhattacharyya, D. et al. (2012). Characterization of Staphylococcus aureus: a review. [Cited in thesis introduction].
7. Chang, S. et al. (2003). Infection with vancomycin-resistant Staphylococcus aureus containing the vanA resistance gene. New England Journal of Medicine, 348, 1342–1347.
8. Courvalin, P. (2006). Vancomycin Resistance in Gram-Positive Cocci. Clinical Infectious Diseases, 42, 25–34.
9. Cui, L. et al. (2003). Cell wall thickening is a common feature of vancomycin resistance in S. aureus. Journal of Clinical Microbiology, 41, 5–14.
10. Dilnessa, T. and Bitew, A. (2016). Prevalence and AMR Pattern of Methicillin Resistant S. aureus from Addis Ababa. BMC Infectious Diseases, 16, 398.
11. EUCAST (2018). Breakpoint tables for interpretation of MICs and zone diameters, Version 8.0. www.eucast.org.
12. Fong, J.N. and Yildiz, F.H. (2015). Biofilm matrix proteins. Microbiology Spectra, 3, 1–27.
13. Gilbert, P. et al. (2002). Biofilms in vitro and in vivo: do singular mechanisms imply cross-resistance? Symposium Series (Society for Applied Microbiology), 92, 98–110.
14. Holden, M.T. et al. (2014). Complete genomes of two clinical S. aureus strains: rapid evolution of virulence and drug resistance. PNAS, 101(26), 9786–9791.
15. Ibrahim, S. et al. (2018). Prevalence and Antibiotic Sensitivity Pattern of S. aureus from Aminu Kano Teaching Hospital. Microbiology Research Journal International, 1–9.
16. Jamal, M. et al. (2015). Bacterial Biofilm: Its Composition, Formation and Role in

Human Infections. Journal of Microbiology and Biotechnology, 3.

17. Jayakumar, S. et al. (2013). Prevalence of high- and low-level mupirocin resistance among Staphylococcus spp. Journal of Clinical Diagnostic Research, 7(2), 238–242.
18. Kitur, K. et al. (2015). Toxin-induced necroptosis is a major mechanism of S. aureus lung damage. PLoS Pathogens, 11(4), e1004820.
19. Kumurya, A.S. (2015). Detection of Inducible Clindamycin Resistance among Staphylococcus Isolates in North-western Nigeria. International Journal of Preventive Medicines Research, 1(2), 35–39.
20. Loomba, P.S. et al. (2010). Methicillin and Vancomycin Resistant S. aureus in Hospitalized Patients. Journal of Global Infectious Diseases, 2(3), 275–283.
21. Magiorakos, A.P. et al. (2012). Multidrug-resistant, extensively drug-resistant and pandrug-resistant bacteria: an international expert proposal for interim standard definitions. Clinical Microbiology and Infection, 18, 268–281.
22. McGuinness, W.A. et al. (2017). Vancomycin Resistance in Staphylococcus aureus. Yale Journal of Biology and Medicine, 90, 269–281.
23. Mustafi, H.I.S. (2014). S. aureus can Produce Catalase Enzyme when React with Human WBCs. Journal of Medical Microbiology and Diagnosis, 3, 4.
24. Nwankwo, E.O.K. et al. (2010). Methicillin resistant S. aureus (MRSA) and antibiotic sensitivity in Kano. African Journal of Clinical and Experimental Microbiology, 11(1), 129–136.
25. Ode, R.O. et al. (2015). Characterization of S. aureus SCV from ABU Teaching Hospital, Zaria. 13th National Conference, Nigeria Association of Pharmacists in Academia.
26. Park, S.H. et al. (2015). In vitro antimicrobial activities of fusidic acid and retapamulin against mupirocin- and methicillin-resistant S. aureus. Annals of Dermatology, 27(5), 551–556.
27. Proctor, R.A. et al. (2014). S. aureus SCVs: A Road Map for the Metabolic Pathways in Persistent Infections. Frontiers in Cellular and Infection Microbiology, 4, 99.
28. Sharma, D. et al. (2019). Antibiotics versus biofilm: an emerging battleground. Antimicrobial Resistance & Infection Control, 8(1), 76.
29. Siddiqui, A.H. and Koirala, J. (2018). Methicillin Resistance Staphylococcus aureus. StatPearls Publishing, Treasure Island (FL).
30. Stepanovic, S. et al. (2007). Quantification of Biofilm in Microtiter Plates. APMIS, 115, 891–899.

31. Taddesse, S. (2014). Antimicrobial Profile of S. aureus from Clinical Specimens. Thesis, Addis Ababa University, Ethiopia.
32. Tong, S.Y. et al. (2015). Staphylococcus aureus infections: epidemiology, pathophysiology, clinical manifestations, and management. Clinical Microbiology Reviews, 28(3), 603–661.
33. Tuchscherr, L. et al. (2016). S. aureus develops increased resistance by forming dynamic SCVs during chronic osteomyelitis. Journal of Antimicrobial Chemotherapy, 71, 438–448.
34. Udobi, C.E. et al. (2013). Prevalence and Antibiotics Resistance Pattern of MRSA from an Orthopaedic Hospital in Nigeria. BioMed Research International, 860467, 1–4.
35. Wertheim, H.F.L. et al. (2005). The role of nasal carriage in S. aureus infections. The Lancet Infectious Diseases, 5(12), 751–762.
36. Wolter, D.J. et al. (2013). S. aureus SCVs are independently associated with worse lung disease in CF children. Clinical Infectious Diseases, 57, 384–391.
37. Yagci, S. et al. (2013). Prevalence and Genetic Diversity of S. aureus SCVs in CF Patients. Clinical Microbiology and Infection, 19, 77–84.